

DNA Recovery From Saliva Sponges

Once collected, Oragene®•DNA/saliva samples are stable at room temperature for years without processing. Heating the samples as indicated below (step 1) ensures the DNA will be uniformly distributed in the sponges and the free liquid. If 5 sponges are used to collect saliva, about ½ of the liquid will be trapped in the sponges and the other ½ will be free.

To recover DNA from saliva sponge samples, we recommend the following methods. If you require DNA for a small number of tests or want a fast and accurate estimate of the total amount of DNA collected, proceed with **method A**. If you wish to recover DNA from the total sample for a battery of tests, proceed with **method B**. Alternatively, one can purify DNA from a sample with both method A and B. For instance, one can quickly estimate the total amount of DNA in a sample and 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 DNA. DNA recovered from the same donor using method A and B can be pooled.

Method A) Purification of DNA from an 0.5 mL aliquot

1. Ensure that the cap of the Oragene•DNA vial is tight. Mix gently by inversion 5 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 0.5 mL of the free liquid.
3. Purify DNA according to the *Laboratory Protocol for Manual Purification of DNA from 0.5 mL of Oragene•DNA/saliva*. Inclusion of glycogen, described as an optional step in the Protocol, will make the DNA pellet more visible and may improve yield.

Method B) Purification of DNA from the total sample

1. Ensure that the cap of the Oragene•DNA vial is tight. Mix gently by inversion 5 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 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 blue base into the barrel of the syringe (see Figure).
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 min at 20°C.
6. Remove and discard the syringe barrel containing the dry sponges.
7. DNA can be extracted from the saliva/Oragene•DNA liquid in the centrifuge tube, following instructions in the *Laboratory Protocol for Manual Purification of DNA from 4.0 mL of Oragene•DNA/saliva*. Ensure the volumes of reagents used in this protocol for a 4 mL sample are adjusted for the actual volume of saliva/Oragene•DNA liquid recovered from samples collected with saliva sponges.



Other Notes

1. This protocol has been created as a suggested method for collecting DNA saliva samples from infants or young children who are unable to spit. If you have any recommendations for improvements, please contact Adele.Jackson@dnagenotek.com.

References

1. Oragene®•DNA Laboratory Protocol for Manual Purification of DNA from 0.5 mL of Oragene®•DNA/saliva (2006). *DNA Genotek*. http://www.dnagenotek.com/techsupport_documents.htm
1. Oragene®•DNA Laboratory Protocol for Manual Purification of DNA from 4.0 mL of Oragene®•DNA/saliva (2006). *DNA Genotek*. http://www.dnagenotek.com/techsupport_documents.htm
2. Chartier, J., Egan, Birnboim, H.C. (2004) DNA Yield is Proportional to Saliva Volume. *DNA Genotek* http://www.dnagenotek.com/techsupport_documents.htm.

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