TANBead® Nucleic Acid Extraction Kit

Plant RNA Auto Plate

(For use with the Maelstrom 8 series and Maelstrom 4800 series)

RUO

M6K3A46

(For Research Use Only) V5

1. Intended Use

TANBead® Nucleic Acid Extraction Kit (M6K3A46) 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 (M6K3A46) 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 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 Plate	6	Auto Plate with reagent buffers
Lysis Buffer	90 mL x 1	Sodium salt, Tris buffer, surfactants
Elution Buffer	1.5 mL x 1	Nuclease-Free Water
Spin tips	96 tips	Spin tip assembled box
Protocol	1	Instruction guide for user

6. Auto Plate Content

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

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 Plates in an oven (preheated 42~60°C) for 5 to 10 minutes.
4) 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 Plates to air. The evaporation would lead to pH change or effect on the extraction effectiveness.
7) Please check the integrity of the Auto Plates 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.
10) Reagent solution contains guanidine salt, avoid using bleach-containing detergent.
11) 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

1) TANBead® 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.

■ **Specimen 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**

1) Carefully remove the aluminum foil on Auto Plate.
2) Transfer **500 µL supernatant** into wells of column **#1 / #7** of Auto Plate.
3) Add **500 µL IPA** into wells of column **#1 / #7.**
4) 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 press the mount tips region.
5) Push Auto Plate completely to the bottom of the plate rack. Make sure that the chamfer of the plate is at the lower left.
6) Close the door panel.
7) 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.
8) Once the program has ended, buzzer shall alarm. Take out Auto Plate carefully.
9) Use a micropipette to transfer the purified nucleic acids from well of column **#6 / #12** to a clean tube.
10) Discard used Auto Plates and spin tips into the waste recycling bin.

12. Program

■ **Maelstrom 8 series**

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

Step	Well	Action	RPM	Time (Second)	CW/CCW (Second)	Temp.	Temp. 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	MB	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 (°C)**	**Mixing (M)**	**Mixing Speed (RPM)**	**Collect (M)**	**Vapor (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		**Consult instructions for use**
15°C 25°C	**Temperature limit**		**Contains sufficient for test**
REF	**Catalogue number**		**Caution**
LOT	**Batch code**	NON STERILE	**Non-sterile**
	Do not re-use		**Keep away from sunlight**
	Date of manufacture		**Use-by date**
RUO	**For research use only**		