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Ref. SB0101

MANUAL

Expiry date: 1 year
Store at 4^0C

GENEKAM RNA ISOLATION KIT (MAGNETIC BEADS) **-Only for research use-**
-To be used by a technical person-

Contents:

- Tube A (Lysis buffer)
- Tube K (Proteinase K)
- Tube B (Washing buffer 1)
- Tube M (Magnetic beads)
- Tube E (Elution buffer)

Chemicals and equipments needed:

- Pipettes and Pipette tips
- Heat block
- Centrifuge
- Magnetic rack (check whether microtubes fit in them properly)
- RNase free tube
- Microtubes (1.5 or 2ml)

Procedure:

Standard Step (this can be used with any sample):

1. Add 100-250µl plasma / serum / cell culture fluid / vaccine / body fluid and 600µl of tube A together in one tube. Add 15µl of tube M (magnetic beads) to it.
2. To it add 20µl of tube K.
3. Incubate at room temperature for 8 minutes.
4. Separate the beads with magnetic rack. Discard the supernant through removing with pipettor (Once again, use pipette to remove supernant!).
5. Add 500µl of tube B (add 10 µl of tube K; it should be freshly prepared therefore add Tube K freshly) to resuspend the beads. Keep at room temperature for 10 minutes. Separate beads with magnet. Discard the supernant with pipettor.
6. Add 500µl of tube B (add 10µl of Tube K to it. It should be freshly prepared, therefore Tube K should added before use) to resuspend the beads. Keep at room temperature for 1 minute. Separate the beads with magnet and discard the supernant with pipettor.
7. Add 100µl of tube E and resuspend it. Incubate it at 80° C for 10 minutes (during heating, remix it or reshake it). Separate the beads with magnet und store the supernant containing RNA in RNAase free tube.

Hint: 1. How to isolate RNA from blood samples: to 50µl of blood, add 760µl of tube A and 15µl of tube M (magnetic beads). To these, add 20µl of tube K. Now keep it at room temperature for 10 minutes and proceed ahead with number 4 of standard step onwards.

2. How to isolate RNA from buccal swabs: add 300 µl of tube A to buccal swab. Add to it 25µl of tube K. Keep it at 56 ^{0}C for 15 minutes. Remove the buccal swabs (very important!) and add 300µl of tube A to it. Now proceed with step 2 of standard step (i.e. adding 15µl of beads) till elution.

3. How to isolate RNA from cell pellet: add cell pellet to 600µl of tube A. After that add 15µl of tube M. To them add 20µl of tube K. Now proceed with step 3 of standard step i.e. keep it at room temperature till elution.

If you should find any mistakes, please let us know. Thank you.

Suggestion: **This manual has been written specifically for beginners, hence persons with experience in PCR must use their experience to keep each step as small as possible e.g. you should calculate the amount of the needed chemicals, before starting with testing.** **Last update: 21-11-2018** **v1.2**	**Genekam Biotechnology AG** **Duissernstr. 65a** **47058 Duisburg** **Germany** **Tel. (+49) 203 / 555858-31,-32,-33** **Fax (+49) 203 / 35 82 99** **anfrage@genekam.de** **http://www.genekam.de**