



M6K3S46

(For Research Use Only) V3

### 1. Intended Use

TANBead® Nucleic Acid Extraction Kit (M6K3S46) is suitable for isolating RNA from a wide range of plant species. Pretreated samples can be processed through a series of extraction steps, which is operated by the magnetic bead-based technology of TANBead® Nucleic Acid Extractor (Maelstrom 8 series, Maelstrom 4800 series). With the features of high quality and quantity, the purified extracts can be applied for downstream assays including real-time PCR and next generation sequencing.

#### 2. Purpose

TANBead® Nucleic Acid Extraction Kit (M6K3S46) is employed in RNA isolation from a variety of plant samples. After pretreatment and transferring of the sample, with automated nucleic acids extractor (Maelstrom 8 series & Maelstrom 4800 series) your precious time will be saved, and the isolation of RNA will be remarkably consistent. The isolated nucleic acids samples can be used in subsequent applications, such as real time PCR and sequencing. It is suitable for laboratories with high throughput requirement.

### 3. The basic principle

The silicon dioxide layer coated on the magnetic beads can adsorb the negatively charged molecules to purify nucleic acids from samples.

### 4. Specification

| Starting Materials | 30 – 50 mg plant sample |
|--------------------|-------------------------|
| Elution Volume     | 70 – 100 μL             |
| Typical RNA yield  | Up to 5 µg              |

# 5. Component Supplied with the Kit Auto Tube 8 Trays Auto Tube with reagent buffers Lysis Buffer 90 mL x 1 Sodium salt, Tris buffer, surfact

| Lysis Buffer      | 90 mL x 1         | Sodium salt, Tris buffer, surfactants |
|-------------------|-------------------|---------------------------------------|
| Elution<br>Buffer | 1.5 mL x 1        | Nuclease-Free Water                   |
| Base              | 2                 | A rack for 8 Auto Tubes               |
| Spin tips         | 48 tips x 2 boxes | Spin tip assembled box                |
| Protocol          | 1                 | Instruction guide for user            |

# 6. Auto Tube Content

| Well  | Buffer           | Volume (μL) |
|-------|------------------|-------------|
| 1/ 7  | -                | -           |
| 2/ 8  | Washing Buffer 1 | 800         |
| 3/ 9  | Magnetic Beads   | 800         |
| 4/ 10 | Washing Buffer 2 | 800         |
| 5/ 11 | Washing Buffer 2 | 800         |
| 6/ 12 | Elution Buffer   | 100         |

### 7. Kit Storage and Shelf Life

1) Components under room temperature (15~35°C) can be stored until the expiration date labeled on the box.

# 8. Precautions

- 1) For research use only.
- 2) Avoid using expired reagents.
- 3) When the temperature is below 20°C, place the Auto Tubes in an oven (preheated 42~60°C) 5 to 10 minutes.
- Avoid vigorous shaking, in order to avoid excessive formation of foam.
- 5) Carefully remove aluminum foil to avoid splashing.
- 6) Do not expose the opened reagents or Auto Tubes to air. The evaporation would lead to pH change, or effect on the extraction effectiveness.
- Please check the integrity of the Auto Tubes and remember to mount the spin tips into the appropriate position of the suitable instrument before operating them.
- 8) Please wear a mask and disposable gloves when handling.
- 9) Use sterile consumables to avoid nuclease contamination.

- Reagent solution contains guanidine salt, avoid using bleach containing detergent.
- Avoid eyes, skin, and clothing contact with reagents. In case of any contact, flush with flowing water.
- 12) If any serious incident occurs, please report to the manufacturer and the competent authority of the member state in which the user and/or the patient is established.
- 13) The Washing Buffer 2 and Elution Buffer are colorless and transparent. Colored reagent may be due to the contamination, please replace it with a new one before use.

### 9. Materials required, Not Supplied

- TANBead<sup>®</sup> Nucleic Acid Extraction System Model: Maelstrom 8 series, Maelstrom 4800 series (non-sterile)
- 2) Disposable gloves
- 3) Scissors, utility knives
- 4) Micropipette, disposable tips (10 μL / 200 μL / 1000 μL)
- 5) 1.5 mL microcentrifuge tube
- 6) 15 mL / 50 mL conical tube
- 7) IPA: Isopropanol for molecular biology
- 8) Liquid nitrogen

### 10. Sample Collection, Transportation, and Storage

### ■ Sample collection and storage

Freshly collected plant sample is suggested for use or stored it at -20 or -80°C for long-term conservation.

### Sample transportation

Transportation of plant samples should follow specific plant transportation related law. Plant samples should be kept between 2 - 8°C during transportation.

# 11. Nucleic Acids Extraction Protocol Sample preparation

- 1) Add liquid nitrogen to the sample and grind it.
- 2) Collect the sample and add 800 µL Lysis Buffer.
- 3) Mix well and stand for 10 minutes on ice.
- 4) Centrifuge at 6000 RPM for 5 minutes and take 500 uL supernatant as the sample for the following step.

# **Automatic process**

- Prepare the Assembled Auto Tubes by inserting Auto Tubes into the Base completely.
- 2) Carefully remove the aluminum foil from Auto Tube.
- 3) Transfer 500 µL supernatant into wells of column #1 / #7 of Auto
- Add 500 μL IPA into wells of column #1 / #7.
- 5) Set up spin tips:

**Maelstrom 8 series**: Handle to mount tips and make sure that there is no gap between the necks of spin tips and the spin shaft. **Maelstrom 4800 series**: Go to Tip page and mount tips region by region.

- 6) Push Assembled Auto Tubes completely to the bottom of the plate rack. Make sure that the chamfer of the reagent plate is at the lower left.
- 7) Close the door panel.
- 8) Select the program:

Maelstrom 8 series: Press "6K3-1" or "6K3-7" for input samples at wells of column #1 or column #7, respectively.

Maelstrom 4800 series: Press "6K3".

The parameters are given in following section.

- Once the program has ended, buzzer shall alarm. Take out Auto Plate carefully.
- Use micropipette to transfer the purified nucleic acids from well of column #6/ #12 to a clean tube.
- 11) Discard the used Auto Tube and spin tips into the waste recycling bin.

### 12. Program

### ■ Maelstrom 8 series

| Program Name: 6K3 |          |          |          |          |          |          |
|-------------------|----------|----------|----------|----------|----------|----------|
| Well              | 1/7      | 2/8      | 3/9      | 4 / 10   | 5 / 11   | 6 / 12   |
| Volume            | 800 (µl) | 150 (µl) |

| Step | Well  | Action     | RPM  | Time<br>(Second) | CW/CCW<br>(Second) | Temp. | Temp.<br>Control |
|------|-------|------------|------|------------------|--------------------|-------|------------------|
| 1    | 3/9   | Mixing     | 3000 | 10               | 0                  | 55    | YES              |
| 2    | 3/9   | Collection | 0    | 30               | 0                  | 55    | YES              |
| 3    | 2/8   | Mixing     | 3000 | 30               | 0                  | 55    | YES              |
| 4    | 2/8   | Collection | 0    | 30               | 0                  | 55    | YES              |
| 5    | 1/7   | Mixing     | 3000 | 600              | 0                  | 55    | YES              |
| 6    | 1/7   | Collection | 0    | 30               | 0                  | 55    | YES              |
| 7    | 2/8   | Mixing     | 3000 | 120              | 0                  | 45    | YES              |
| 8    | 2/8   | Collection | 0    | 30               | 0                  | 45    | YES              |
| 9    | 3/9   | Mixing     | 3000 | 120              | 0                  | 45    | YES              |
| 10   | 3/9   | Collection | 0    | 30               | 0                  | 45    | YES              |
| 11   | 4/ 10 | Mixing     | 3000 | 120              | 0                  | 45    | YES              |
| 12   | 4/ 10 | Collection | 0    | 30               | 0                  | 45    | YES              |
| 13   | 5/ 11 | Mixing     | 3000 | 120              | 0                  | 45    | YES              |
| 14   | 5/ 11 | Collection | 0    | 30               | 0                  | 45    | YES              |
| 15   | 5/ 11 | Vapor      | 0    | 600              | 0                  | 45    | YES              |
| 16   | 6/ 12 | Mixing     | 3000 | 600              | 0                  | 45    | YES              |
| 17   | 6/ 12 | Collection | 0    | 60               | 0                  | 45    | YES              |
| 18   | 5/ 11 | Mixing     | 3000 | 30               | 0                  | 0     | NO               |

### ■ Maelstrom 4800 series

| Program Name: 6K3 |       |              | Model: Maelstrom 4800 series |                          |                |              |       |
|-------------------|-------|--------------|------------------------------|--------------------------|----------------|--------------|-------|
| Temp1             | Temp2 |              |                              |                          |                |              |       |
| 45                | 40    |              |                              |                          |                |              |       |
| Well              | Name  | Volume       | Action                       | Mixing                   | Collect        |              |       |
| 1/7               | LB    | 800          | For.                         | Low                      | Low            |              |       |
| 2/8               | WB1   | 800          | For                          | Low                      | Low            |              |       |
| 3/9               | МВ    | 800          | For                          | Low                      | Low            |              |       |
| 4 / 10            | WB2   | 800          | For                          | Low                      | Low            |              |       |
| 5 / 11            | WB2   | 800          | For                          | Low                      | Low            |              |       |
| 6 / 12            | EB    | 100          | For                          | Low                      | Low            |              |       |
| Step              | Well  | Temp<br>(°C) | Mixing<br>(M)                | Mixing<br>Speed<br>(RPM) | Collect<br>(M) | Vapor<br>(M) | Pause |
| 1                 | 3/9   |              | 0.2                          | 3000                     | 0.5            | 0            | Off   |
| 2                 | 2/8   |              | 0.5                          | 2500                     | 0.5            | 0            | Off   |
| 3                 | 1/7   | 55           | 10                           | 2500                     | 0.5            | 0            | Off   |
| 4                 | 2/8   |              | 2                            | 2500                     | 0.5            | 0            | Off   |
| 5                 | 3/9   |              | 1                            | 1500                     | 0.5            | 0            | Off   |
| 6                 | 4/ 10 |              | 1                            | 1500                     | 0.5            | 0            | Off   |
| 7                 | 5/ 11 |              | 1                            | 1500                     | 0.5            | 10           | Off   |
| 8                 | 6/ 12 | 45           | 5                            | 2500                     | 0.5            | 0            | Off   |
| 9                 | 3/9   |              | 0.2                          | 3000                     | 0              | 0            | Off   |

### 13. Result

Nucleic acid product purified by TANBead® nucleic acid extraction kit can perform qualitative / quantitative analysis of specific genes by PCR, RT-PCR, Q-PCR or qRT-PCR. Please refer to the molecular diagnostic kit manual.

# 14. Reagent performance

### ■ Repeatability

Under repeatability conditions where nucleic acids are extracted with the same reagent kit on the same source samples by the same operator. The coefficient of variation of nucleic acids extraction concentration is less than 5%.

### ■ Reproducibility

A five-day reproducibility test was carried out with the same source samples for 5 consecutive days with the same reagent kit by different operators. The coefficient of variation of nucleic acids extraction concentration is less than 5%.

# ■ The stability of extracted RNA

| Storage Conditions | RNA stability |
|--------------------|---------------|
| -80°C              | Over 90 days  |
| -20°C              | 28 days       |
| 4°C                | 14 days       |
| 25°C               | 2 days        |
| Freeze-thaw        | 5 times       |

### 15. Explanation of Symbols

|           | Manufacturer          | (i          | Consult instructions for use |
|-----------|-----------------------|-------------|------------------------------|
| 15°C 35°C | Temperature limit     | Σ           | Contains sufficient for test |
| REF       | Catalogue number      | $\triangle$ | Caution                      |
| LOT       | Batch code            | NOM         | Non-sterile                  |
| 8         | Do not re-use         | 誉           | Keep away from<br>sunlight   |
| سا        | Date of manufacture   | 8           | Use-by date                  |
| RUO       | For research use only |             |                              |