Roche use only:

Internal Reference No.: _____

Key Account Manager Email: _____

Design Specification Form

SeqCap RNA Enrichment System

Complete this form in its entirety to ensure accurate processing of your order by Roche Sequencing Solutions (Roche). Note that a NimbleDesign account is now required before completing this form (see *sequencing.roche.com/products/software/nimbledesign-software.html*). If you are ordering multiple designs, complete one form for each design. If you have questions, contact Customer Service (refer to page 6 for contact details).

Background Information

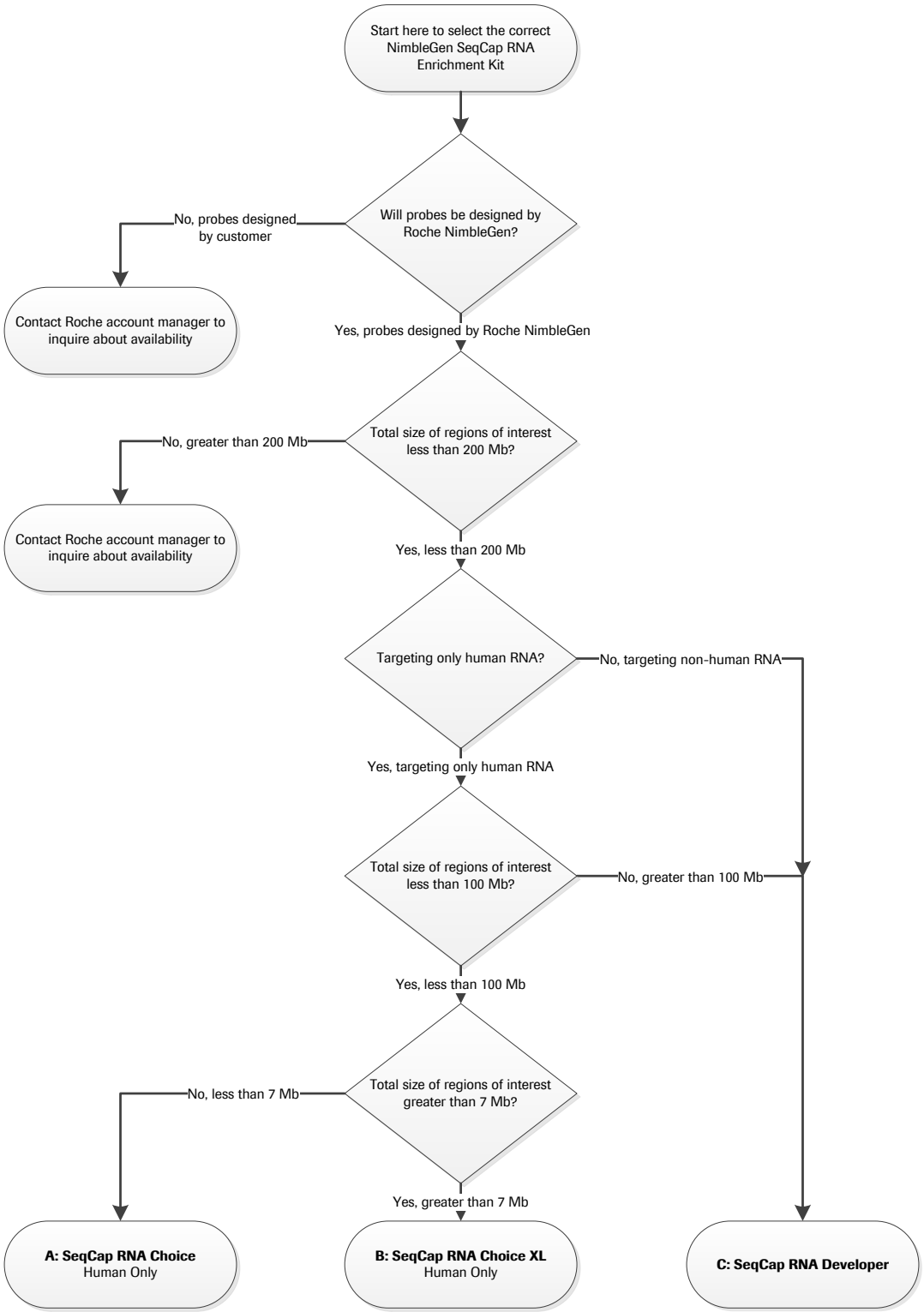
The SeqCap RNA Enrichment System provides a convenient method to enrich for targeted transcripts from cDNA in the human or any other sequenced genome. The region(s) targeted for enrichment can be a gene panel of interest or any genomic region(s) where a researcher intends to study transcriptional activity. Based on regions selected by the researcher, Roche will design capture probes using proprietary algorithms and send the design proposal to the researcher for approval. In a standard SeqCap RNA design, capture probes are designed using genomic sequences instead of transcriptome sequences in order to avoid probes that overlap splice junctions; this prevents capture bias for specific spliced variants. After the design has been approved, Roche will produce the capture probes and ship the probes to the address on the Purchase Order.

The performance of targeted enrichment methods may be unpredictable due to variability among transcriptomes and other properties intrinsic to the targeted region(s). Examples of such factors include: target sequence complexity (*e.g.* G/C content), repetitive element distribution (*e.g.* Alu, LINE, LTR), evolutionary history of the targeted region (*e.g.* conserved pseudogenes and gene family relationships), and population variation from the reference genome (*e.g.* CNV and hypervariable regions). When using a previously untested capture design, it is recommended to perform a small-scale experiment to determine the capture characteristics of a specific target before beginning a larger study.

Select Enrichment Product(s)

Three custom SeqCap RNA Enrichment System products are available, which differ in target genome, the size of the custom regions, and/or the nature of the regions of interest. Please use the flowchart on the next page to select the appropriate product(s) for your order.

Enrichment Product Selection Flowchart



ENRICHMENT KIT INFORMATION

Specify the kit quantity.

You may combine two or more configurations if the number of reactions needed is not listed below. For example, you can order two 48-reaction kits and two 12-reaction kits to get a total of 120 reactions for your design.

A. If ordering SeqCap RNA Choice kits, please enter the quantity needed in the table below.

Enrichment Kit Name	Cat. No.	Quantity
SeqCap RNA Choice Enrichment Kit, 12 Reactions	07277300001	
SeqCap RNA Choice Enrichment Kit, 48 Reactions	07279078001	
SeqCap RNA Choice Enrichment Kit, 384 Reactions	07279086001	

B. If ordering SeqCap RNA Choice XL kits, please enter the quantity needed in the table below.

Enrichment Kit Name	Cat. No.	Quantity
SeqCap RNA Choice XL Enrichment Kit, 12 Reactions	07279183001	
SeqCap RNA Choice XL Enrichment Kit, 48 Reactions	07279191001	
SeqCap RNA Choice XL Enrichment Kit, 384 Reactions	07279205001	

C. If ordering SeqCap RNA Developer kits, please enter the quantity needed in the table below.

Enrichment Kit Name	Cat. No.	Quantity
SeqCap RNA Developer Enrichment Kit, 12 Reactions	07279213001	
SeqCap RNA Developer Enrichment Kit, 48 Reactions	07279221001	
SeqCap RNA Developer Enrichment Kit, 384 Reactions	07279230001	

DESIGN INFORMATION

1. **Please specify the taxonomic name and genomic build for your design.** Provide the genome build information and source for the build. Many genome builds can be found in the UCSC Genome database (*genome.ucsc.edu*) or ENSEMBL (*www.ensembl.org*). If the genome build is not available in either of these databases, provide a link to a publicly available source for the build. If your reference sequence is not publicly available, please provide a FASTA-formatted genome sequence when possible. This reference sequence will be used to screen out probes targeting repetitive elements of the genome in order to enhance capture efficiency.

   ☐ *Homo sapiens* (HG19/GRCh37)

   ☐ *Homo sapiens* (HG38/GRCh38)

   ☐ Other: _____

   Example: *Mus musculus* (mm10) from UCSC.

2. **Provide a brief description of the design:**

   Description: _____

   Example: design targeting transcripts from a cancer gene panel, version 3.

3. **Provide a design identifier (optional).** Roche can include a design identifier of up to 10 characters in the final design name. The provided design identifier will go after the build in the final design name.

   Design Identifier: _____

   Example: CancerV3 (final design name will be 130130_HG19_CancerV3_RNA).

4. **Submit (attach) the regions of interest in one of the following formats:**

   ☐ Coordinates for the regions of interest. Coordinates must be supplied in tab-delimited BED format; (*genome.ucsc.edu/FAQ/FAQformat.html#format1*). Exact coordinates of each region are required.

   ☐ List of gene/transcript identifiers. Please provide the identifier list source (database or URL) and database version.

   Source: _____ Version: _____

   Examples: Ensembl Gene IDs, Ensembl Transcript IDs, Ensembl Exon IDs, HGNC Gene Symbols, RefSeq ncRNA IDs, RefSeq mRNA IDs, or CCDS IDs.

   ☐ FASTA formatted target sequences (if coordinates or identifiers cannot be obtained).

Regions less than 30 bp (*e.g.* micro-exons) will not be directly targeted by SeqCap RNA probes. These regions will be removed and captures of the adjacent regions will be relied upon for capture. For small target regions in the range of 30 to 50 bp in size, Roche extends the regions to 50 bp.

5. **If known, specify the total size of your regions of interest contained in the BED file and/or FASTA files you are providing:**

   Total size of regions of interest: _____

6. **Email the BED file, FASTA files, and/or list of identifiers and this form to Customer Service.** (*madison.customerservice@roche.com*).

7. **Review and approve the design.** Roche will email probe coverage summaries for each design. Instructions for viewing and approving the design are provided along with these files. Approval of the design is required before processing of your order can continue. By approving the design, you represent to Roche that you have the right to submit the data to Roche for purposes of manufacture, and confirm that the design is according to your specifications.

CONTACT INFORMATION

Provide contact information for correspondence:

Product Design

Name: _____

Company/Institution: _____

Address 1: _____

Address 2: _____

City: _____ State: _____

Zip/Postal Code: _____ Country: _____

Telephone: _____

NimbleDesign User ID: _____

Email: _____

Delivery Address

Orders are delivered to the address listed on your Purchase Order. Contact your Purchasing Agent to ensure your delivery address is up to date.

ADDITIONAL INFORMATION

Include any additional information regarding your order:

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CUSTOMER SERVICE

If you have questions about completing this form, contact Customer Service:

Email *madison.customerservice@roche.com*

TECHNICAL SUPPORT

If you have technical questions, contact your local Roche Technical Support. Go to *sequencing.roche.com/support.html* for contact information.

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Pleasanton, CA 94588 USA

Notice to Purchaser
For patent license limitations for individual products please refer to: ***www.technical-support.roche.com***.

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