

GenHunter Corporation

624 Grassmere Park Dr, Ste 17
 Nashville, TN 37211
 Hours of Operation: 9am-5pm M-F
 Vanderbilt Pickups: 10:30am-Noon M-F

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 Toll-free: (800) 311-8260
 Email: sequencing@genhunter.com
 Web: www.genhunter.com/sequencing

SEQUENCING INFORMATION

HOW TO ORDER

Email the [order form](#) to us - we pick up at Vanderbilt from 10:30am-noon & accept drop-offs from 9am-5pm.

Deadlines for Next-Day Results <i>Results are usually next-morning, 1-3 business days max</i>		Location
Order Form	Submit by 10 am (M-F)	From the convenience of your computer
Sample Pickups at Vanderbilt	Pickups are from 10:30 am-Noon (M-F) Samples need to be ready by 10:30 am	At your lab, create a designated pickup location with a GenHunter-labeled box or rack in a -20C, 4C, or cold room for our courier to find. Place tubes in the same order as they occur on form.
Sample Drop-offs at GenHunter	Deliver samples to us by noon (M-F)	624 Grassmere Park Dr, Ste 17 Nashville, TN 37211

SAMPLE PREP - Use a high-quality purification kit and elute DNA with nuclease-free dH₂O (*instead of the kit buffer*)

PRIMER PREP - Primers should be exact match with template, 17-25 bp length, annealing site 30+ bp from target
 To convert ng/μL to μM: $\mu\text{M} = [(\text{ng}/\mu\text{L})/(\text{Molecular Weight})] \times 1000$ {Molecular Weight ≈ (# bases in primer) x 32}

TYPE <i>Samples or Primers</i>	RECOMMENDED CONCENTRATION <i>Dilute samples if > 50% above range</i>	MIN. VOLUME PER TUBE <i>Provide a separate tube for each reaction: 2 rxns = 2 sample tubes + 2 primer tubes (if not using stock)</i>	MIN. VOLUME PER TUBE WHEN USING THE SAME SAMPLE OR PRIMER FOR > 8 RXNS <i>Provide only 8 tubes but with extra volume per tube</i>
PCR Products	20 - 40 ng/μL	8 μL	8 tubes w/ 8 μL for 1st rxn + 8 μL per additional rxn <i>Example: 16 rxns = 8 tubes w/ 16 μL each</i>
Plasmids	100 - 300 ng/μL	8 μL <i>10 μL if conc. < 50 ng/μL</i>	Same as above <i>Sub 10 μL if low concentration</i>
BACs & gDNA	500 - 1000 ng/μL	8 μL <i>10 μL if conc. < 500 ng/μL</i>	Same as above
Primers	1 - 2 μM	8 μL	8 tubes w/ 8 μL for 1st rxn + 2 μL per additional rxn <i>Example: 16 rxns = 8 tubes w/ 10 μL each</i>

STOCK PRIMERS - Supplied by us (no prep needed)!

T7	TAATACGACTCACTATAGGG	M13F (-20)	GTAAAACGACGGCCAG	(our default M13F)
T7-Rev	CTAGTTATTGCTCAGCGGTG	M13F (-40)	GTTTCCCAGTCACGAC	
T3	ATTAACCCTCACTAAAGGGA	M13R	CAGGAAACAGCTATGAC	
CMV-For	CGCAAATGGGCGGTAGGCGTG	SP6	ATTTAGGTGACACTATA	
BGH-Rev	TAGAAGGCACAGTCGAGG			(Primers are also available for GenHunter vectors pAptag & PCR-TRAP)

STANDARD SEQUENCING (TUBES) - Submit samples and primers in separate 1.5 mL tubes or 8-strip PCR tubes

- **1.5 mL flip-top tubes**
 - Label lids clearly with the sample or primer name (brief enough to be readable) **OR**
 - Label lids clearly with your initials and number of sample or primer (indicate sample or primer with S or P), and on the side of one tube also include your lab name
- **NEW! 8-strip PCR tubes** (for exact multiples of 8: no partial strips or empty tubes)
 - Label the side of the 1st tube of each strip with your lab name (may be abbreviated), your initials, and the number of that sample (S1, S9, etc) or primer (P1, P9, etc) - no need to label every tube, use tape if needed

BULK SEQUENCING (96-WELL PLATES) - Submit multiple samples or primers in separate 96-well plates. A single sample or primer that is to be used for all 96 reactions may be submitted in a single tube.

- Use this [order form for plates](#)
- Aliquot vertically (A to H) to correspond with the wells on the order form (leave no empty spaces between wells)
- Spin down, seal, and freeze plate prior to submission
- Label the side of the plate with your lab name, initials, and any other info needed to differentiate multiple plates

RESULTS - Usually ready the following morning, and guaranteed within 1-3 business days

- Sequencing text files and chromatograms (.seq and .ab1 files) are emailed upon completion
- Chromatograms (.ab1 files) can be viewed on free programs such as [ABI Sequence Scanner](#) or [Chromas Lite](#) (for PC), or [FinchTV](#) (for Mac or PC).
- Repeats are run as soon as space is available, with one free repeat per reaction as needed
- If results will take more than one business day, you will receive a notification with an estimated time of arrival
- If you did not receive your results the next day and did not receive a notification stating when you may expect them, please do the following: check your spam folder, add us to your address book to avoid future emails from going to spam, and let us know that you did not receive your results so we may re-send them immediately.

SAVING TUBES - Tubes are automatically kept for 2 weeks from the submission date

- You may request any tubes submitted within the past 2 weeks to be used for additional reactions
 - Simply submit a new order form as well as any tubes not already in our possession
- **NEW!** Request tubes to be saved for a longer period, such as primers that are frequently used for your lab so you do not have to re-submit them each time!

BILLING - Invoices are sent the following month (once a month), with new accounts set up automatically

- Complete the billing information on the order form prior to submission, including the purchase order or credit card information
 - Purchase orders - submit to your institution for approval, and provide the purchase order number on the form. If waiting to receive the PO number, you may enter it as "pending" on the form and provide it to us once received and before the end of the month
 - Credit cards - for security, submit credit card information over the phone and enter it as "on file" on the form with only the expiration date and CCV number shown

TROUBLESHOOTING - Use this quick guide to help prevent some common issues

- Run samples on an agarose gel to check the concentration and quality of DNA to help prevent noisy data, failed reactions, or short reads
- Prep samples with high-quality kits such as Qiagen, and be sure to use fresh reagents and elute with dH2O instead of buffer to help prevent noisy data or short reads
- Check samples for presence of multiple priming sites or templates, which will cause mixed/overlapping reads
- Confirm that the primer is an exact match with the template (without even one mismatched base) to help prevent failed reactions
- Use primers that have not been degraded from too many freeze-thaw cycles or from long-term storage at 4C to help prevent noisy data or failed reactions
- If you have any questions before and/or after sequencing, please let us know and we will be happy to help!