

## OMNIGene®•GUT DNA and RNA (OMR-205) - Microbial RNA purification or DNA and RNA co-purification recommendations

Protocol using QIAGEN® RNeasy® PowerMicrobiome® Kit

This laboratory protocol is for the preparation of a fecal sample collected and stabilized in OMNIGene•GUT DNA and RNA (OMR-205) for subsequent purification of microbial RNA or co-purification of microbial DNA and RNA using the QIAGEN RNeasy PowerMicrobiome Kit.

### Required reagents

- QIAGEN RNeasy PowerMicrobiome Kit (Cat. No./ID 26000-50)
- $\beta$ -mercaptoethanol
- Phenol-chloroform-isoamyl alcohol (25:24:1, pH 6.5-8.0)

### Equipment required

- Pipettors and wide-bore 1000  $\mu$ L pipette tips (e.g., Axygen™ TF-1005-WB-R-S)
- Equipment listed as referenced in the RNeasy PowerMicrobiome Kit Handbook (HB-2276-003)

Procedure for RNA purification
1. Warm the PM1 solution at 55°C for 5–10 minutes prior to use.
2. Prepare DNase stock as described in the RNeasy PowerMicrobiome Kit Handbook.
3. Add 100 $\mu$ L phenol–chloroform–isoamyl alcohol to the PowerBead Tube.
4. Shake or vortex the OMNIGene•GUT DNA and RNA collected samples vigorously for 10 seconds.
5. Add 200 $\mu$ L of sample, 500 $\mu$ L of PM1 (pre-warmed) and 6.5 $\mu$ L of $\beta$ -mercaptoethanol to the PowerBead Tube.
6. Continue with the RNeasy PowerMicrobiome Kit, starting at step 3 of the manufacturer’s protocol, making sure to include the optional on-column DNase treatment (steps 11-13).

Procedure for DNA and RNA co-purification
1. Warm the PM1 solution at 55°C for 5–10 minutes prior to use.
2. Add 100 $\mu$ L phenol–chloroform–isoamyl alcohol to the PowerBead Tube.
3. Shake or vortex the OMNIGene•GUT DNA and RNA collected samples vigorously for 10 seconds.
4. Add 200 $\mu$ L of sample, 500 $\mu$ L of PM1 (pre-warmed) and 6.5 $\mu$ L of $\beta$ -mercaptoethanol to the PowerBead Tube.
5. Continue with the RNeasy PowerMicrobiome Kit, starting at step 3 of the manufacturer’s protocol, making sure to skip steps 11-13 to isolate total nucleic acids.
6. To isolate DNA or RNA from the total nucleic acid eluate, perform DNase treatment and/or RNase treatment (see recommendations below).

### Additional recommendations

- For RNA isolation from total nucleic acid eluate, we recommend using Lucigen<sup>®</sup> BaselineZERO™ kit to process >50 µL of eluate.
- DNase-treated samples can be purified with QIAGEN RNeasy<sup>®</sup> MinElute<sup>®</sup> Cleanup Kit, Zymo RNA Clean & Concentrator™-5 or equivalent.
- To isolate DNA from total nucleic acid eluate, we recommend treating >20 µL of the total nucleic acid eluate with RNase A as per the manufacturer's instructions.

**Technical support is available Monday to Friday (9h00 to 17h00 ET):**

- Toll-free (North America): 1.866.813.6354, option 6
- All other countries: 613.723.5757, option 6
- Email: [support@dnagenotek.com](mailto:support@dnagenotek.com)

OMNigene<sup>®</sup>·GUT DNA and RNA (OMR-205) is for Research Use Only, not for use in diagnostic procedures.

Some DNA Genotek products may not be available in all geographic regions.

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All DNA Genotek protocols, white papers and application notes are available in the support section of our website at [www.dnagenotek.com](http://www.dnagenotek.com)