Microsynth

Barcode High-Throughput Run (Prepaid Service)

Sample Preparation

Plasmids, Purified and Non-Purified PCR Products
PCR reactions can be sent directly after PCR in their reaction buffer at room temperature (RT) in the barcoded 96-well plate. The only recommendation is that you check the quality of the sample on an agarose gel (a random subset is sufficient; at least one sample per PCR primer pair) before shipment. To obtain optimal sequencing results it is important that all samples are in the recommended concentration range.

E. coli Cells (for Isolation of Plasmids at our Laboratories)
If you send us E.coli cells, we recommend you that you ship them in the barcoded 96-well plate within Luria Broth (LB) medium at RT. Please make sure that your cells are incubated in 130 µl LB medium (containing the appropriate antibiotics!) for at least 3-4 hours with gentle shaking at 37° C prior to shipping. If no shaker is available at your lab, please let us know. We then perform a longer incubation at our site.

Glycerol Stocks (Additional Service for a Fee)
Please indicate in advance if you want us to make glycerol stocks for you. Shipment of glycerol stocks to customer will be on dry ice. Alternatively, glycerol stocks can be produced in your lab after an overnight incubation.

Shipment
Safe shipment of liquid cultures or solutions requires good sealing of your 96-well plate. It is recommended that you seal your barcoded 96-well plate with 8-cap stripes, 96-cap mats or heat sealing.

General Information
When forwarding us purified plasmids or PCR products, please send them in liquid form at RT in pure water, 10 mM Tris-HCl (pH 8.0) or 10 mM Tris-HCl (pH 8.0) with a maximum of 0.5 mM EDTA. **TE buffer (10 mM Tris-HCL, 1mM EDTA) might cause sequencing problems.** We strongly recommend that you measure the concentration of a random subset of samples. Again, it is very important that all samples fulfill the requested ranges of DNA concentration.

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Sample Amounts Per Sequencing Reaction & Concentrations

DNA Template	Concentration	Effective Amount (12 µl)	Pipetting Scheme for Pre-mixed Option
Plasmid	**40-100 ng/µl[1]**	**480-1'200 ng**	**12 µl DNA template solution + 3 µl sequencing primer solution**
PCR[2]	**18 ng per 100 bases in a volume of 12 µl**		
PCR (200bp)	**3.0 ng/µl**	**36 ng**	
PCR (300bp)[2]	**4.5 ng/µl**	**54 ng**	
PCR (400bp)[2]	**6.0 ng/µl**	**72 ng**	
PCR (>400bp)[2]	**etc.**	**etc.**	
Primer (premixed) **Primer separate**	**2 pmol/µl** **10 pmol/µl = 10µM**	**30 pmol** **-**	

[1] Optimal plasmid concentration is 80 ng/µl.

[2] Regardless of whether the PCR is purified or non-purified

***Remark:** Direct primer synthesis at Microsynth possible*

Order Form Completion

Prior to shipping your sequencing samples to Microsynth, please proceed as follows to complete your order form:

1. Enter our webshop on www.microsynth.ch (click on "LOGIN SHOP")
2. Click on „**Plate Sequencing**" in the green DNA Sequencing area
3. Click on "**Fill Order Form**" under Barcode High-Throughput Run[1,2]
4. Fill in the order form and finally submit your order
5. Pack your samples + the printed order form (**very important!**) into any type of transparent plastic bag
6. Drop your sample package into the closest Microsynth sample drop box or alternatively use our prepaid envelopes for mail shipment

[1] In case you need to define more than one primer source (e.g. 72 samples shall be sequenced with a standard primer from Microsynth's Standard Primer List whereas 24 samples require a specific sequencing primer from your Custom Primer List), please start with **"Fill Order Form (Multiple Primer Sources)"**.
[2] Addtional primer: Copy plate to use same sample pattern for another primer (we receive physically one plate but two electronical plates.

Need More Information?

In case you need help or more information, please do not hesitate to

- call us at +41-71-726 10 03
- or email us at sanger.support@microsynth.ch

We are looking forward to receiving and sequencing your samples.