SEQme
SEQ & QPCR COMPANY

Sanger Sequencing & Fragment Analysis

Services à la carte

www.seqme.eu

From a one-time job to the long-term partnership

SEQme – Your partner in Sanger Sequencing and Fragment Analysis Services

At SEQme we believe that Sanger sequencing and Fragment analysis are techniques that have still much to say, regardless of fast development of DNA sequencing technologies in recent years. In fact, our company has been established to provide full portfolio of services in DNA sequencing and as you may guess or perhaps witnessed yourself Sanger sequencing services were present right at our onset.

We pride ourselves on being able to meet various needs you as our customers in Sanger sequencing may have – standard and low-price driven sample analyses in our lab, special protocols enabling analysis of difficult samples, add-on services like sample cleanup as well as more challenging tasks, for example direct sequencing of high-molecular weight DNA. Additionally, we offer five fragment analysis protocols enabling sizing of your samples up to 1.200 bp using four combinations of fluorescent dyes. Of course, all orders are processed and data delivered via a secure access account on our website. To use our services, you can either register personally or your lab can register as a team. All services can be prepaid and lab members can easily access funds deposited on the lab account which simplifies order processing significantly and allows you to stay focused on your research.

A dedicated team of professionals knowledgeable in Sanger sequencing to the very last detail processes all samples as fast as possible and – a rather unique approach in this automatic world – *performs manual analysis of every single trace* coming out from our sequencers to give you the best electropherograms you can get. We do not rely on automatic data analysis algorithms, in our long-term experience all Sanger basecallers are error prone.

In case of interest we provide also expert consultancy on all steps of the workflow, experiment optimization recommendations, or problem-solving suggestions.

Last but not least, you can take part in our courses or workshops - old they may be, these techniques still present challenges for newbies. When participating at our courses you gain an overall picture of the Sanger workflow and have the opportunity of directly interacting with our team members, including lab specialists. Should you have access to your own sequencer, you can still benefit when joining our workshops for instrument users - you will learn how to operate these machines efficiently to save your time and lower costs per sample. As you can see, you can either work with us or learn from us since we are always open to share our know-how. In any case, when thinking about Sanger sequencing or fragment analysis, rely on us.

Give it a try and let us offer our Sanger Sequencing services to you for the first time or become our loyal customer and partner to profit most from the long-term cooperation.

Your SEQme team

There are no good or bad, cheap or expensive Sanger sequencing protocols in our portfolio, there are only those more or less appropriate for the task in your mind.

Sanger Sequencing Services at SEQme

StandardSeq

Standard single read sequencing service for plasmids or PCR products

- You supply - PCR product or plasmid mixed together with sequencing primer
- We perform - Sequencing reaction, electrophoresis and data analysis
- Time to results - Up to 24 hours after arrival of samples to our lab

CleanSeq

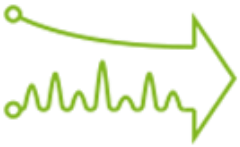
Sequencing of PCR products including their enzymatic cleanup

- You supply - Unpurified PCR product or other suitable template and a sequencing primer
- We perform - Enzymatic cleanup of your PCR product, sequencing reaction, electrophoresis and data analysis
- Time to results - Up to 36 hours after arrival of samples to our lab

HairpinSeq

Special sequencing protocol for hairpins, GC-rich and other problematic templates

- You supply - Plasmid or PCR product and two primers
- We perform - Bidirectional sequencing reaction using a special sequencing chemistry and both primers you supply, electrophoresis and data analysis
- Time to results - Up to 24 hours after arrival of samples to our lab



ReadySeq

Electrophoresis of ready sequencing products in our sequencers

- You supply - Ready sequencing reaction
- We perform - Electrophoresis and data analysis
- Time to results - Up to 12 hours after arrival of samples to our lab

… order all protocols online at www.seqme.eu/order

Any application, any scientific goal…

Fragment Analysis Services at SEQme

Fragment analysis involves labelling DNA fragments (PCR products) with fluorescent dyes and multiple different colored fragments can then be detected in one sample. One of the dye colors is used for a labelled size standard present in each sample and used to extrapolate the base-pair sizes of the sample product peaks.

We perform capillary electrophoresis of labelled DNA fragments, sizing typically up to 1200 bp.

We support following dye combinations (dye sets):

Dye set	Dyes	Size standard and sizing range
DS-33	6-FAM™, VIC®, NED™, PET®	GeneScan-600 LIZ, 20-600 bp or GeneScan-1200 LIZ, 20-1200 bp
DS-30	6-FAM™, HEX, NED™	GeneScan-500 ROX, 35-500 bp
PowerPlex	Fluorescein, JOE, TMR-ET, CXR-ET (Promega)	Your choice, WEN-labelled (previously CC5)
BT5	6-FAM, BTG, BTY, BTR, BTO (Qiagen)	Your choice, BT5-labelled

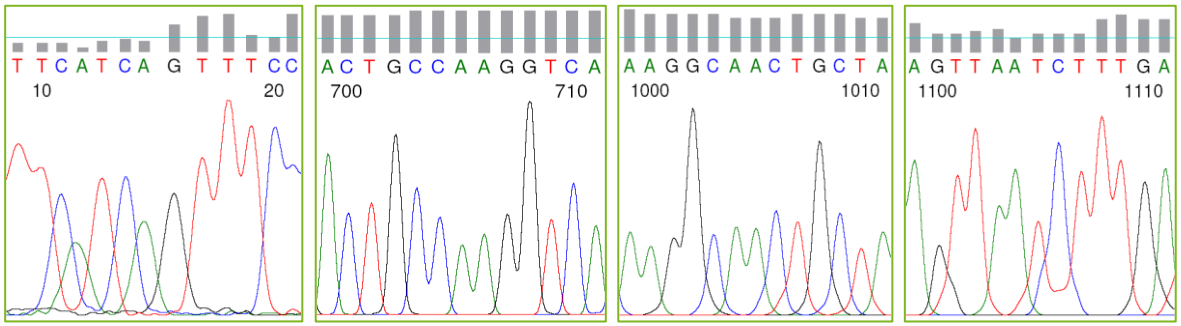
Fragment analysis applications include:

- Microsatellite (STR) analysis - linkage mapping, animal breeding, human, animal, and plant typing, pathogen sub-typing and others
- Relative Fluorescence applications compare peak height or area between two samples. Common techniques include namely Qualitative Fluorescence (QF) PCR and Multiplex Ligation-dependent Probe Amplification (MLPA). Applications include LOH in tumor samples, Copy Number Variation (CNV), Aneuploidy detection

Just as your preferences are diverse, so are the strategies we use to give you the best results you deserve.

3 good reasons why choosing SEQme as your Sanger sequencing service provider:

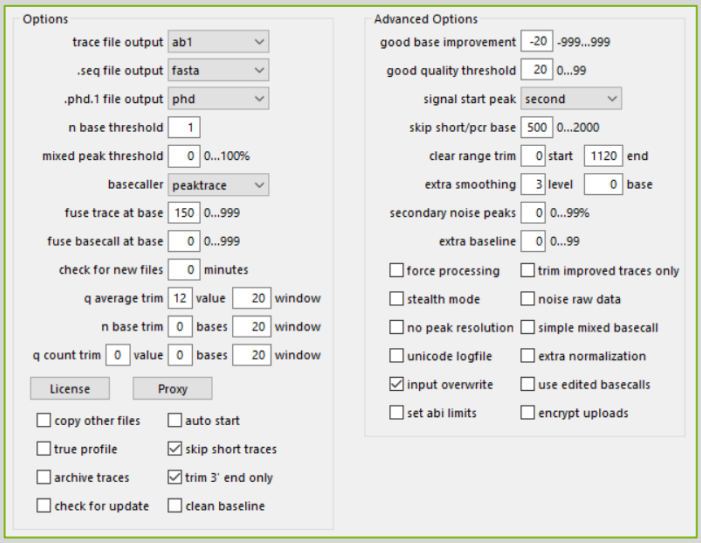
1. High quality data – Length of read up to 1100 bp, data accuracy typically > 99 %



2. Our specialists monitor every single trace processed in our lab before sending to you!

Our experts check every electropherogram we generate and optimize data analysis settings, including but not limited to choosing the proper basecaller and peak start and stop positions to get the most of it.

We believe that quality matters first!
Knowing how important results are to you we insist on human touch. Traditional it may be, we have chosen our way. After all, have you ever seen how many parameters professionals can set when analyzing Sanger traces?



3. Direct access to qualified, professional and experienced personnel

At SEQme, you can talk directly to people involved in processing your samples. And this is something we really pride on. After all, we all have experience talking to call centers and anonymous operators, have we not? But not with us!

… meet our people at www.seqme.eu/company

We strive to spread our knowledge and advice on how to get the most of Sanger Sequencing technology

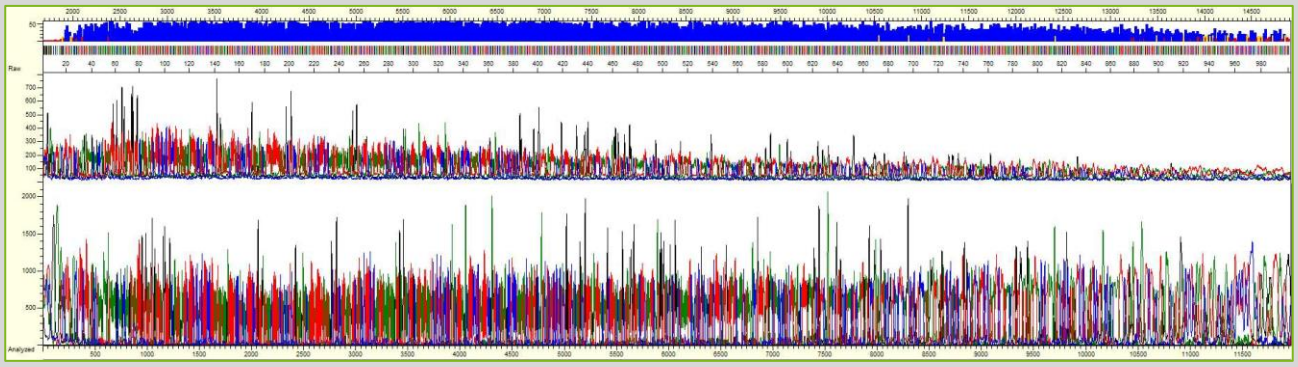
Sanger Sequencing Magazine at SEQme website

We encourage collaboration and the open exploration of ideas. We therefore share our know-how openly not only with our customers but all users interested. Therefore, we publish our Magazine on Sanger sequencing tips and tricks.

Here are some examples:

How do I sequence large templates directly?

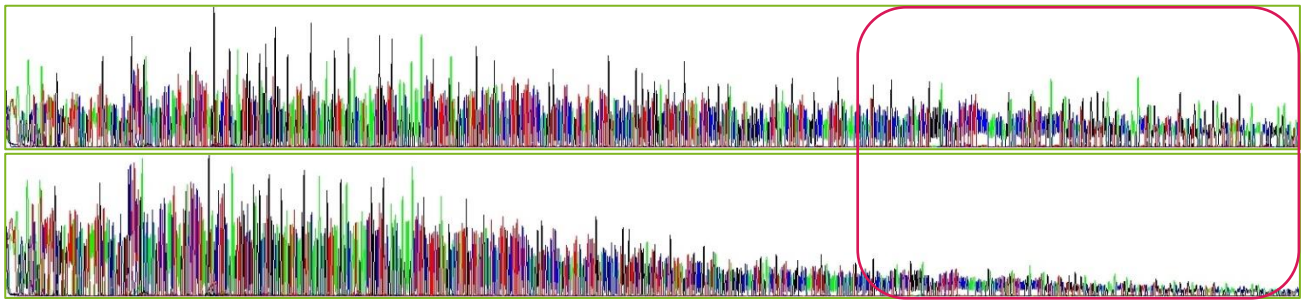
Similarly, as for standard samples also here the key prerequisite is the template amount. For example, if you consider sequencing of bacterial chromosomal DNA having a size of approx. 3-4 Mb you must supply approx. 3 µg of this DNA mixed together with 25 pmol of your sequencing primer …



Direct chromosomal DNA sequencing, bottom pane: raw data, upper pane: electropherogram with excellent sequence up to 1000 bp

Salts and other inhibitors in the sequencing reaction:

The presence of inhibitors has a strong influence on polymerase processivity during sequencing reaction and it can kill the reaction completely. We have run two reactions to show you this effect – the picture below shows two samples differing only in NaCl concentration, there is no other difference there. Please note the decline of signal in one of electropherograms! It speaks for itself.



Upper pane - Correct concentration of salts, Bottom pane - Too much salts – Signal dies out…

… check all our posts at www.seqme.eu/magazine

Expert Sanger sequencing training for expert users – to save your time and lower costs per sample

Wet lab training for lab personnel and instrument operators

Many labs use genetic analyzers by Applied Biosystems for years. Present users and operators of these instruments may have never participated at training which had been delivered long time ago (typically after the instrument installation) and therefore users and operators may lack the necessary knowledge, instruments are not effectively used, and analysis costs are too high.

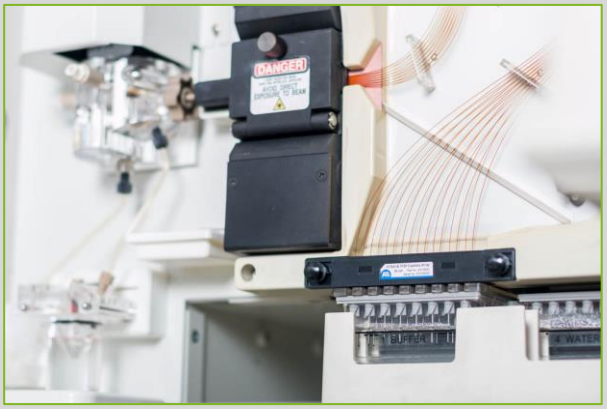
Even if you do not possess your own sequencer, you may still be interested in mastering the old but widely used technique of Sanger sequencing. All Sanger sequencing and instrument operator trainings are always organized in your lab. The training is conducted by Applied Biosystems certified application specialist and the training agenda is tailored to your needs.

Instruments supported - Sequencers / genetic analyzers Applied Biosystems:

- ABI Prism 310
- Applied Biosystems 3100 / 3100 Avant
- Applied Biosystems 3130 / 3130xl
- Applied Biosystems 3500 / 3500xl
- Applied Biosystems 3730 / 3730xl

Training agenda – Customized to your needs, typically includes:

- Principles of Sanger sequencing, chemistry, kits, template preparation, cycling reaction and cleanup
- Get to know your instrument – capillary electrophoresis, DataCollection software, reagents and consumables
- Data analysis – Sequencing analysis software, GeneMapper (including ID versions), etc.
- Troubleshooting



We have long experience with servicing and technical support of DNA sequencers by Applied Biosystems.

Our trainers possess certificates provided by the manufacturer.

… find all our events at www.seqme.eu/courses

Deep insight into software algorithms – understand and customize your data analysis tools and make your daily routine smart

Data analysis software training

We – like you – are not software developers but users. And users can really benefit from software tools only if they understand them to the very last detail. Knowing not only what parameters you need to set but also why and what effect your settings have on the output is definitely a must. We have tested and mastered various software packages for Sanger sequencing and fragment analysis in our daily routine and – according to our philosophy – we are eager to share our know-how.

In our courses on Sanger sequencing and fragment analysis data processing we use software tools by Applied Biosystems exclusively. We have selected these tools carefully and you will learn why during the course. Of course, you will also learn how to use them efficiently.

Our courses on data analysis tools:

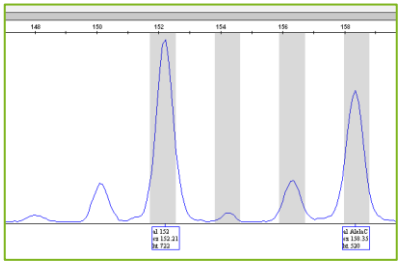
- Software by Applied Biosystems
  - Sequencing Analysis
  - SeqScape / VariantReporter
  - GeneMapper
  - GeneMapper ID

Training agenda – Customized to your needs, typically includes:

- Theoretical session on software architecture and algorithms
- Hands-on session demonstrating basic software workflow for various applications
- Hands on session showing tips and tricks how to analyze suboptimal data and perform troubleshooting
- Hands on session using your own data – to make you feel you are in your lab

Our trainings are either public and you are expected to bring your own laptop with a valid software license (demo license, if applicable) or alternatively our trainers come to your lab to run a private training session for you.

You choose the format, we deliver the content.



… find your course at www.seqme.eu/courses or contact us!

There is nothing more rewarding than a satisfied customer inquiring about our services again…

Testimonials

Libor M., Institute of Molecular Genetics, Prague
We are very happy with sequencing results of our hairpins. Thanks for your excellent service.

Astrid H., Biology Centre, Budweis
...but how to best test a new sequencing service if not with difficult samples? You have done a very good job. We are very happy about the change from Macrogen to SEQme!

Dusan T., Mendel University, Brno
I want to thank you for the great job you do – data quality, read length and time to results, everything is extraordinary.

Tomas C., Institute of Soil Biology, Budweis
... you did excellent job, the data are simply great. After your notice that direct sequencing of chromosomal DNA may not yield optimal results I was very surprised by the data I received. Perfect read length and a strong signal. Hats off and many thanks, this has helped me a lot.

You know what you want.
We know how to do it.

We will be happy to offer our Sanger sequencing and Fragment analysis services to you – give it a try!

Quick Links

? Choose your service at www.seqme.eu/sanger-sequencing

? Order on-line at www.seqme.eu/order

? Follow our Sample submission guidelines at www.seqme.eu/documents to prepare your samples

? If you need an advice, see www.seqme.eu/magazine or check our Q & A at www.seqme.eu/sanger-sequencing/instructions

? If looking for an expert training, choose from our portfolio of courses at www.seqme.eu/courses

? Interested in prepayment of our services? Available on line after registration.

? Another question? See our contact details at www.seqme.eu/company or go to www.seqme.eu/contact-us

info@seqme.eu | www.seqme.eu