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<b>SOPs For Automatic Nucleic Acid Extraction</b>	Reviewed By	
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### **1: SCOPE:**

This SOP describes the method of extracting NUCLEIC ACID from respiratory tract samples such as throat swab and nasopharyngeal by using ZYBIO (MEGANTIC bead method) nucleic acid extraction kit.

### **2: PURPOSE:**

The purpose of this Standard operating procedure (SOP) is to established an outline procedure for extracting Nucleic Acid from various samples in Covid-19 testing Laboratory.

### **3: AIM:**

The aim of this SOP is to extract high quality RNA from various samples for diagnostic and research purpose.

### **4: Responsibility:**

**4.1:** Laboratory supervisors are responsible for making sure that technical staff (Technician, technologist and Molecular Biologist) for extraction unit are properly trained and equipment and facility are maintained in good working order.

#### **Training must be given on the following topics:**

- Wearing and use of personal protective equipment and clothing;
- Handling of potentially infectious materials;
- Prevention of incidents and steps to be taken by workers in case incidents (including biological, chemical, electrical and fire hazards) occur;
- Procedures;
- Waste management;
- Impact of results for patient management and research.

#### **Training must be provided:**

- When a new staff member takes up post;
- Annually;

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- When there is a change in conditions or best practices

**4.2:** Laboratory personnel are responsible for reading and understanding the SOP and related documents and to perform these tasks in accordance with the SOPs.

## 5: Abbreviations

RNA Ribonucleic ACID

SOP Standard Operating Procedure

µL microlitre

## 6: Material and Methods:

The material and equipment and forms and listed in the following list are recommended only and alternative products are suitable may be substituted for the site-specific task or procedure.

### Materials and Equipment supplied:

1. Zybio-3000 ExtractionSystem
2. Zybio extractor Plates
3. Lysis buffer (20 ul)
4. Disposable strip caps
5. Instruction for use

### Material required but not included:

1. Micro pipettes single channel ( 5ul-200 ul)
2. Sterile filter tips for micro pipettes
3. Disposable gloves
4. Biohazard waste containers.

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**Procedure:**

**Principle:**

The ZYbio nucleic Acid kit is designed to extract RNA from different respiratory samples. Cells are lysed and RNA released in a protease-containing buffer. The released RNA is bound to a MEGANTIC BEADS and purified using two washes that remove contaminants. The purified RNA is released from the column using distilled water or buffer.

This method makes it possible to obtain RNA suitable for PCR. It is a fast, high quality, and effective method of RNA extraction. As this is the first step in the PCR process, it is important to maintain sterile technique to prevent contamination of extracted RNA.

**Sample type:**

serum, plasma, nasopharyngeal swab, cell preservation solution, tissue fluid, urine and s

**Sample volume**

up to 100 /200 uL of samples secretions.

**General Protocol :**

**Step 1:** Take out all the components in the kit, keep them at room temperature to be ready for use. Dump the liquid that adheres to the aluminum film and the well wall of the 96-well plates to the bottom of the plates. Let them stand for 3-5 minutes.

**Step 2:** Carefully open the aluminum film of the 96-well plates, and add 15µL [Proteinase K] (B-100: 10µL) to the position A1~H1 and A7~H7 in order, then add 200 µL sample (B-100: 100µL) in order.

**Step 3:** Turn on the nucleic acid Extraction system, enter the page < Program Edit >, and set the extraction process according to table 4:

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### 1. Table 4 Running Program Setting

No.	Position	Name	Waiting Time (min)	Mixing Time (min)	Absorption Magnetic Bead Time (sec)	Mixture Velocity	Volume	Temperature State	Temperature(°C)
1	2	Move	0	0	30	Slow	150	Closed	0
2	1	Lysis	0	4	60	Slow	500	Heating for Lysis	55
3	3	Wash	0	1	60	Slow	600	Closed	0
4	6	Elution	0	2	30	Slow	50	Heating for Elution	80
5	1	Move	0	0	0	Slow	300	Closed	0

**Step 4:** Click “Start” to run the extraction program. The process takes about 10 minutes.

**Step 5:** Take out 96-well plates and pipette inventory nucleic acid solution from the position A6-H6 and A12-H12 into 1.5 mL centrifuge tube for following operation. (a small amount magnetic beads could be removed by centrifuge or magnetic separator.)