

WOLF – a device to facilitate cell-based immunity testing

Anja Garritsen¹, Wilfred Teunissen², Freerk van Oudheusden², Milou Kouwizjer¹, Anja Scholzen¹, Richard Kneusel³, Hannes Stockinger⁴, Herman van der Vegt²



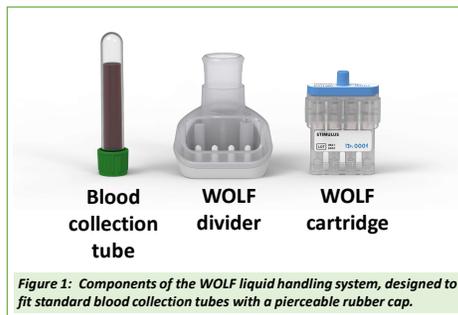
- Innatoss Laboratories B.V., Oss, The Netherlands
- npk design B.V., Leiden, The Netherlands
- DIARECT AG, Freiburg im Breisgau, Germany
- Institute for Hygiene and Applied Immunology, Center for Pathophysiology, Infectiology and Immunology, Medical University of Vienna, Austria

A need to simplify logistics of cell-mediated immunity tests

- Cell-mediated immunity (CMI) tests are used to diagnose (prior) infection by intracellular pathogens and to establish antigenicity of vaccines.
- T-Spot-TB (Oxford Immunotec) and LymeSpot (AID) use PBMCs for antigen stimulation, whereas QuantiFERON-TB (Qiagen) and Q-detect (Innatoss) are whole blood-based tests.
- The use of CMI tests is limited by the fact that living cells are needed. Consequently, blood or cell samples need to be stimulated within hours from collection.
- With this time constraint, Innatoss faced a huge logistic challenge in a pilot study exploring a novel CMI test for Lyme Borreliosis in the Netherlands, which is a relatively small country. In remote regions where new diagnostics and vaccines for infectious disease are most needed, logistics for CMI-based tests would be even more challenging.
- Supported by the Fast Track to Innovation program (Horizon 2020) with academic and industrial partners from Wien, Oss, Leiden and Freiburg we therefore designed a liquid handling device (termed WOLF) to facilitate the logistics of CMI testing and enable blood stimulation directly at the point of blood collection.**¹

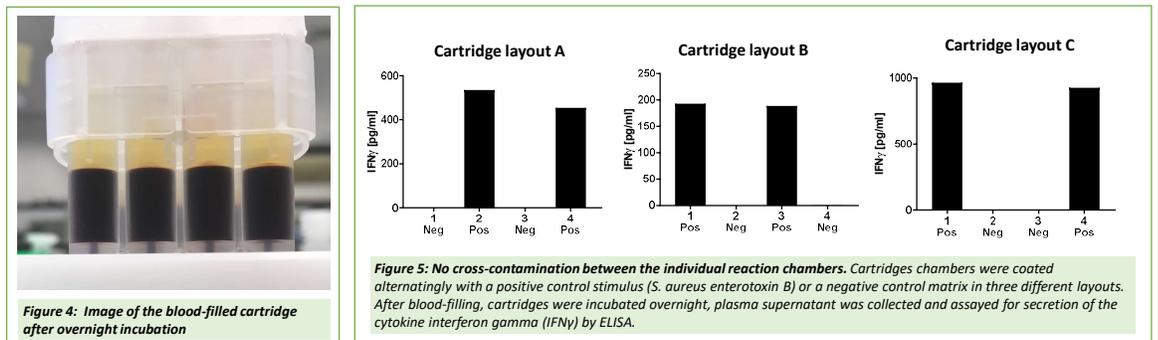
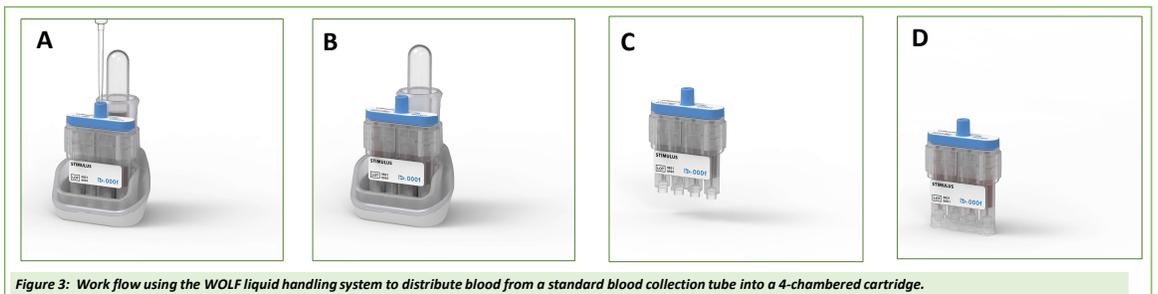
Materials and methods

- A demonstration product series of the WOLF liquid handling system with the required characteristics and tolerances was produced.
- Several iterations and revisions of molds were needed before having a fully functional product.
- The manufactured prototype consists of a splitter device with an integrated blood tube holder and a 4-chambered cartridge with a lid (Fig 1). The chambers can be filled with disease-specific stimuli for CMI testing
- The lid includes a filter that is sealed onto the lid housing. The filter is instrumental in stopping the blood flow when the chamber is filled. Further optimization was achieved by introducing a one-way valve to stop air from entering the reservoir, thus precluding contamination and leakage (Fig 2).



Results

- A blood collection tube is placed on the divider and under-pressure is applied (Fig 3A), resulting in blood flowing from the tube through microfluidic channels into the separate chambers (Fig 3B). The blood-filled cartridge is then removed from the divider (Fig 3C) and closed with a plug (Fig 3D).
- Following stimulation, the cartridges can be shipped to a central laboratory for analysis of cytokine secretion. The cartridge withstands even rough handling/shipment conditions (not shown).
- Removal of the lid after incubation of the blood-filled cartridge allows for collection of stimulated whole blood or plasma.
- We demonstrate that filling of the chambers of the cartridges is even and reproducible (Fig 4).
- No cross-contamination between the chambers occurred (Fig 5).



Conclusion & Outlook

- The WOLF device is functional and broadly applicable for whole blood-based testing. It allows stimulation directly at the point of blood collection without the need for specialized personnel and laboratory facilities such as biosafety cabinets, which limits logistic hurdles, time and labor.
- The availability of this novel liquid handling device will stimulate development of novel CMI-based tests.

References

- Patent WO2019038437; <https://patentscope.wipo.int/search/en/de?tail.jsf?docId=WO2019038437>