

# **XpressDNA Milk kit**

## Protocol for extraction of milk DNA using XpressDNA Milk kit

#### **Kit Contents**

Components	<b>Storage Conditions</b>	<b>Shipping Conditions</b>
Milk Lysis Buffer	RT	RT
Proteinase K	2 - 8 °C	RT
Proteinase K Buffer	2 - 8 °C	RT
Milk MagNa Mix	RT	RT
Milk Wash Buffer 1	RT	RT
Milk Wash Buffer 2	RT	RT
Milk Elution Buffer	RT	RT
MagNa Stand (optional)	RT	RT

<sup>\*</sup> RT denotes 15 - 25°C.

Note: Materials not provided with the kit

- 0.5M EDTA
- TE Buffer
- RNase A solution

#### **Important**

Pay attention to standard lab practices and safety information before beginning the procedure. For more information, refer the appropriate Material Safety Data Sheet (MSDS) available from the product supplier or download from our website <a href="http://www.maggenome.com/">http://www.maggenome.com/</a>

#### **Technical Support**

For any product related queries please write to us on <a href="mailto:info@maggenome.com">info@maggenome.com</a>, <a href="maggenome.com">sales@maggenome.com</a>, <a href="maggenome.com">sales@maggenome.com</a>,

### **Pretreatment**

- 1. To a sterile microcentrifuge tube, add **1ml of milk** sample
- 2. Centrifuge at 10000 rpm for 5 minutes.
- 3. Carefully discard the supernatant by pipetting and add 100µl of 0.5M EDTA and 100µl of TE buffer.
- 4. Vortex the tube for 20 seconds and Centrifuge at 14500 rpm for 20 seconds.
- 5. Carefully discard the supernatant by pipetting.

### **Preparation of lysate**

- 6. To the pellet add 500µl of Milk lysis Buffer (completely resuspend the pelletby pipette mixing).
- 7. Add **10ul of RNase A**, mix and keep at RT for 15 minutes.
- 8. Add 20 µl of Proteinase K and mix the contents by pipetting.
- 9. Incubate at **56°C** for 1 hour
- 10. Centrifuge at 14,000 rpm for 5 minutes at room temperature.
- 11. Transfer the supernatant to a fresh microcentrifuge tube.

## **Binding and Washing DNA**

- 12. To the lysate, add **350μl of Milk MagNa Mix.** Gently mix the contents by inverting the tube 10-12 times. Incubate at room temperature for 5 minutes
- 13. Place the tube on MagNa Stand for 5 minutes.
- 14. Carefully discard the supernatant keeping the tube on MagNa Stand (*Make sure that the pellet is not disturbed*).
- 15. Add **500μl of Milk Wash Buffer-1**, remove the tube from MagNa Stand and resuspend the pellets by pipette mixing for about 5-8 times (*ensure complete dispersion of the particles*).
- 16. Place the tube back on MagNa Stand until the solution becomes clear (30 sec-1 min.).
- 17. Carefully discard the supernatant with the tube on MagNa Stand (*Make sure that the pellet is not disturbed*).
- 18. Add **500μl of Milk Wash Buffer-2** and gently invert the tube placed on MagNa Stand for 5-6 times to wash the pellet (*surface wash only*)
- 19. Discard the supernatant without disturbing the pellet.
- 20. Repeat step 17-18.
- 21. Air dry the pellet with the tube on MagNa Stand at RT for 10-15 minutes.

**NOTE**: Do not over dry the pellet.

## Elute DNA

- 1. After drying, remove the tube from MagNa Stand.
- 2. Add **50μl of Elution Buffer** and resuspend thepellet by pipette mixing 10-12 times (*ensure complete dispersion of particles*).
- 3. Incubate at **56°C for 5 minutes.**

- 4. Place the tube on MagNa Stand for 5 minutes or until the solution gets cleared.
- 5. Carefully transfer the supernatant containing DNA to a sterile microcentrifuge tube, with the tube on MagNa Stand. Make sure that the pellet is not disturbed.
- 6. Discard the MagNa particles.

**Note:** In the elution step, if the MagNa particles take more than 10 minutes for clearing, then spin the tubes at 14,000 rpm for 5 minutes and collect the supernatant for pure DNA.