

Purification Protocol for 4 ml of Oragene®•DNA Saliva Using the chemagic Magnetic Separation Module I and the chemagic Dispenser

Protocol name: chemagic DNA Saliva 4k prefilling V081124.che

Positioning Racks on the Tracking System

Can be done manually or by an integrated robotic system

Position 1: Rack with disposable tips

Position 2: 50 ml tubes containing 4 ml Oragene saliva

See "Before You Start" and "Preparing Steps"

Position 3: 50 ml tubes prefilled with 400 µl Magnetic Beads (for 5 ml Wash Buffer 3)

Position 4: Empty 50 ml tubes (for 5 ml Wash Buffer 4)

Position 5: Empty 50 ml tubes (for 5 ml Wash Buffer 5)

Position 6: Empty 50 ml tubes (for 5 ml Wash Buffer 5)

Position 7: Empty 50 ml tubes (for 5 ml Wash Buffer 6)

Position 8: 4 ml tubes prefilled with 400 µl Elution Buffer 7

Before You Start

- Collect the saliva sample as described in Oragene•DNA user instructions provided with each kit
- Incubate the Oragene•DNA saliva sample over night at 50 °C
- Transfer 4 ml of the Oragene•DNA saliva into a 50 ml tube

Preparing Steps

- Fill 400 µl Elution Buffer 7 into 4 ml tubes.
- Fill 400 µl Magnetic Beads into 50 ml tubes (before transfer resuspend the beads carefully).
- Select the protocol "chemagic DNA Saliva 4k drying prefilling ext wash V090526.che" and press the [Insert IDs] button.
- If the enhanced functions are deactivated continue without pressing the [Insert IDs] button.
- Check the volumes in the buffer supply containers (see above) and confirm by pressing the [OK] button.
- Select the number of samples by using the pull-down-menu.
- The scheme for positioning the tubes will be shown after selecting.
- If the enhanced functions are activated follow the instructions as given in the software to enter the IDs for samples, eluates and racks.
- Position all racks with the required number of tubes on the tracking system according to the instructions.
- Check all racks and tubes for correct fitting on the tracking system.
- After closing the cover start the automated isolation process by pressing the [OK] button (enhanced functions activated) or the [Start] button (without enhanced functions); the lysate will be mixed for 10 minutes. Afterwards the other buffers will be prefilled by the chemagic Dispenser. The isolation of the nucleic acids will be performed by the chemagic MSM I without further manual manipulation.

Clean the chemagic Dispenser (at least once a week; see manual). Take care to drain the waste container frequently.

General Remarks

The Elution Buffer 7 included in this kit is 10 mM.

Tris-HCl pH 8.0. TE buffer pH 8.0 can also be used without any protocol adjustments. Water pH 8.0 may also be used, but the yield could be slightly decreased.

Binding Buffer 2, Wash Buffer 3, Wash Buffer 4, Wash Buffer 5 contain ethanol. Longer storage of the buffers without lids should be avoided.

If ethanol evaporates the optimal yield can not be guaranteed.

The Magnetic Bead suspension should be mixed vigorously before dispensing, otherwise the suspension is not homogenous and the DNA yield could be low.

UV Measurements/Real Time PCR

In some cases you may find some traces of magnetic beads left in the eluate. Such particles will not interfere with standard PCR and most

downstream applications but may increase the background in UV measurements or could influence real time PCR. In such a case we recommend to perform an additional separation step using the chemagic Stand 2x12 (art. No. 300) in order to separate traces of particles.

For questions regarding this protocol, call Technical Support at chemagen +49 (0) 2401 805-501.