

Authors

Sarah Anne Hanauer
Umwelt-Campus Birkenfeld, Germany

Dr. Wolfgang Peter
Stefan-Morsch-Stiftung, Birkenfeld, Germany

Dr. Stephan Jacobs
PerkinElmer chemagen Technologie GmbH
Arnold-Sommerfeld-Ring 2, 52499 Baesweiler, Germany

chemagen Technology chemagic Seq Pure Kit

The **chemagic Seq Pure Kit** was developed to purify sequencing reactions, including BigDye® terminator reactions. Clean-up is performed using the unique **chemagic Magnetic Bead** based technology.

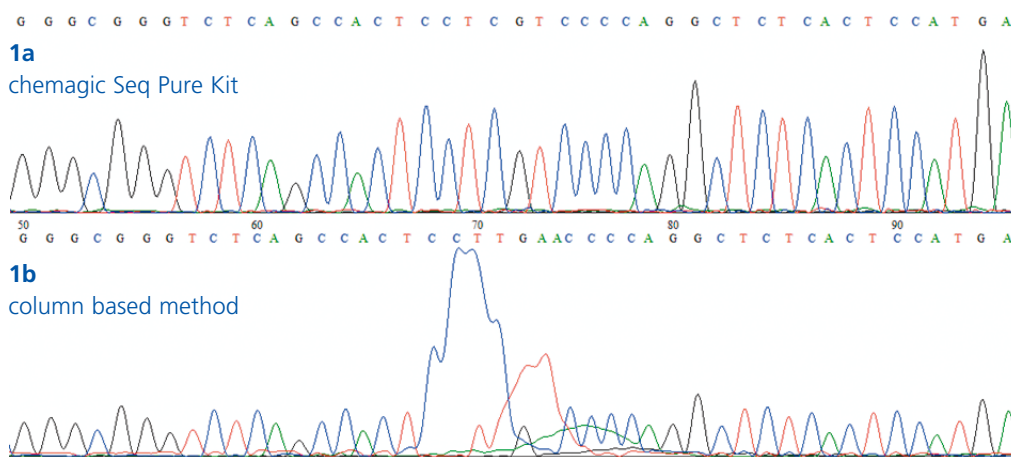


Figure 1a/b: HLA-Typing (SBT)

Comparison of two Electropherograms of purified BigDye® terminator v3.1 sequencing reactions. **Figure 1a** shows a sequencing reaction mix purified with the **chemagic Seq Pure Kit** adapted on a PerkinElmer® JANUS® Mini MDT® Liquid Handling Platform. **Figure 1b** shows the same sequencing reaction mix purified using a standard column based manual protocol. Sequencing reactions were performed using 1/16 x BigDye® terminator v3.1 chemistry, gDNA (isolated with **chemagic Blood Kit special** in combination with the **chemagic Magnetic Separation Module I**) and an oligonucleotide primer annealing within the HLA-A- Locus. After purification, the samples were analysed on an Applied Biosystems 3130® DNA Analyzer. With the **chemagic Seq Pure Kit** long and high quality reads are achieved, the appearance of dye blobs (**Fig. 1b**) is eliminated (**Fig. 1a**).

The clean-up process is simple, fast and convenient as it requires neither centrifugation, nor filtration steps and removes any type of dye terminator from 5-20 µl sequencing reactions. The kit has been optimized for use with Amersham DYEnamic ET and BigDye® chemistries (including v1.1 and v3.1) and yields highly purified sequencing reaction products. The protocol recommends 1/4- 1/16 x scale reactions to save on reagent costs, to reduce dye blobs and to improve sequence data quality. The obtained results are of the highest quality with high fluorescent signal intensities, eliminated dye blob occurrence and routinely long read lengths (> 900 bp). They achieve 98 % accuracy to 900 bases and Phred 20 quality scores > 825 bases.

chemagic Seq Pure Kit

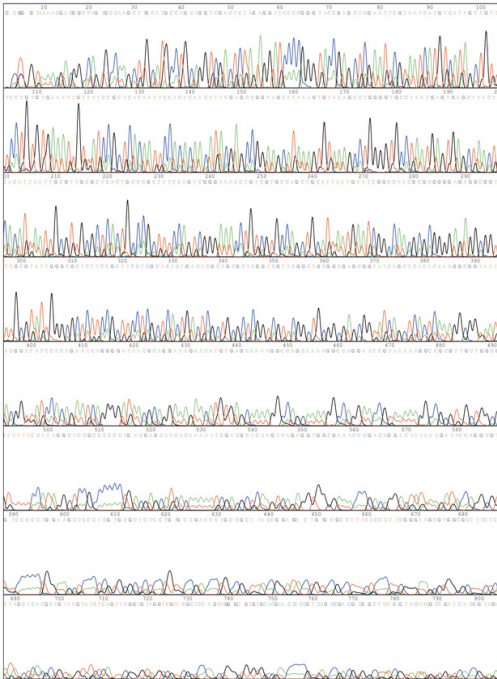


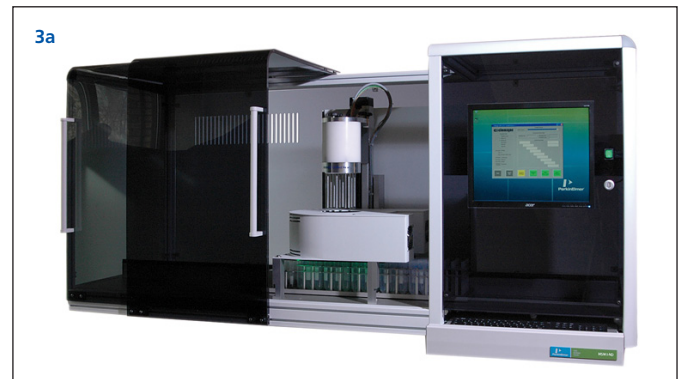
Figure 2: Plasmid Sequencing

Electropherogram of a purified BigDye® terminator v3.1 sequencing reaction. It was purified using the **chemagic Seq Pure Kit** and **chemagic Magnetic Separation Module I**.

Sequencing reactions were performed using 1/4 x BigDye® terminator v3.1 chemistry, pGEM-3Zf(+) (400 ng), and pUC/M13 (-21) oligonucleotide primer. After purification, the samples were analysed on an Applied Biosystems 3730 DNA® Analyzer. This electropherogram is typical for the high quality reads achieved using this clean-up method (Phred 20 > 825).

For high throughput sequence clean-up reactions the **chemagic Seq Pure Kit** can be adapted on commonly used liquid-handling systems e.g. PerkinElmer® JANUS® Mini MDT® (**Fig. 1, Fig. 3b**).

The **chemagic Seq Pure Kit** is best used with the **chemagic Stand 96** (**Fig. 3c**), the ideal accessory for automated or manual purifications. Furthermore, the **chemagic Magnetic Separation Module I** (**Fig. 2, Fig. 3a**), as a validated high throughput system, is an ideal solution for the automated sequence clean-up.



PerkinElmer, Inc.
940 Winter Street
Waltham, MA 02451 USA
Phone: (+1) 800-762-4000
or (+1) 203-925-4602
www.perkinelmer.com

PerkinElmer chemagen Technologie GmbH
Arnold-Sommerfeld-Ring 2
52499 Baesweiler, Germany
Phone: (+49) 2401 805-501
Fax: (+49) 2401 805-519
info@chemagen.com, www.chemagen.com



For a complete listing of our global offices, visit www.perkinelmer.com/ContactUs

Copyright ©2013, PerkinElmer, Inc. All rights reserved. PerkinElmer® is a registered trademark of PerkinElmer, Inc. All other trademarks are the property of their respective owners.

CT13/24/0813-01 Printed in Germany