User-Developed Protocol:

Isolation of mRNA from adipose tissue using Oligotex® Direct mRNA Kits

This procedure has been adapted by customers from the QIAGEN® Oligotex® Direct mRNA Protocols, and is for use with Oligotex Direct mRNA Kits. **It has not been thoroughly tested and optimized by QIAGEN.**

The procedure has been successfully used for isolation of mRNA from adipose tissue by customers. Because of the nature of adipose tissue it cannot be used immediately in Oligotex Direct mRNA Protocols; simple centrifugation, however, eliminates the problem, and the procedure works fine.

Please be sure to read the *Oligotex Handbook* and the detailed Oligotex Direct mRNA Protocol for Isolation of Poly A^+ mRNA from Animal Tissues carefully before beginning this procedure.

Procedure

1. **Homogenize tissue sample according to steps 1 and 2 of the Oligotex Direct mRNA Protocol for Isolation of Poly A^+ mRNA from Animal Tissues.**
2. **Centrifuge at 14,000 x *g* at 4°C for 10 minutes.**
   The sample will separate into three layers.
3. **Remove the top oily layer (~20–50 µl for 100 mg tissue).**
   Because the recommended sample size for the Oligotex Direct mRNA Micro and Mini preps is 100 mg, it may be difficult to see the three different layers.

   If the tube is held up to the light, the distinction between layers is more obvious. Alternatively, it is recommended to homogenize a large tissue sample and divide the lysate up into separate preps.
4. **Isolate the aqueous phase, and mix with 2 volumes of dilution Buffer ODB.**
5. **Follow the Oligotex Direct mRNA Protocol for Isolation of Poly A^+ mRNA from Animal Tissues from step 4.**

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Selected handbooks can be downloaded from **www.qiagen.com/literature/handbooks/default.asp**.
Material safety data sheets (MSDS) for any QIAGEN product can be downloaded from **www.qiagen.com/ts/msds.asp**.