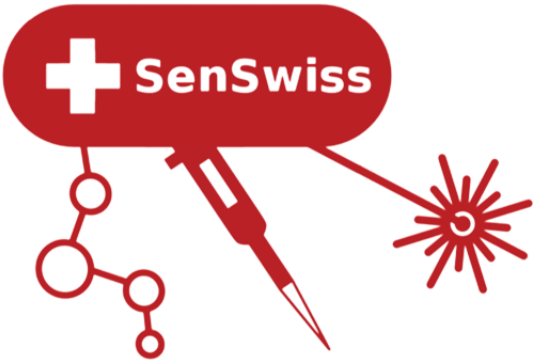

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1 Summary for SensUs website

SenSwiss is a point-of-care device that measures the concentration of an antibiotic, vancomycin, in blood plasma. Vancomycin is known to recognize and bind a specific cell-wall peptide precursor in Gram-positive bacteria. We took advantage of this by synthesizing our own fluorescently-labeled peptide that binds vancomycin in a similar fashion. Based on this, we developed a device that detects vancomycin by the principle of fluorescence polarization. If we shine polarized light on a plasma sample containing vancomycin, our fluorescent peptide emits a weak signal when unbound to vancomycin, and a much stronger signal when bound to vancomycin. Using the strength of the signal to measure the amount of vancomycin bound to our peptide, we can measure vancomycin concentration. The measurements are done with a miniaturized fluorescent microscope, with the help of 3D-printed parts.

We envision our device targeting both undiagnosed, critically-ill patients, as well as patients undergoing longer-term vancomycin treatment. Having effective therapeutic drug monitoring at the point of care, we hope to offer better treatments with improved clinical outcomes, which can drastically reduce hospitalization time and ease the burden on both patients and hospitals alike.

2 Biosensor System and Assay

2.1 General description of the detection technique

The SenSwiss biosensor uses direct fluorescence polarization (FP) as its method of detection. This technique relies on a fluorescently-labeled probe molecule binding the target molecule, vancomycin, with an affinity that is suitable for our application. A direct fluorescence polarization assay is a homogeneous assay, which is run entirely in solution and does not require surface functionalization or washing. It is therefore fast, simple and cheap to develop, making it an interesting and viable technique for point-of-care biosensors.

When a static fluorescent molecule is excited with plane-polarized light, it emits polarized light in the same plane of polarization. Since the fluorescent probe in a direct fluorescence polarization assay is free-floating in solution, the light it emits is quickly depolarized as it randomly rotates in all planes under Brownian motion. If, however, this fluorescent probe is bound to another molecule in a complex, it becomes heavier and rotates slower in solution. Consequently, the bound complex is able to retain the polarization of light for longer, giving a stronger detectable signal through a polarizing filter. The degree of polarization detected, or fluorescence anisotropy, is thus linked to the amount of target molecule in the sample, providing a method for concentration measurement.

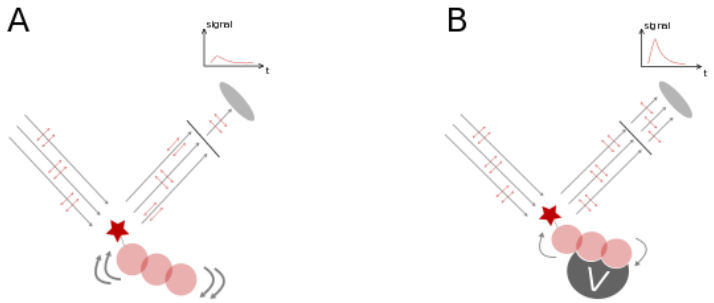

Figure 1: Fluorescence polarization assay. (A) The fluorescently-labeled peptide is free in solution, the light emitted from the fluorescent molecule is quickly depolarized due to its rotation and the signal detected is weak. (B) When vancomycin binds the peptide, its rotation is hindered, allowing for more polarized light to be emitted back from the fluorophore and increasing the observed signal.

SenSwiss adapted this principle to make a direct fluorescence polarization assay for detecting vancomycin, by taking advantage of its selective binding to the L-Lys-D-Ala-D-Ala (KAA) moiety of Lipid II, a peptide precursor of the bacterial cell wall. To test this system, different peptides were synthesized by solid-phase peptide synthesis (SPPS) and coupled to a fluorophore, fluorescein isothiocyanate (FITC). All peptides contained the KAA moiety, but were different in terms of amino acid sequence and structure. [1]. The final peptide used in our device is a branched-chain heptapeptide, Ac-L-Lys-Tyr-Glu-Leu-Lys(Tyr-Glu-Thr)-D-Ala-D-Ala (KYELKAA) (Appendix I, Figure 4). This peptide is the only reagent required to run our assay.

We chose to use a fluorescent peptide as a probe, since there is a wide pool of peptides to choose from that vancomycin selectively binds. In addition, peptides have considerably lower production costs and improved stability when compared to other potential binding partners, such as monoclonal antibodies. Finally, peptide chemistry is an area of research in which extensive expertise was available to us at our institution.

2.2 Biosensor system

The SenSwiss device operates with a 4 x 4 cm 3-well cartridge made of PMMA, a thermoplastic which has the advantage of being transparent and non-autofluorescent. The sample to analyze is split in three and applied to the wells with our peptide probe, before being inserted into a dedicated frame in the device through a small slot. This frame comprises a Peltier element to achieve optimal heating of the wells, a crucial step before taking optical measurements (see Section 3). A servomotor allows for automated scanning of the three wells.

To translate the incoming signal from the assay into a vancomycin concentration value, an optical setup sensitive to light polarization was built by our team. The SenSwiss biosensor contains a miniaturized fluorescent microscope, with two SMOS photodetectors, mounted in a custom 3D-printed case (Fig. 2).

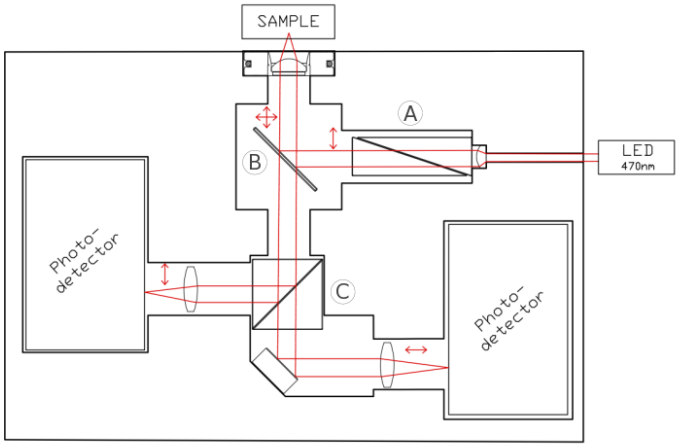

Figure 2: Optical system for the detection of the sample signal; the red arrows represent the direction of light polarization. An unpolarized coupled-fiber LED, emitting light at 470 nm, passes through a single band excitation filter with a wavelength of 475 nm and a bandwidth of 35 nm. This light is collimated with a lens and goes through a polarizer (A); only vertically-polarized light is retained. The optical beam is reflected up by a dichroic mirror (B) and focused on the sample. The fluorescent molecules in the sample emit light at a higher wavelength of 525 nm, which is transmitted through the dichroic mirror with a lower threshold of 499 nm. The beam is then split in two by a beam splitter (C), separating both vertically- and horizontally-polarized light, each collected by a photodetector.

The signal received by each photodetector is filtered and amplified by an electrical circuit embedded within the photodetectors, before acquisition is performed by an A/D converter communicating with an Arduino microcontroller. The latter subsequently processes the converted signal, comparing it with pre-stored calibration data, to retrieve a vancomycin concentration value interpretable by medical staff. The user interface of our device consists of a touch screen, communicating with the Arduino to coordinate different tasks, such as the heating and scanning steps required for the completion of an accurate vancomycin concentration measurement.

3 Analytical Performance

3.1 Off-Chip

Prior to use in our device, we carried out several experiments to validate the performance of our synthesized fluorescent peptides. Blood plasma has a high protein content and it has been demonstrated in the literature that vancomycin non-selectively binds to plasma proteins such as immunoglobulins and albumin [2]. These unwanted binding events interfere with our assay, disrupting its range of detection (Appendix I, Figure 5). To achieve more reliable detection, the plasma sample containing vancomycin needed to be heated at 80°C for 60 seconds prior to measurement, in order to denature the plasma proteins and break putative bonds with vancomycin. In addition, heat inactivation favorably increases the viscosity of the sample, yielding higher fluorescence anisotropy.

Upon heating, the plasma was centrifuged at 15,000 rcf for 120 seconds. The plasma supernatant was used to prepare samples at various vancomycin concentrations. Finally, we added 5 µL of our fluorescent peptide to 10 µL of each sample. Automated fluorescence polarization measurements were carried out in six biological replicates in a 384-well plate, using a *Tecan Infinite F200 Pro* plate reader.

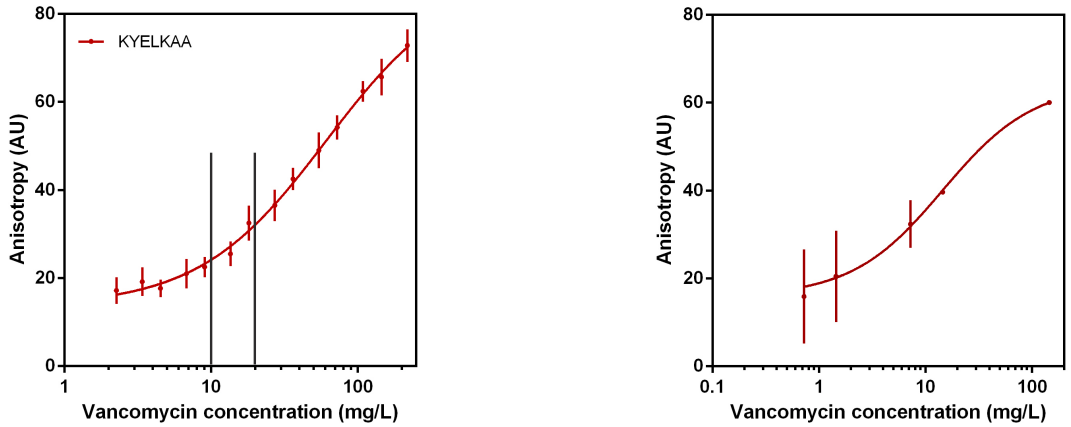

Figure 3: Fluorescence polarization titration curve for our peptide using a *Tecan Infinite F200 Pro* plate reader (left). The peptide was added at a concentration of 1 µM and titrated with a wide range of vancomycin concentrations in heat-inactivated plasma. The target therapeutic range of 10-20 mg/L is indicated. Using our optical setup, we have been able to obtain similar readings in PBS (right). This setup is still in the process of optimization.

3.2 On-Chip

In the context of the competition, our device will run on pre-centrifuged plasma samples, eliminating the need for a centrifugation step after heating.

In our device, the time-to-result is predominantly limited by the sample heating step. Our assay is run on a 15 µL sample volume. The sample volume is divided into 3 measurement wells. The wells first undergo heat-inactivation for 60 seconds, after which the peptide is added to the plasma. They are then read sequentially by the optical setup to generate triplicates after a 30-second incubation. The data acquisition for each reading, as well as processing the data to give a final concentration read-out, is almost instantaneous. In total, the time-to-result is approximately 3-4 minutes.

4 Novelty and Creativity

4.1 Already available

- Fluorescence polarization has been widely used and is still in use today in different diagnostics platforms for measuring vancomycin concentration, such as *Cobas Integra* by Roche [3].
- The direct fluorescence polarization approach we selected, using a fluorescent peptide probe to bind vancomycin, was inspired by a paper published in Analytical Chemistry in 2010 [4].
- The optical setup used in our biosensor was adapted from doctoral research conducted at EPFL [5].

4.2 New developments

Both previous and current technologies using fluorescence polarization for vancomycin TDM rely on a more complex assay; a competitive immunoassay that uses fluorescently-labeled vancomycin and capture antibodies [6]. The SenSwiss solution, for the first time, greatly simplifies the assay, replacing both antibodies and fluorescent vancomycin with a labeled peptide and reducing the number of steps involved.

Referring to a publication by Yao et al.[1], we successfully adapted four alternative peptides to our direct FP assay. Using one of these peptides, we were able to increase anisotropy and achieve noticeable signal improvement compared to the previous direct FP assay.

One of the greater challenges of our device was to test and adapt direct fluorescence polarization of vancomycin in blood plasma, which had previously only been demonstrated in PBS and FBS. We managed to develop a functional assay with simple heat inactivation protocols to greatly improve the reliability and reproducibility of the assay in plasma.

Beyond adapting the assay for plasma, we were able to take this one step further by successfully developing an assay that requires almost no plasma dilution. Using a highly-concentrated peptide stock and requiring minimal peptide probe concentration in the assay means that we require a very small volume of added probe. Adding 1 µL of fluorescent peptide probe to a 15 µL sample divided across three wells corresponds to a 1.2 dilution factor. This is in stark contrast to previous fluorescence polarization assays in plasma, with the competitive immunoassay having approximately 100,000-fold dilution [6] and the direct fluorescence polarization assay of teicoplanin in blood plasma using a 40-fold dilution [4]. In addition, the sample volume of 15 µL represents a significant improvement, compared to 400 µL in the previous direct FP assay [4].

4.3 Future developments

Bridging how our device is today with how we see it in the market, we want to use our strategic partnership with DBS System SA to develop smart, innovate solutions for blood sample processing. Currently, their microfluidic technology allows direct plasma extraction from 5 and 10 µL blood samples, allowing direct adaptation of our device to blood samples. In addition, we would dedicate R&D efforts to adapt their technology to plasma filtration, eliminating the need for centrifugation altogether. We also envision implementing a barcode reader interfacing with an Internet connection, allowing instantaneous storage of test results on the cloud for doctors to review. Finally, we would want to produce chips that are pre-loaded with the peptide reagent, since our peptide has demonstrated long-term stability at room temperature (Appendix I, Figure 4), as well as containing a stored 'internal standard' of known concentration to be read each time the test is run. This will further reduce the time-to-result, as well as providing an essential, additional level of quality control.

5 Translation Potential

Vancomycin is primarily administered in two situations: to critically-ill patients who have not yet been diagnosed, and to patients with allergies or resistance to first-line antibiotics. Due to its narrow therapeutic window, a generic dosage does not exist for vancomycin and treatments should be adapted to every case. In addition to receiving inefficient treatments, under-dosed patients are at risk of developing antibiotic resistant infections. Antibiotic resistance is a global health problem that costs USD 20 billion in the US and EUR 1.6 billion in the EU every year, due to extended treatments and hospital bed-days [7]. On the other hand, doses higher than the therapeutic limit have been linked to harmful side effects, principally in the kidneys and the inner ear [8].

Proper regulation of dosage can only be made through careful therapeutic drug monitoring (TDM) prior to administration. However, reports have shown that good practices are not universally enforced [9] and where they are, doctors have admitted that available solutions do not allow sufficiently rapid adjustment of doses, says Dr. Huttner from Geneva University Hospital. Vancomycin TDM is currently outsourced to analysis laboratories that, in spite of their high-throughput and performance, multiply human errors and cause delays; one TDM takes 50 minutes of medical manpower, according to Céline Fischer from Sion Hospital. Dr. Christin from Nyon Hospital adds that delays can be as long as 24 hours in small and rural hospitals that do not benefit from 24/7 laboratory services. These factors discourage doctors from running sufficient tests to provide their patients with the appropriate dosage in the first days of treatment. In fact, SenSwiss estimates that, on average, 1 in 15 usual hospitalization days [10] could be saved on vancomycin treatments if a faster test was available. Based on our interviews with Swiss doctors and reports from the literature, we found that France, Italy, Germany, the Netherlands, the UK, Switzerland, and the US, which constitute our primary market, have a combined total of 10.5 million vancomycin treatments every year [11][12][13][14]. As one hospitalization day costs, on average, EUR 300 [15], a total of EUR 3.15 billion could be saved annually in these countries if patients were properly monitored for vancomycin alone. These savings would be shared by both service providers (governments, hospitals) and patients/insurance companies. Beyond quantifiable numbers, lower mortality rates among severe infections and allowing patients to return home sooner are invaluable advantages that a new solution for vancomycin TDM would provide. Problems and solutions are summarized in Appendix II.

To address these needs, SenSwiss developed a point-of-care (POC) device to monitor vancomycin. To meet this objective, our company focused primarily on two aspects: providing fast and reliable vancomycin TDM tests and making tests easily accessible to both hospitals and patients.

The first goal was achieved technically. Our team developed a small POC device with a simple and user-friendly interface adapted to nurses and doctors. The patient is painlessly pricked on the finger, a single drop of blood (15 µL) is collected by the nurse on the test chip. Powered by the technology of our business partners, DBS System SA, the chip extracts plasma from the blood drop, which readily mixes with the reagent. The barcode on the chip is scanned by the instrument, the chip is inserted in the SenSwiss device and results are available on the cloud within 5 minutes. Decreasing manpower from 50 minutes to 15 minutes per test - by which the need for lab analyst work is also no longer required - corresponds to a reduction in associated overhead costs from EUR 26 to EUR 4.5 (Appendix IIIA). For doctors, considerable time-to-result reduction gives the possibility to readily adjust subsequent vancomycin doses, thereby improving the quality of the treatment.

The DBS System technology also allows us to store the sample safely for later analysis. As long-term

vancomycin treatments still require a weekly TDM, using this system in conjunction with our cartridge may allow nurses who visit patients for drug administration to collect blood samples at the same time. The extracted plasma is securely stored on a paper matrix and can be easily retrieved in the hospital with a simple protocol. Home monitoring is therefore compatible with the SenSwiss solution, and saves time for patients who would no longer need to make weekly visits to the hospital.

The second goal was reached by adapting our business model to the stakeholders' needs. The projections in the following section were done for years 1, 2 and 5 post-launch (y1, y2 and y5). As there should be no barrier for hospitals to adopt our solution, we decided to distribute SenSwiss instruments for free. Through this, SenSwiss aims to capture the vancomycin market at great pace; our market penetration growth is summarized in Appendix IIIB. In order to minimize the economic burden brought by free distribution, our device was conceived to be mass-produced at low cost. Most electrical and mechanical components (Appendix IIIC) are generically produced in large quantity at low cost by large distributors such as Distrelec or Misumi. Customized parts, including the housing, the optical setup box and the chip holder, can be easily mass-produced by injection molding. Only the optical components of the detection system are less compatible with mass-production. However, this problem can be overcome by creating strategic partnerships with optical manufacturers such as ThorLabs and plans for large-scale orders at advantageous prices. Instrument costs are summarized in Appendix IIIC, together with assembly costs, taxes and shipping. The total cost to bring a SenSwiss instrument to a hospital culminates at EUR 2'000.

SenSwiss plans on generating revenues from the sale of test cartridges, first in Europe, and also in the US one year later. Cartridges would have simple microfluidic geometry and contain pre-loaded reagent, produced at high quantity and very low cost. The chip is made of PMMA, a plastic that can be mass-produced by thermo-injection. The use of thermo-injection and the gradual improvement of our production channels will decrease the unit cost over time and increase production. The unit costs of the cartridges have been projected to be EUR 0.28, EUR 0.16 and EUR 0.07 for years 1, 2 and 5 post-launch. To these costs, we added taxes and transport (15% of price) [16] and a share to our distributors (15% y1, 7.5% y2, 7.5% y5), the latter decreasing over the years as SenSwiss aims to gradually handle its own distribution (50% y1, 25% y2 and 25% y3) (Appendix II). Cartridge costs are summarized in Appendix IIID. To further compensate the burden of free instrument distribution, we decided to sell the test at a price of EUR 19. Although it appears to be more expensive than EUR 3.5 tests run on big analysis platforms such as Abbott, we draw attention to the fact that processing our test costs EUR 4.5 of manpower against EUR 26 for currently implemented solutions. In total, the SenSwiss alternative would save up EUR 6 per test, which corresponds to annual savings of EUR 39'600 for a 1200-bed hospital in Switzerland. This benefit will be shared between hospitals and patients/insurance companies, although the exact division is difficult to determine.

A vancomycin treatment requires, on average, 5 TDM tests for proper dose adjustment. With 10.5 million treatments provided every year in target countries [11][12][13][14], 52.5 million vancomycin tests would be done. Based on these numbers and our estimated market penetration over the years, our company projected its revenues. By subtracting additional costs - instrument distribution, R&D, taxes, wages and company maintenance - we established our projected yearly profits over y1, y2 and y5 (Appendix IIIE and IIIF). We make a profit of EUR 7.6 million in the first 2 years, which fully covers the burden of 7 million required to launch our activities in the first 4 years pre-sales [16]. After 5 years of sales, our annual profit is expected to rise to EUR 36.1 million.

The thriving activity of our company gives us an incentive to tackle upcoming challenges in POC monitoring. SenSwiss plans on developing new tests compatible with its instrument. Aminoglycosides, another

class of antibiotics, is our first target. As a favored alternative for treating the rising threat of tuberculosis, its market is believed to grow 10% by 2022 [17], giving an estimated market of 5'000'000 patients in the US, UK and 5 European countries [18] [19]. With a high dose-dependent efficacy and 10% toxicity incidence [20], it demands careful POC monitoring that our device can provide.

6 Team and Support

6.1 Contribution of the team members

Aly: Worked on the synthesis of several fluorescent peptides for use in the device, as well as running fluorescence polarization assays for testing and optimization. Worked on entrepreneurship and various administrative tasks.

Paul: Worked on fluorescent peptide synthesis. Developed and optimized the fluorescence polarization assays in terms of peptide concentrations and plasma treatment. Worked on the translation of these assays to our own optical setup. Helped with administrative tasks and entrepreneurship.

David: Worked on the biological part of the project, including fluorescent peptide synthesis and fluorescence polarization assays. Worked on entrepreneurship, including the business model and conducting interviews.

Mathieu J.: First worked on the design and fabrication of the microfluidic chip. During the summer, started optimizing the optical setup and was involved in the electronics, data acquisition, data processing and Arduino software.

Matthieu P.: Worked on developing the optical setup of the device, which includes building the 3D-printed microscope. Electronic assembly of the photodiode-attached circuits, data analysis and co-designer of the SenSwiss device.

Alix: Worked on a separate detection method at the beginning of the project, then switched to developing the software on Arduino and joined the entrepreneurship team along the way. Team leader of SenSwiss, taking care of different organizational aspects.

Victor: Entrepreneurship guru for SenSwiss, from research on vancomycin use in health-care to the establishment of our business model. He was also co-designer of the SenSwiss device and participated in software development.

Jacopo: Worked on the hardware of the sensor, with special focus on the electronics. Also involved in the design and fabrication of the cartridge.

6.2 People who have given support

Prof. Christian Heinis: Head of the Laboratory of Therapeutic Proteins and Peptides (LPPT), where the assay was developed.

Gontran Sangouard: PhD advisor from LPPT who assisted with peptide synthesis and assay development.

Prof. Aleksandra Radenovic: Head of the Laboratory of Nanoscale Biology (LBEN), and her lab members, supported the project at the initial stage when ELISA was investigated as a potential detection method

Colin Darbellay: Assistant in the Microsystems Laboratory (LMIS4). Helped with work on microfluidics and regularly attended team meetings.

Prof. Dr. Jean-Manuel Segura: Professor at the Haute Ecole Spécialisée (HES) in Sion, who advised about the use of direct fluorescence polarization and provided valuable information about previous work.

Ronald Gianotti: Member of the Laboratory of Applied Photonics Devices (LAPD), who assisted with electronics and photodetector assembly.

Konstantinos Kaloulis: Organizer of the Tech/Business Case Club in the Technology Transfer Office (TTO) at EPFL. Helped us with various aspects of the translation potential of the device.

Dr. Toralf Scharf: Senior Scientist in the Nanophotonics and Metrology Laboratory (NAM), who provided his expertise in optics.

Dre. Angela Huttner: Doctor at Hôpital Universitaire de Genève (HUG), who gave us useful insights into the use of Vancomycin and its monitoring in Swiss hospitals

Pierre Lescuyer: Analyst at HUG who gave valuable insight about the use of vancomycin in healthcare.

David Tonoli: Analyst at HUG who gave valuable insight about the use of vancomycin in healthcare.

Dr. André Christin: Doctor at Hôpital de Nyon (GHOL) who greatly helped us to understand Vancomycin use and monitoring in the context of medium-sized Swiss hospitals.

Céline Fischer: Biomedical engineer working in the Biomedical and Sales department of the "Institut Central des Hôpitaux" (ICH) in Sion. Helped us understand the medical device acquisition process by hospitals reviewed our final prototype.

Dr. Dominique Werner: Clinical chemistry laboratory analyst at "Centre Universitaire des Hôpitaux Vaudois" (CHUV) who helped us understand how vancomycin monitoring is performed and how we could improve it.

Dr. André Pascal: Clinical pharmacology pharmacist at CHUV who helped us understand the importance of Vancomycin monitoring and how it is performed nowadays in Swiss hospitals.

Dr. Julien Déglon: Co-founder and CTO of DBS System, a Swiss company specialized in blood collection devices, who met with us to discuss their technology and possible partnership.

Dr. David Forchelet: Product development engineer at DBS System.

6.3 Sponsors

DBS System SA: Technology partners with an interest in our device. Discussed ways to combine our technology with theirs to offer a better POC biosensor.

Unipix: Have offered us their high-precision pipette calibration technology for use in the competition.

Lunaphore: Start-up company involved in medical diagnostics, who gave us support and mentorship in the development of our business model.

Hôpital de Nyon: Knowledge partners that have given invaluable information related to vancomycin, POC, TDM and health-care management.

7 Final remarks

We had a great year, full of challenges and setbacks, but we also learned a lot and have gotten to be very close as a team. We don't know at this point what lies ahead for SenSwiss, but it was definitely a pleasure to work on this project and to bring the idea to life as much as we could!

We would like to thank Professor Renaud for bringing our team together and for his ongoing support and valuable advice throughout the project. We would also like to thank Evgenii for always making the time to help us out.

See you in Eindhoven,

SenSwiss

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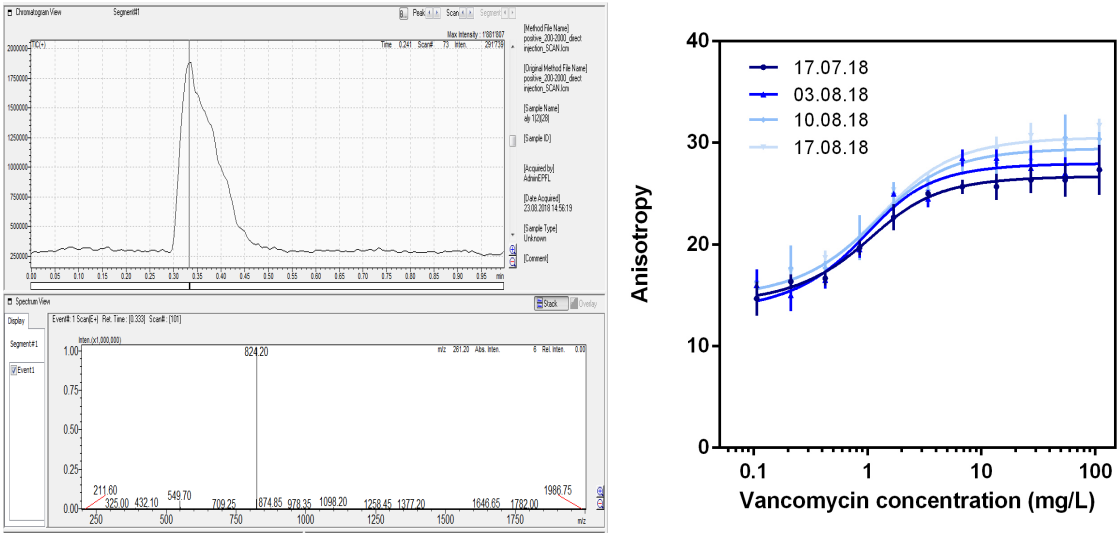
Appendix I



(a) (b)

Figure 4: Following peptide synthesis, the synthesis mixture was purified using high-performance liquid chromatography (HPLC) and the peptide fraction was verified using mass spectrometry. Our fluorescent peptide probe has a monoisotopic mass of 1646.6 g/mol. Figure 4(a) shows the direct-injection mass spectrum of our purified fluorescent peptide. The total ion count (TOC) reading shows a clear peak corresponding to a mass:charge ratio of 824.20. This precisely corresponds to a +2 cationized adduct of our fluorescent peptide. Figure 4(b) shows highly reproducible fluorescence polarization titration curves of our peptide in PBS, stored at room temperature over the course of one month.

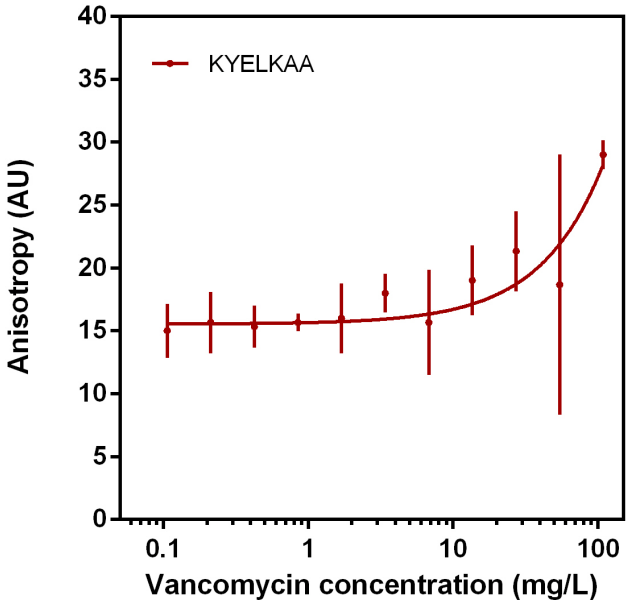


Figure 5: Fluorescence polarization titration curve for our peptide in non-heat-inactivated plasma. The peptide was added at a concentration of 1 µM. The presence of interfering plasma proteins significantly alters the detection range of the assay. A large loss in anisotropy is observed, as well as larger error bars.

Appendix II

Summary of stakeholders' needs and value proposition of the SenSwiss solution

Problem	Impact on stakeholders	SenSwiss Solution	Outcome
Unadapted dosages make treatments and hospitalization longer and decrease prognosis	**Patients**: - experience longer and difficult stays - pay for long hospitalization - are cut from their home and work	- **Small**, **easy-to-use** device - **Fast & accurate** results (**5 minutes**) - **Low volume** of sample (**15 uL**) - Vancomycin monitoring is **more accessible** to hospitals that neglected this practice. - Other hospitals have access to an **easier method of monitoring**. - It leads to **better adjustment of vancomycin dosage**, which shortens the treatment duration.	- **1 out of 15 days** of treatment **saved** on average. - **€ 126 millions saved** if we reach 4% penetration within 1 year in France, Italy, Germany, the Netherlands, the UK, Switzerland. - These **benefits are shared between governments, hospitals, patients and insurance companies**
	Doctors/Nurses: - involve more work in extended hospitalization		
	Insurance companies: - cover extra hospitalization days		
	Hospital management: - slow patient turnover leading to crowded surface promoting infection transmission		**4 beds per day could be freed** in a 1200-bed hospital using the SenSwiss solution (CH data)
Unadapted dosage increases rise of antibiotic resistance	**Governments**: - have to invest to find alternatives to the rise of antibiotics. - Resistances are a yearly burden of **€ 18.6 billion every year** worldwide	- Easy-to-use solution providing results within 5 minutes greatly **accelerates the adaptation of the dose to each treatment**.	**Rise of resistance to vancomycin is slowed**. As a result, saves important global healthcare costs to governments.
	Doctors, patients, insurance companies, hospitals: - **antibiotic resistance** contribute in the **long term** to **increased hospitalization**		**Decrease the fast growth of bed demand** in hospitals by preventing higher vancomycin-resistant strains to arise.
Long delays in vancomycin TDM time-to-result slow down the dose adaptation of treatment	**Patients**: - **Receive adapted dose late**, which may **reduce the quality of the treatment** and **extend their hospitalization stay**	- Easy-to-use solution providing results within 5 minutes greatly **accelerates the adaptation of the dose to each treatment**. - There is **no need for intermediate laboratory** to conduct the test.	Reduction of **1 in 15 days** of hospitalization (see above)
	Doctors, nurses, analysts: - A **lot of manpower** involved in the current solution - **More actors in the chain** promote human errors.		Doctors and nurses are **empowered** and conduct vancomycin monitoring themselves. Doing so, they can **provide a more precise and adapted treatment.**
Weekly vancomycin checkup for patients	**Patients**: - have to **go to the hospital weekly** in long treatments to check their vancomycin levels.	**Partnered with DBS System** technology, we give the possibility to visiting nurses to **collect blood samples from patients' home** These samples are **safely stored** on paper and easily processed at the hospital for further reading.	- Patients **do not need to go** for weekly checkups at the **hospital** anymore. - **Follow up** can be done **at distance** by the doctor.
	Insurance companies: - try to **lower their expenses** in healthcare	Price of single vancomycin test is **reduced from € 29.5 to € 23.5**	**€ 43'200 saved in a 1200-bed hospital**

Appendix III

A. Vancomycin test cost - current situation vs. SenSwiss

Average nurse salary (EU) [21]	€ 3'000.00	**Average analyst salary (EU)** [15]	€ 5'500.00	**Average working hours per day in EU**	7h 36m
	Nurse time	**Analyst time**	**Manpower costs**	**Test cost**	**Total unit cost**
Current solution	20 minutes	30 minutes	26 € (c.f. Part 5)	3.5 € [26]	€ 29.50
SenSwiss solution	15 minutes	0 minute	4,5 €	19 €	23.5 €

B. Market penetration and sales projections

Annual # of vancomycin tests performed (EU*) [11, 12, 13, 14]	12'468'400	**Annual # of vancomycin tests performed (US)** [11, 12, 13, 14]	39'858'000
Test cost	€ 19.00		
*EU means in this context France, Italy, Germany, the Netherlands, the UK, Switzerland			
	year 1	**year 2**	**year 5**
EU penetration	4%	10%	30%
Annual # of tests sold (EU*)	500'000	1'246'800	3'740'500
US	not entered	4%	20%
Annual # of tests sold (US*)	0	1'590'000	7'971'600
Annual number of tests sold	500'000	2'836'800	11'712'100
Total annual revenue	€ 9'500'000.00	53'900'000	€ 222'530'000.00

C. Instrument unit costs

Electronics [23]	€ 100.00
Mechanics [22, 24]	
Generic parts [24]	€ 30.00
Housing [22]	€ 100.00
Optics	€ 770.00
Production total	**1000** €
Assembly (5% production total)	€ 100.00
Distributor shares (30% production total)	€ 600.00
Transport and taxes (15% production total)	€ 300.00
Total unit cost	**2000** €

D. Test chip unit costs

Prod + assembly [22]	€ 0.28	€ 0.16	€ 0.07
Distributor share (% of price) [12]	15% of price 2.85 €	7.5% of price 1.42 €	7.5% of price 1.42 €
Transport & taxes (15% of price)	€ 2.85	€ 2.85	€ 2.85
Total unitary cost	€ 5.98	€ 4.43	€ 4.34

E. Instrument and test chip total annual costs

	year 1	**year 2**	**year 5**
#chips sold / year	500'000	2'836'800	11'712'100
Total chip costs	€ 2'990'000.00	€ 12'567'000.00	€ 50'830'500.00
#instruments distributed / year	342	745	868
Total instrument costs	€ 684'000.00	€ 1'490'000.00	€ 1'736'000.00
Total annual instrument and chip costs	3'674'000 €	14'057'000 €	52'566'500 €

F. Total annual benefit and expenses

	Revenues from tests	**Chip and instrument costs**	**R&D (35% of revenues)**	**Taxes (15% of revenues)** [25]	**Company costs (10% of revenues)**	**Annual total**
year 1	€ 9'500'000.00	-€ 3'674'000.00	-€ 3'325'000	-€ 1'425'000.00	-€ 950'000.00	**€ 126'000.00**
year 2	€ 53'900'000.00	-€ 14'057'000.00	-€ 18'865'000	-€ 8'085'000.00	-€ 5'390'000.00	**€ 7'503'000.00**
year 5	€ 222'530'000.00	-€ 52'566'500.00	-€ 77'885'500.00	-€ 33'370'000.00	-€ 22'530'000.00	**€ 36'178'000.00**