



MTP sequencing (96/384-well sequencing)



Genomics

Microtiter plate (MTP) sequencing is also known as 96/384-well sequencing.

Guideline content

1. Online ordering / Registration
2. Completing the online order form
3. Sample shipment
 - a) Plasmids (bacterial colonies on agar)
 - b) PCR products or plasmid DNA
 - c) Primers
4. Acknowledgement of receipt
5. Processing samples
6. Delivery of sequence data

1. Online ordering / Registration

- Go to the LGC Genomics online ordering system at www.lgcgenomics.com
- Click on the button 'new user account'
- Fill in the registration forms as completely as possible
- Please note that the email address you have entered will be used for all subsequent communication. It is therefore recommended that each user of the service has his/her own account.

2. Completing the online order form

- To ensure smooth processing it is recommended that the 'remarks' field in the online order form is used for the sequence(s) of the primer(s) in case the sender is not the account owner an alternative email address can be provided during the online ordering process.
- Orders without a completed accompanying online order form cannot be processed.
- For any questions regarding completing the online order form, please contact us on **Tel: +49 (0)30 5304 2240**.

For further information please contact us on
Tel: +49 (0)30 5304 2240 or genomics@lgcgenomics.com

3. Sample shipment

- Special sample bags for safe shipping of the MTPs can be ordered at the LGC Genomics online ordering system at www.lgcgenomics.com
- Please ensure that each MTP is labeled with the customer's name, the customer code and the plate name as defined in the online order form.

LGC Genomics offers courier pick-up services for MTPs upon request.

Samples plates for sending agar stabs and PCR fragments or plasmid DNA are also available upon request.

Please send your samples to:

via courier service

LGC Genomics GmbH
Ostendstr. 25 / TGS Haus 8
12459 Berlin
Germany

via post (envelopes only)

LGC Genomics GmbH
P.O. Box 940327
12443 Berlin
Germany

3 a) Plasmids (bacterial colonies on agar)

- Please use regular MTPs with a flat or round bottom, not PCR plates, total volume of 350 µl / well.
- Pour 200 - 250 µl of 1.5 % LB-medium agar, together with the appropriate antibiotic, into the wells. Non-sterile plates are sufficient if you take them directly from the original package. Avoid overfilling and bubbles in the agar.
- Inoculate single colonies from a fresh plate using a sterile toothpick by stabbing it into the agar. After inoculation seal the plate loosely with an appropriate plastic cover and incubate the microtiter plate at 37°C overnight.
- We do not recommend the transfer of small amounts of a liquid bacterial culture to the wells filled with agar.
- To prepare the MTP for transport, please secure the cover with tape.
- Please ensure you include information about the vector used (antibiotic resistance).
- Please quote special requirements e.g. low copy number vector, high GC content.

For further information please contact us on

Tel: +49 (0)30 5304 2240 or genomics@lgcgenomics.com

3 b) PCR products or plasmid DNA

- Optimal volume of each sample is 20 μ l.
- Please use 8-strip caps to seal the plates.
Film or foil sealing of plates is not sufficiently secure for shipping and the plates are likely to leak. Use mechanically stable plates (skirted plates).
- Since an equal volume of sample will be used for all sequencing reactions, please ensure that the fragments do not vary in size by more than a factor of three (i.e. the largest fragment is not more than three times the size of the smallest) and the plasmid DNA should have a similar DNA concentration.
- To ensure high quality results, PCR products have to be at least 150 bp in length. A size larger than 300 bp is recommended.

3 c) Primers

- Please provide at least 10 μ l of stock solution of the respective primer for each 96-well plate ordered (100 μ M [100pmol/ μ l] in 'safe-lock' tubes individually labelled with **name and concentration** of primer).
- Please note that we accept a maximum of 6 primers in tubes for each MTP. Templates for given primers have to be arranged in blocks (either in lines or rows). Where more than 6 primers are required or an arrangement in lines or rows is not possible, we recommend that a second - a 'mirror' plate - of primers is used (primer in position A1 for template A1, primer in A2 for template in A2...etc...primer in H12 for template in H12). The mirror plate should comprise 15 μ l primer solution (5 pmol/ μ l [5 μ M]).
- For the primer plate, please use 8-strip caps to securely seal the plate.

Free universal primers:

M13-21F	TGTA AACGACGGCCAGT
M13-29R	CAGGAAACAGCTATGACC
Sp6	ATTTAGGTGACACTATAG
T7	TAATACGACTCACTATAGGG
pAD-Gal4-AD	GTTTGAATCACTACAGGGATG

Continued free primer list see next page.

For further information please contact us on
Tel: +49 (0)30 5304 2240 or genomics@igcgenomics.com

Free universal primers (continued):

27F	GAGTTTGATCMTGGCTCAG
1492R	GGYTACCTTGTTACGACTT
ITS1	TCCGTAGGTGAACCTGCGG
ITS4	TCCTCCGCTTATTGATATGC
pGEM1	CGACTCACTATAGGGCGAATTGGG
pGEM2	TTGGGAGCTCTCCCATATGGTCG
pQE276	GGCAACCGAGCGTTCTGAAC
pQE65	TGAGCGGATAACAATTTACACAG

Customer primers can be synthesised and stored upon request.

4. Acknowledgement of receipt

On arrival at LGC Genomics in Berlin, the plates are visually checked for any transport-related physical damage and the customer (sender) is contacted by email or phone.

5. Processing samples

As a special service – and at no extra charge to the customer – LGC Genomics carries out a clean-up treatment of all PCR products and performs a mini plasmid prep for all bacterial colonies received.

An additional quality check of the samples on agarose gels is performed and the samples are stored for 3 months as part of our standard operating procedures.

6. Sequencing results

The customer will be notified via email when the sequencing data are available in the download area of the LGC Genomics online order system, which is protected by password. We provide the chromatograms (.abi and .scf files) and the text sequence (.fsa file) extracted by our software.

For further information please contact us on
Tel: +49 (0)30 5304 2240 or genomics@lgcgenomics.com

LGC Genomics offers
Next generation sequencing

Please check our webpage
www.lgcgenomics.com
for further information



LGC Genomics GmbH

Ostendstrasse 25 • 12459 Berlin • Germany

Tel: +49 (0)30 5304 2240 • Fax: +49 (0)30 5304 2201

Email: genomics@lgcgenomics.com

Web: www.lgcgenomics.com

LGC Genomics

Queens Road • Teddington • Middlesex TW11 0LY • UK

Tel: +44 (0)20 8943 7000 • www.lgcgenomics.com