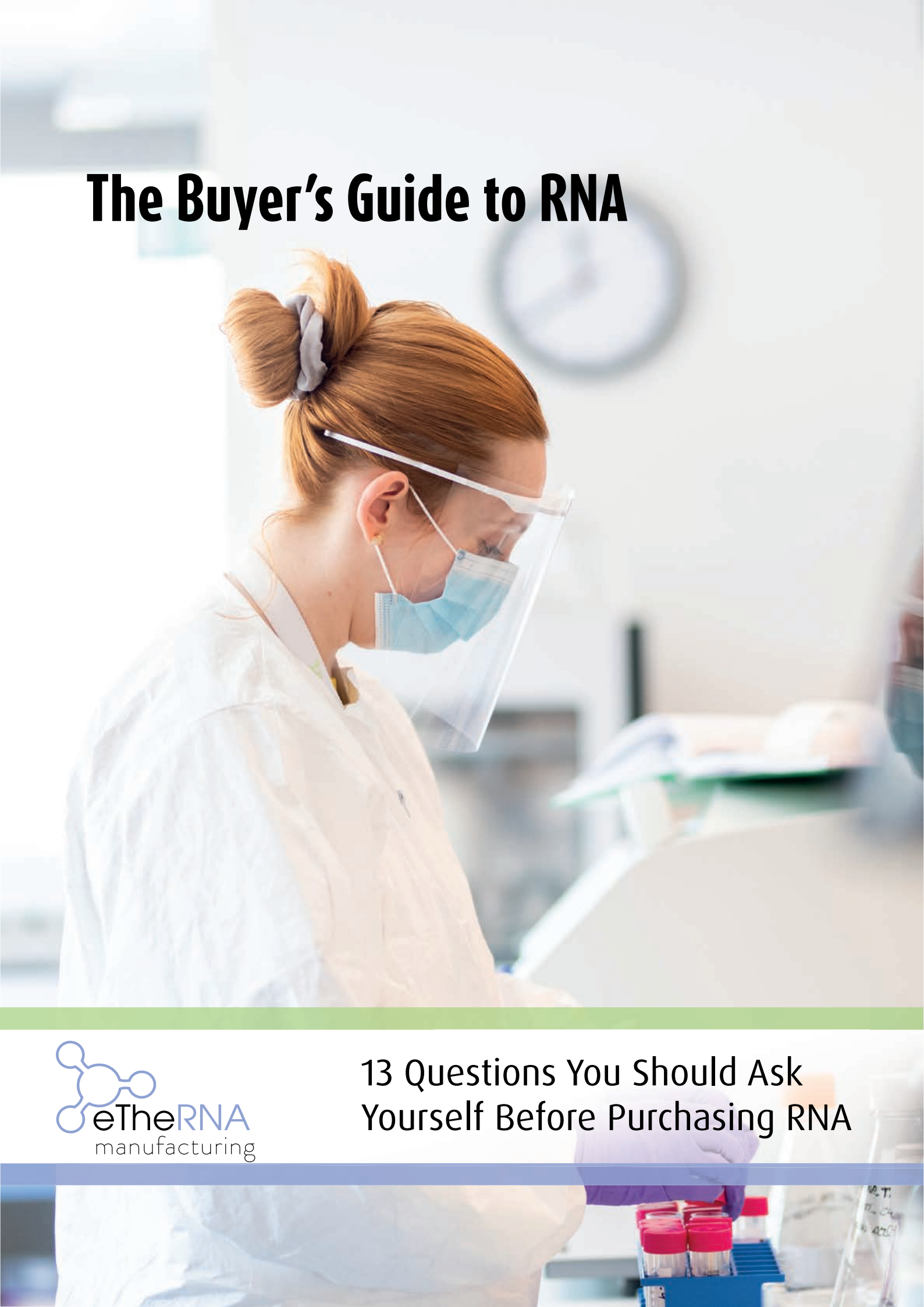
# The Buyer's Guide to RNA



eTheRNA manufacturing

## 13 Questions You Should Ask Yourself Before Purchasing RNA



eTheRNA builds on more than 30 years of research grade mRNA manufacturing, and more than 5 years of GMP mRNA production experience.

We know that ordering RNA can be a complex task and so we've put together this brief guide answering many of the common questions that we receive from our customers. We hope you find it informative. If you would like to ask us a question or talk directly, please do not hesitate to contact our Contract Manufacturing Organisation: cmo@etherna.be

You can also visit our website for more information: www.etherna.be

### What is the difference between research grade RNA and GMP grade RNA?

*GMP grade RNA is produced according to the strict rules of good manufacturing procedures in dedicated clean rooms and can be used for preclinical and clinical applications.*

*Research grade RNA is produced following the same procedure, with the same reagents but without the GMP documentation and restricted clean rooms.*

### How will RNA be shipped?

*RNA is typically stored in water for injection and shipped frozen on dry ice. During transportation the temperature is monitored and remain below -15°C, which is also the long term storage termperature.*

### How long does it take to deliver RNA?

*This depends on many factors. Of course, R&D grade RNA delivery is much faster than GMP grade. Depending on whether the production also entails the creation of the RNA production plasmid, delivery of R&D grade material can vary from weeks to months. This timeline also depends on the purification method, the amount needed, the size of the aliquots, potentially extra QC assays to performed. The production of a large batch of GMP grade RNA will take several months. Again, this will depend on many parameters such as availability of the plasmid, the intended use of the RNA, the amounts etc.*

### How can I store RNA?

*RNA is very stable when stored in water at temperatures below -15°C. For GMP material an elaborate stability program must be in place. We have stability data from previous batches up to 3 years and beyond.*

### Which types of RNA can be produced? Is the size of the RNA limited?

*Every RNA type that can be produced by in vitro transcription can be produced. This is usually mRNA, but very short RNA types such as guide RNA for CRISPR-CAS, or very long types such as Self Amplifying RNA are also possible.*

### Which cap systems are available?

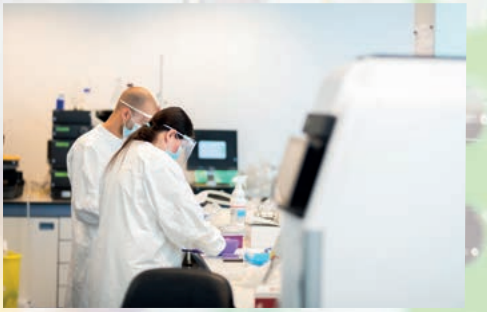
*Capping can either be performed cotranscriptionally (ARCA or CleanCap) or after in vitro transcription with Vaccinia capping enzymes. The plasmid must be suited for the chosen capping system. Some capping systems are subject to intellectual Property.*

### Are there any limitations in the amounts of RNA that can be ordered?

*Generally speaking, R&D grade RNA can be ordered from amounts as small as 1mg up to gram amounts. GMP grade production usually starts at higher quantities, preferably starting at around 100mg up to gram range. Very small or very large batches may restrict the options for purification.*

### Can I order RNA that contains modified nucleotides?

*Any modified nucleotide can be incorporated in the IVT product when commercially available. However, you should be aware that many modified nucleotides are protected by IP.*



## Which purification options are available and which one should I choose?

*Several purification methods can be offered. This can be basic silica purification for smaller amounts of R&D grade material. For GMP productions, multiple step purifications can be performed, including a specific double strand RNA removal step. The purification option chosen should depend on the intended use of the material. We can guide customers to what is needed in the specific situation.*

## Can eTheRNA provide the DNA template?

*Yes, we can assist in all phases from optimizing the sequence, cloning in an appropriate plasmid, selecting the correct clone and creating a working cell bank up to the final clinical RNA product. Some steps in this process will be outsourced to validated providers.*

## What are the key production steps in *in vitro* transcription RNA manufacturing?

*The main production steps involved in RNA manufacture include:*

*1. Synthesis of synthetic DNA*
*2. Plasmid cloning*
*3. Plasmid purification*
*4. Full DNA sequencing documentation*
*5. PCR amplification of insert, purification and characterization*
*6. In vitro transcription*
*7. Purification and characterization*
*8. Bioburden reduction or sterile filtration*

## Can I ask for specific GMP Grade RNA specifications?

*Yes, we offer the following specification options according to your requirements:*

- *Visual inspection*
- *Appearance*
- *Integrity*
- *Identity*
- *pDNA content*
- *Protein content*
- *Endotoxin content*
- *Concentration*
- *dsRNA content*
- *Sterility*
- *RNA sequence*

## What information do I need to have in order to order mRNA manufacturing services?

*Before ordering mRNA services, you should have the following information available:*

- *Target mRNA sequence or length or protein of interest*
- *Base composition or modifications*
- *Desired 5' and 3'-UTR sequence*
- *Plasmid map and scale*

eTheRNA manufacturing

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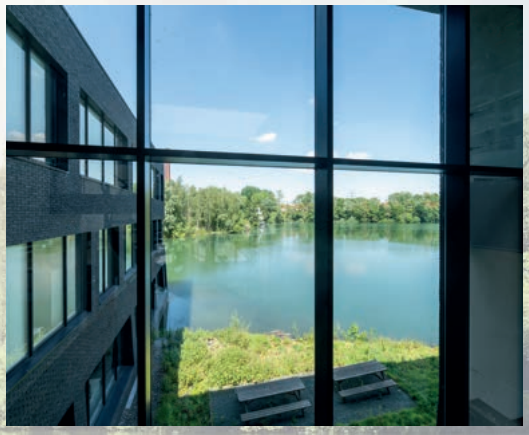
## About us



eTheRNA builds on more than 30 years of research grade mRNA manufacturing, and more than 5 years of GMP mRNA production. The initial experience was built at the labs of the VUB in Brussels. Since 2018, all RNA production is performed in the brand new GMP authorized facility in Niel, Belgium.

We now offer our expertise and capability to other parties interested in having their RNA manufactured according to their needs. We will allocate part of our capacity for external parties as a contract devel-opment and manufacturing organization.

We offer the eTheRNA expertise and capability as a one stop service to bring RNA based research and products from the lab to the clinic. Our standard approaches and processes are scalable, from ng to g level, and we produce according to customer specification and with clinical phase appropriate controls. Depending on your needs, we produce both non-GMP or research grade material and GMP grade material to be used in clinical trials.

Services can include plasmid development and production, research grade mRNA production and GMP grade mRNA production, including different possibilities with regards of process and purification.

Given the experience with own clinical trials, eTheRNA can also provide support in CMC file writing.

For a no obligation discussion about your project, please email us or visit our website for more information:

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www.etherna.be