

Übung: Riboswitchin'

SS 2007 Übung zu RNA-Biochemie (RNA) (00.939)

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The last part of our 3-part Übung is a case study of a realistic problem which could occur in any molecular biology laboratory. The idea is, we give you a minimum of information – just the problem description – and you have to find your own solution.

Of course, the theoretical background for this Übung was entirely discussed during the lectures, while the practical issues should be covered by the previous Übungen.

So there is nothing really new here. You just have to consider the problem, make a plan and convert it into a practical approach.

1 Addresses

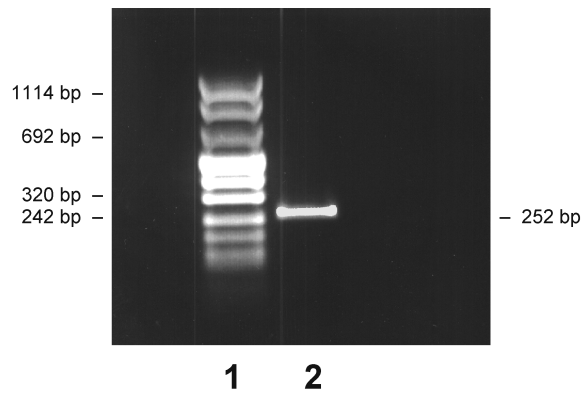
Use every tool and data source you consider to be useful for this Übung.

2 Gathering your Sequence

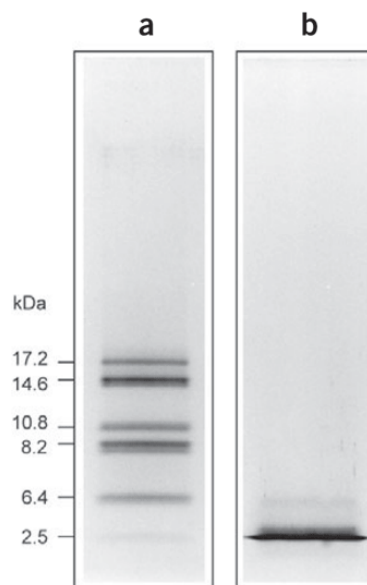
A copy of the sequence described below can be found in `/home/sbienert/data/part03/endofgene.fas`.

3 Once upon a Time...

A friend of yours, a highly trained biologist a world renowned philosopher, is trying to purify a protein, worthy of a noble prize. Since the production of this protein is known to be very low *in vivo*, she tries the molecular biology standard approach: Clone the gene of the protein into a vector, excite its expression and then purify it. After cloning it, your friend tries, as a first check for success, a gel electrophoresis of the protein's mRNA. As expected, there is a bright signal around 250nt in the following photo, which perfectly meets the length of the cloned gene (don't ask where we got your friend's photo from, just accept that we got it). The first column is the marker and column 2 is supposed to be the cDNA belonging to the gene:



Since these results are very promising, she tries to extract the translated mRNA, next, and uses a SDS-PAGE to quantify the amount of protein. For a sequence around 250nt long, she expects a signal near 6.4kDa. But as you can see on the following photo of the gel, there is only a very weak signal around that value, which also appears somewhat smeared. On lane a you find the marker and on lane b the protein is supposed to give a signal:



Because nobody is perfect, she repeats the whole experiment (gel electrophoresis & SDS-PAGE) several times until she is sure there is no mistake. She even tries to clone the gene again, but the results do not change.

Because she has no further ideas how to cope with the problem, she asks you for help as she knows that you are one of the world's top-bioinformaticians. Therefore she shows you her results of the gels and the sequence of the gene, which she has cloned into *E. coli*. You see the end of this sequence:

```
> End of the noble prizing gene
GGGTGTTCCCAAGGAGGACGTCTCGATCGTCGCTTATCGCGTTCCCTCCTGGCCATTGTTTCCTCCTTAC
```

4 Assignment

1. Give your solution to the problem to your Übungsleiter at the end of this Übung. This will not be formally marked, but is just a check that you participated in the Übung.
2. Solve the problem of your friend.