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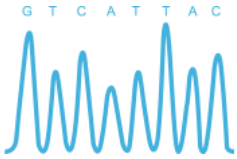
DNA SEQUENCING SAMPLE SUBMISSION GUIDELINES



For the best Sanger sequencing results, GENEWIZ strongly recommends you follow our Sample Submission Guidelines as closely as possible. If you have any questions, our Technical Support team is here to help!

Email (mailto:DNASeq@genewiz.com) | Phone (1-877-GENEWIZ (436-3949), option 2) | Live Chat

US SAMPLE PREPARATION

To prepare samples for DNA sequencing, please follow these easy steps:

1. For orders with <48 samples, please use 8-strip PCR tubes to streamline preparation and processing. Label your tubes on the side with your initials and sample number. For orders with ≥48 samples, you can receive a discount by using a 96-well PCR plate and arranging the samples vertically (A1 to H1). See the Tubes and Plates section for more details.
2. Dilute your sequencing primer to 5 µM (pmol/µl) using water. You will need 5 µl for each sequencing reaction. If you want to use a GENEWIZ Universal Primer, we will add it for you at no charge. Remember that only one primer is used in a sequencing reaction. See the Technical Notes section for tips on designing primers for sequencing.
3. For the amount of template needed in each of our DNA Sequencing Services (Pre-Mixed, Pre-Defined, and Custom), please refer to the tables below. Prepare template in 10 µl for each sequencing reaction. Please make dilutions in water or Tris. For best results, do not use Tris-EDTA (TE) because EDTA will inhibit the sequencing reaction.

UK SAMPLE PREPARATION

To prepare samples for DNA sequencing, please follow these easy steps:

1. For orders with <48 samples, please use 8-strip PCR tubes if you can, to streamline preparation and processing, but we also accept other containers. Label your tubes on the side with your initials and sample number. For orders with ≥48 samples, you can receive a discount by using a 96-well PCR plate and arranging the samples vertically (A1 to H1). See the Tubes and Plates section for more details.
2. Dilute your sequencing primer to 5 µM (pmol/µl) using water. You will need 1 µl (minimum volume of 10 µl) for each sequencing reaction. If you want to use a GENEWIZ Universal Primer, we will add it for you at no charge. Remember that only one primer is used in a sequencing reaction. See the Technical Notes section for tips on designing primers for sequencing.
3. For the amount of template needed in each of our DNA Sequencing Services (Pre-Mixed, Pre-Defined, and Custom), please refer to the tables below. Prepare template in 10 µl for each sequencing reaction. Please make dilutions in water or Tris. For best results, do not use Tris-EDTA (TE) because EDTA will inhibit the sequencing reaction.

US & UK PreMixed

In the same tube, mix template (10 µl) and your primer (5 µl) according to the table below. To use a GENEWIZ Universal Primer, simply submit template at the requested concentration in 10 µl. See the Technical Notes section for tips on how to purify PCR products

DNA Type	DNA Length (include vector)	Template Concentration in 10 µl	Template Total Mass	Your Primer Total Picomoles	Premixed Volume* (Template + Your Primer)
Plasmids	<6 kb	~50 ng / µl	~500 ng	25 pmol	15 µl
	6 - 10 kb	~80 ng / µl	~800 ng		
	> 10 kb	~100 ng / µl	~1000 ng		
Purified PCR Products	<500 bp	~1 ng / µl	~10 ng	25 pmol	15 µl
	500 - 1000 bp	~2 ng / µl	~20 ng		
	1000 - 2000 bp	~4 ng / µl	~40 ng		
	2000 - 4000 bp	~6 ng / µl	~60 ng		
	> 4000 bp	Treat as plasmid	Treat as plasmid		

*If you use a GENEWIZ Universal Primer (/Public/Resources/Free-Universal-Primers), submit the required amount of template in 10 µl.

*For PCR products purified with an enzymatic cleanup protocol (e.g. ExoSAP-IT), a spectrophotometer cannot be used for accurate DNA quantitation. Reaction components, such as degraded primers and nucleotides, will absorb UV light and inflate the calculated DNA concentration. Instead, use the band intensity on an agarose gel relative to that of mass standards to estimate DNA concentration. Alternatively, a fluorometer, which employs a dye that specifically binds dsDNA, can provide accurate quantitation.

US Pre-Defined

In separate tubes, provide template (10 µl) and your primer (5 µl) according to the table below. See the Technical Notes section for tips on how to purify PCR products.

DNA Type	DNA Length (include vector)	Template Concentration in 10 µl	Template Total Mass	Template Volume Per Reaction	Your Primer Concentration µM (pmol/µl)	Your Primer Volume Per Reaction
Plasmids	<6 kb	~50 ng / µl	~500 ng	10 µl	5 µM	5 µl
	6 - 10 kb	~80 ng / µl	~800 ng			

	> 10 kb	~100 ng / µl	~1000 ng			
Purified PCR Products	<500 bp	~1 ng / µl	~10 ng	10 µl	5 µM	5 µl
	500 - 1000 bp	~2 ng / µl	~20 ng			
	1000 - 2000 bp	~4 ng / µl	~40 ng			
	2000 - 4000 bp	~6 ng / µl	~60 ng			
	> 4000 bp	Treat as plasmid	Treat as plasmid			

UK Pre-Defined

In separate tubes, provide template (10 µl) and your primer (5 µl) according to the table below. See the Technical Notes section for tips on how to purify PCR products.

DNA Type	DNA Length (include vector)	Template Concentration in 10 µl	Template Total Mass	Template Volume Per Reaction	Your Primer Concentration µM (pmol/µl)	Your Primer Volume Per Reaction
Plasmids	<6 kb	~50 ng / µl	~500 ng	10 µl	5 µM	5 µl
	6 - 10 kb	~80 ng / µl	~800 ng			
	> 10 kb	~100 ng / µl	~1000 ng			
Purified PCR Products	<1000 bp	~15 ng / µl	~75 ng	5 µl	5 µM	2 µl
	1000 - 2000 bp	~25 ng / µl	~125 ng			
	2000 - 3000 bp	~35 ng / µl	~175 ng			
	> 3000 bp	Treat as plasmid	Treat as plasmid			

*For PCR products purified with an enzymatic cleanup protocol (e.g. ExoSAP-IT), a spectrophotometer cannot be used for accurate DNA quantitation. Reaction components, such as degraded primers and nucleotides, will absorb UV light and inflate the calculated DNA concentration. Instead, use the band intensity on an agarose gel relative to that of mass standards to estimate DNA concentration. Alternatively, a fluorometer, which employs a dye that specifically binds dsDNA, can provide accurate quantitation.

US & UK Custom

With this service, GENEWIZ will determine template concentration, optimize for sequencing, and mix with primer. Please provide template concentration if available.

Choose this option to sequence plasmids or purified PCR products of unknown concentration, un-purified PCR products, bacteria, phage with circular genome, and BAC DNA. For details on sequencing bacteria, phage with circular genome, and BAC DNA, please refer to the Bacteria and Phage section.

DNA Type	DNA Length (include vector)	Template Concentration in 10 µl	Template Total Mass (recommended)	Template Volume Per Reaction	Your Primer Concentration µM (pmol/µl)	Your Primer Volume Per Reaction
Plasmids	Any size	Unknown	At least 500 ng	10 µl	5 µM	5 µl
PCR Products*	Any size	Unknown	At least 500 ng	10 µl	5 µM	5 µl

*(US) If you are sending unpurified PCR products, please send 10 µl of undiluted crude product x (# reactions) and provide a gel image of the product to determine concentration.

*(EU) If you are sending unpurified PCR products, please send 30 µl of undiluted crude product x (# reactions) and provide a gel image of the product to determine concentration.

GENEWIZ UK SANGER SERVICE

For customers submitting Sanger samples to the GENEWIZ Takeley (UK) site, please use the following link (/en/Public/Company/Europe) to access ordering information for our UK laboratory. If you have any questions, please contact GENEWIZ Europe customer support at +44 (0) 1279 873837.

SHIPPING INSTRUCTIONS

1. GENEWIZ Drop Box or Fridge - Place your samples and a copy of your order form in a GENEWIZ drop box or fridge before the cut-off time. Call 1-877-436-3949 for your nearest GENEWIZ drop-off location.
2. GENEWIZ Courier Service - Request for courier pickup if available at your location. Please call 1-877-436-3949 to find out if a GENEWIZ courier picks up at your site. For San Diego customers within the pickup zone, specify the pickup location when placing your order. Note that GENEWIZ couriers pick up room temperature samples only.
3. If a drop box or courier is not available, and for International shipments, ship your samples via overnight delivery service (FedEx if possible) to us at the following location:

   GENEWIZ
   115 Corporate Blvd.
   South Plainfield, NJ 07080
   USA
   908-222-0711

UK SHIPPING INSTRUCTIONS

1. GENEWIZ DROP BOX: Place your samples and a copy of your order form in the GENEWIZ Drop Box before the cut-off time. Call +44 (0)1279 873 837 (or 0811 23 00 01 from France) for your nearest GENEWIZ Drop Box Location.