

Frequent Questions for Protein Expression Service

1. What types of expression systems and protein services do you offer?

- *E. coli* /Yeast /Insect cell /Mammalian cell /Cell free systems
- Services: tag removal (tag-free), endotoxin removal, fermentation and C13/N15 labeling
- Upstream/downstream: gene synthesis, antibody (pAb/mAb) production, lyophilization

2. When should I use an *E.coli* expression system?

- Protein with no or only a few post-translational modifications
- High yield - easier to move forward to scale-up production to get gram level protein
- Low budget
- Downstream applications: For example, if you plan on antibody production, there is no need to produce protein with high activity and full structure.
- No issues with endotoxin level
- Refolding is required - may have issue with structure or activity

3. What do you need to get started? Can you use my construct?

We prefer to start with protein sequence. We prefer to do the codon optimization in-house. If we are able to confirm the optimized sequence then we can start with wide type gene. If you have proven expression level with a plasmid you have, and you can provide us with your expression protocol, we can begin your protein project by using your own plasmid.

4. Are there limits on what proteins you can produce?

MW: We provide guaranteed service for 10kD-80kD protein. If there are no other issues, the gene synthesis cost will be covered. Our production limitation is 8kD to 100kD, 150 kD occasionally. If this size is exceeded, there will be a non-refundable gene synthesis cost due to increased protein size. There may be a degradation issues if we produce protein with high MW. This, in turn, could affect the purity.

SP and TMD: hydrophobic domains. For specific details you can request a project outline which will indicate this for your specific protein. Typically we try and delete the domains, or ask the customer if he/she is interested in partial sequences.

Protein toxicity: This can be tricky. We would have to look closely at the sequence and evaluate the specific protein to see if the project is feasible

5. What type of QC do you provide?

For His-tagged or GST-tagged proteins: SDS-PAGE and WB for free (non-reduced and reduced gels in mammalian system). For tag-free protein: SDS-PAGE, we also recommend to add a MALDI-TOF to identify the protein (additional cost). For endotoxin level we use gel clot assay.

6. Can Biomatik produce bulk (milligrams to grams) amounts of protein?

Yes, we can do fermentation, but we begin with flask culture level production first. We need to know the exact yield. We recommend running a pilot fermentation study to see if this is possible with your protein of interest. If there is a high or moderate yield, where hundreds of mg protein needed, then it is possible to scale-up production.

7. What is your policy for cancellations?

For any protein, the customer may cancel the order after reaching an agreement:

- i) You may cancel without any cancellation fees if we have received the order but production has not begun.
- ii) If the project has begun, you are subject to pay for the completed stages upon cancellation of the order.
- iii) If the project is completed, you may cancel the order but are still subject to pay the project price without shipping costs.

8. What about Patents?

Our role is simply to produce the protein you are interested in; it is your sole responsibility to ensure there is no patent infringement.

9. What is my chance at success with a custom protein working for me?

We guarantee the project with extensive experience and over 95% success rate. Unfortunately we don't guarantee protein activity.

10. Can I track my protein's progress during the synthesis?

Yes, simply make a request when you place your order and our technical sales team will be happy to oblige.

11. How do I re suspend my lyophilized protein and how do I store my custom protein??

We recommend you resuspend in 1 ml of sterile water. For long term storage we recommend you aliquot and store at -80°C. Try and avoiding frequent freeze-thaws.

Example Citations of Custom Protein and Antibody Services:

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- Wadwa, J., Chu, Y-H., Nguyen, N., Henson, T., Figueroa, A., Llanos, R., Ackland, M.L., Michalczyk, A., Fullriede, H., Brennan, G., Mercer, J.F.B. and Linder, M.C. (2014). Effects of ATP7A overexpression in mice on copper transport and metabolism in lactation and gestation. *Physl. Rep.* **2**, e00195.