ORAgene®•RNA

RNA recovery from saliva sponges

Once collected, Oragene®•RNA/saliva samples are stable at room temperature for at least 8 weeks without processing. Heating the samples as indicated below (step 1) ensures the RNA will be uniformly distributed in the sponges and the free liquid. If 5 sponges are used to collect saliva, about half of the liquid will be trapped in the sponges and the other half will be free.

To recover RNA from saliva sponge samples, we recommend the following methods:

- If you require RNA for a small number of tests proceed with method A.
- If you wish to recover RNA from the total sample for a battery of tests, proceed with method B.
- Alternatively, you can purify RNA from a sample with both method A and B. For instance, you can quickly run some preliminary tests by purifying a small aliquot (method A), store the sample at room temperature until all the samples have been collected for the study, and then proceed with method B to recover the remainder of the RNA. RNA recovered from the same donor using method A and B can be pooled.

Method A – purification of RNA from an 250 µL aliquot

1. Ensure that the cap of the Oragene•RNA vial is tight; mix gently by inversion 10 times. Incubate at 50°C for 1 hour in a water bath or for 2 hours in an air incubator.
2. Carefully open the vial and remove 250 µL of the free liquid.
3. Purify RNA according to the Oragene•RNA purification protocol for volumes up to 1,000 µL[1].

Method B – purification of Oragene•RNA from an 1 mL aliquot

1. Ensure that the cap of the Oragene•RNA vial is tight; mix gently by inversion 10 times. Incubate at 50°C for 1 hour in a water bath or for 2 hours in an air incubator.
2. Remove as much of the free liquid from the vial as possible and transfer to a 15 mL conical centrifuge tube.
3. Place the barrel of a 5 mL disposable plastic syringe (i.e., without the plunger) into the same 15 mL conical tube.
4. Using fine forceps, transfer the sponges from the vial base into the barrel of the syringe (see Figure 1).
5. Centrifuge the syringe barrel containing sponges in the conical tube at 200 × *g* (e.g., 1,000 rpm in a Sorvall RT6000D centrifuge) for 10 minutes at 20°C.
6. Remove and discard the syringe barrel containing the dry sponges.
7. RNA can be extracted from the saliva/Oragene•RNA liquid in the centrifuge tube, following instructions in the Oragene•RNA purification protocol for volumes up to 1,000 µL[1].

***Figure 1**: Syringe barrel with sponges inserted in conical tube for centrifuge.*

Notes

This protocol has been created as a suggested method for collecting RNA saliva samples from infants or young children who are unable to spit.

Reference

[1] Oragene®•RNA purification protocol using the Qiagen RNeasy Micro Kit for volumes up to 1,000 µL. DNA Genotek. PD-PR-021.

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

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

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