Total RNA extraction from Rhizobium etli cell or bacteroid pellets

Centre of Microbial and Plant Genetics, Katholieke Universiteit Leuven, Belgium

**CONTEXT**

The laboratory is focused on the study of the genome-wide transcriptome of *R. etli* under diverse conditions. Non-coding RNAs (ncRNAs) play a crucial role in the intricate regulation of bacterial gene expression, allowing bacteria to quickly adapt to changing environments. In this study, we have compared an extensive compilation of these non-coding RNA predictions to intergenic expression data of a whole-genome high-resolution tiling array in the soil-dwelling α-proteobacterium *Rhizobium etli* [1].

**MATERIAL**

- Precellys®24 homogenizer.
- Precellys® kit: 03961-1-005 (glass beads 0.1mm).
- Sample: *Rhizobium etli* cell pellets (from 20-40 ml of bacteria culture) or bacteroid pellets (prepared from *Phaseolus vulgaris* root nodules), RNA stabilized, immediately frozen in liquid nitrogen and stored at -80°C.
- Extraction buffer: 1mL TRIzol Plus RNA Purification kit (Invitrogen).

**PROTOCOL**

- Precellys®24: 6500 rpm, 2x45 sec – 30 sec pause.
- Analyses: RNA isolation, RNA integrity, quantity and purity, cDNA synthesis & RNA detection by tiling microarray.

http://www.biomedcentral.com/1471-2164/11/53

**RESULTS**

All samples had an RNA Quality Indicator value of 10, using Experion RNA StdSens Chips. The ncRNA peak could be detected in each sample (Figure 1). RNA quantity and purity was assessed using the NanoDrop ND-1000. The A260/A280 ratio and A260/A230 ratio of all samples were ≥ 2.

**CONCLUSION**

Transcriptomic studies require high amounts of pure intact RNA. High quality expression data were obtained using the Precellys®24 system allowing for an efficient and reproducible homogenization of different kinds of samples.