A new NGS-based method for chimerism monitoring

Dvysr®

BIOMETRIX DIAGNÓSTICA

Dr. Dan Hauzenberger, Medical Director, Section of Transplantation Immunology at Sweden's Karolinska University Hospital, on how new, highly sensitive and precise methods for mixed chimerism analytics are transforming post-transplant monitoring, enabling earlier detection and treatment to save the lives of transplanted. He also discusses why Karolinska decided to replace their STR-PCR and qPCR methods with a single, NGS-based method and the benefits achieved by doing so.

■■■ Allogenic stem cell transplantations at the Karolinska Hospital

The Karolinska Hospital in Huddinge is home to Sweden's only specialized clinic for allogenic stem cell transplantations (CAST) and performs more transplantations than any other hospital in Sweden. It treats Swedish and international patients with malignant haematological diseases, anemia and immunodeficiency diseases as well as certain metabolic diseases. The specialized clinic also performs advanced cell therapy for cancers such as lymphomas and leukemias (Car-T).

Research is an integrated part of this clinic since more than four decades, resulting in a vast number of scientific publications with focus on managing complications such as Graft vs. Host Disease, infections and disease relapse.

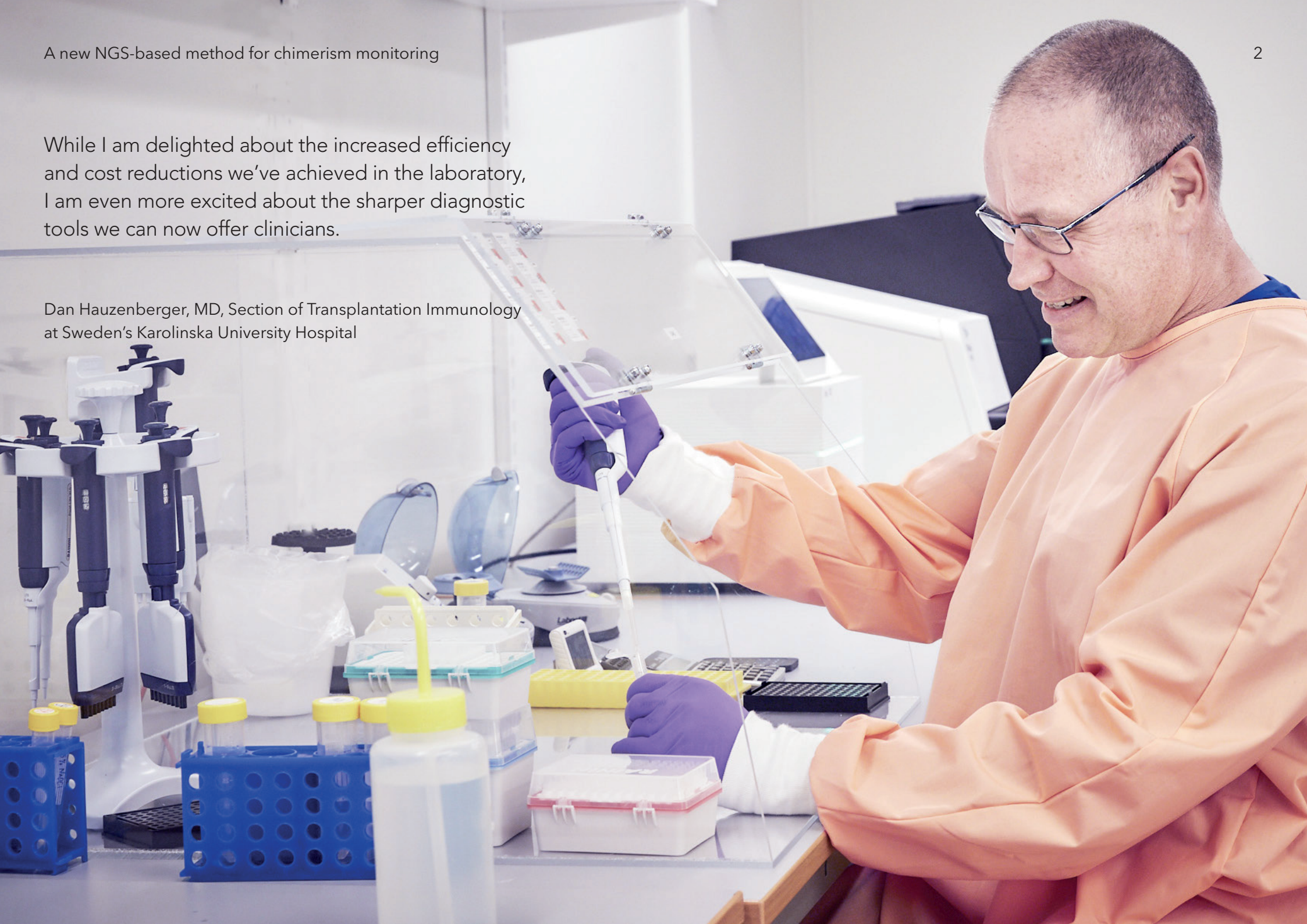
The highly specialized Transplantation Immunology Laboratory supports the transplantation teams at Karolinska. The laboratory follows each patient throughout the entire transplantation process, from finding a suitable stem cell donor, pre-transplantation tissue typing to post-transplantation chimerism monitoring.

Dr. Dan Hauzenberger is associate professor and senior physician at Karolinska University Hospital, as well as Medical Director of the Section for Transplantation Immunology for the past 15 years. Dr. Hauzenberger has focused on chimerism analysis in transplanted patients for more than 20 years and has led the development of new technologies, including highly sensitive molecular diagnostics for chimerism analysis, at his laboratory.

"The pace of diagnostic innovation is extremely high today, and chimerism is no exception. New, highly sensitive and precise methods can significantly increase patient survival as well as reduce costs for the laboratory. As Medical Director of the Transplantation Immunology Laboratory it is my responsibility to not only ensure that we leverage the best diagnostic technologies, but also to help the clinicians find the gold dust in the massive amounts of emerging research and translate this into actionable information that will help save lives."

While I am delighted about the increased efficiency and cost reductions we've achieved in the laboratory, I am even more excited about the sharper diagnostic tools we can now offer clinicians.

Dan Hauzenberger, MD, Section of Transplantation Immunology at Sweden's Karolinska University Hospital



■■■ Two separate methods for detection of mixed chimerism

For at least 15 years Karolinska's Transplantation Immunology Laboratory has used a combination of STR-PCR and qPCR, two well-established methods for detecting mixed chimerism. Both have clear advantages and disadvantages. The STR-PCR-based methods generally show high precision but a restricted limit of detection (LOD); qPCR on the other hand exhibits a high sensitivity but poor precision, especially at higher levels of mixed chimerism.

More transplantations and more frequent testing drove need for new technology

Serving one of the leading transplantation clinics in Europe with 80-100 transplanted patients each year, Karolinska's Transplantation Immunology Laboratory faced a growing demand for pre- and post-transplant testing, as well as life-long chimerism monitoring. Running larger volumes of tests while maintaining two separate protocols required numerous laboratory staff and resources and led to escalating costs — clearly not a sustainable approach. A rising number of transplantations, however, was not the only cause of the increasing volumes. New and more sensitive methods also held the promise of increasing patient survival.

"The more sensitive the method, and the more frequently you test, the earlier you can detect minimal residual disease and in case of disease relapse initiate the curative treatment critical to survival. Survival rates are higher for patients who have undergone just one transplantation and the earlier you treat relapse, the higher the success rate. If a second transplantation is the only remaining option, early detection of a failed transplantation will buy valuable time for the necessary preparations, a process that can take months."

+30 %

Increased capacity

–30 %

Time required in the lab

■■■ Weighing the options for a better method

With an increasing number of transplanted patients requiring life-long monitoring, combined with more frequent testing, Dr. Hauzenberger was strongly incentivized to find a method that would allow Karolinska to process more tests faster.
More specifically, he wanted a robust, more cost-effective method that combined the sensitive analysis of qPCR with the accurate measurements at high chimerism levels of STR fragment analysis. Novel research on NGS-based methods for chimerism analysis showed promising results [1]. One of these assays, based on quantitative detection of selected markers distributed throughout the human genome, showed sensitivity below 0,1% LOD as well as high accuracy over a wide dynamic range [2]. This indicated that NGS platforms may be suitable for developing assays for chimerism analysis with all the advantages of STR and qPCR without the drawbacks.

The solution: a single NGS-based method combining high sensitivity and precision

Karolinska decided to implement Devyser's NGS-based product for chimerism monitoring. It offers one simple protocol for fast chimerism measurement and monitoring in transplanted patients which enabled Karolinska to increase throughput. More importantly, when this sensitive NGS method is combined with cell separation, it allows Karolinska to detect potential disease relapse very early.

Results

I cannot emphasize enough how much easier it is to operate and maintain this single method compared to the previous two, especially in terms of fewer staff required and less hands-on time in the laboratory " says Dr. Hauzenberger. "We can now run the new method with just one to two persons in the lab, compared to 3 to 4 before. The previous methods were also difficult to batch together to run simultaneously, which required us to dedicate one person at all times to handle incoming tests. Now we batch a large number of tests together and run them all at once, with the same turnaround time as before. We still average a turnaround time of three days, with a fraction of the manual work previously required."

Devyser's method is well documented and technically easy to learn, operate and maintain, thereby reducing the risks associated with staff turnover. Interpreting and reporting the results requires specialized competence and is facilitated by the included software.

"While I am delighted about the increased efficiency and cost reductions we've achieved in the laboratory, I am even more excited about the sharper diagnostic tools we can now offer the clinicians. After all, our ultimate goal is to reduce suffering and to save and improve the lives of transplanted patients. Devyser's method, now introduced into our workflow, enables us to detect minimal residual disease and disease with higher precision and a significantly improved workflow in the laboratory. For transplanted patients, sensitive and precise diagnostics saves lives."

High sensitivity for early detection

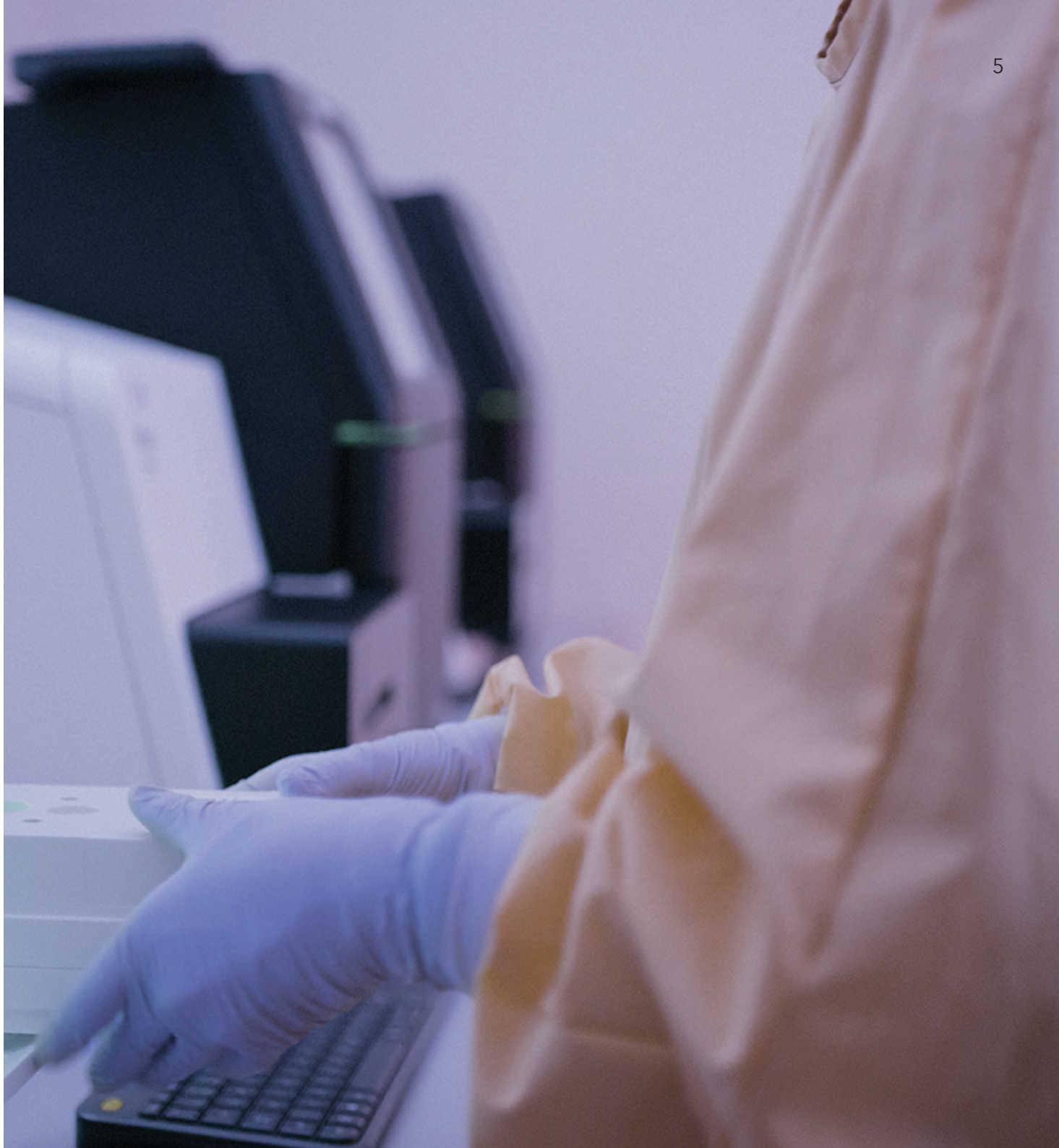
Very early detection of relapse is possible thanks to high sensitivity and accurate measurement down to 0.05% minority fraction chimerism.

Fast and easy workflow from patient sample to report

A complete solution including NGS library preparation and analytical software for a unified workflow from initial screening of recipient/donor pairs to subsequent life-long patient monitoring.

Easy to use

One tube NGS library prep with just 45 minutes hands-on time combined with powerful and intuitive data analysis and reporting software.



About the Karolinska University Hospital

The Department of Transplantation Immunology has been extremely successful in developing new diagnostics for clinical use. Among other things, the clinic has been one of the first laboratories in the first in the world to develop molecular typing for HLA (PCR-SSP) which was developed by Olerup during the 80s and 90s. This technique has been one of the best-selling HLA typing technology in the world for many years. Furthermore, the section has developed the first flowcytometry-based crossmatch before transplantation (Sumitran-Karuppan, Alheim) and has been among one of the first laboratories in the world to develop several molecular chimerism assays (Mattsson, Uzunel and Hauzenberger). All these assays are now part of routine diagnostics at most transplant centers in the world. In addition, all these development projects have yielded many scientific publications and dissertations. Thus, there is both a tradition and competence to conduct research and development at the section. The Department of Transplant Immunology has also conducted clinical research for a long time together with the Clinic for Transplant Surgery and the Center for Allogeneic Stem Cell Transplantation (CAST).

About Dr. Dan Hauzenberger

Dr. Dan Hauzenberger is associate professor and senior physician at Karolinska University Hospital, as well as Medical Director of the Section for Transplantation Immunology for the past 15 years. Dr. Hauzenberger has focused on chimerism analysis in transplanted patients for more than 20 years and has led the development of new technologies, including highly sensitive molecular diagnostics for chimerism analysis, at his laboratory.

References

1. Aloisio et al. 2016
2. Pettersson, L. Et al. 2019

Versão 012023

Dvysr®

Distribuido por:

BIOMETRIX DIAGNÓSTICA

Para maiores informações, consulte-nos: **suporte@biometrix.com.br**

WWW.BIOMETRIX.COM.BR