

#### **INTENDED USE**

AZUL cf-DNA Extraction Kit is an easy and efficient system for the isolation of cf-DNA from plasma and serum samples.

### **SUMMARY AND EXPLANATION**

This kit uses a spin column based extraction for isolating cf-DNA from plasma and serum samples. The eluted cf-DNA is suitable for all sensitive downstream applications such as qPCR and Next-Generation sequencing.

#### **PRODUCT FEATURES**

- Rapid purification of high-quality, ready-to-use cf-DNA.
- No organic extraction or alcohol precipitation.
- · Consistent and high yields.
- Complete removal of contaminants and inhibitors for reliable results.
- Kit formats for low- to high-throughput options for automation of all kits.

#### **PRECAUTIONS**

- 1.AZUL cf-DNA Extraction kits are intended for use as supplied. Do not dilute or add other components to the AZUL cf-DNA Extraction kit.
- 2. Dispose of used reagents, debris, and consumables as hazardous waste according to established laboratory procedures.

#### **DIRECTIONS FOR USE**

## Plasma separation from Blood:

- Take the whole blood samples and centrifuge at 5,400 rpm for 10 mins, and transfer the plasma into a new microfuge tube.
- Centrifuge the separated plasma at 15,000 rpm for 20 mins to separate any remaining cells or particulate matter & transfer it into a new microfuge tube.
- The plasma samples can be stored at  $-20^{\circ}\text{C}$  until further use.

#### cf-DNA Extraction:

- 1.Add 50  $\mu$ L of Lysis Buffer (LB) to 500  $\mu$ L of Plasma and vortex the mixture for 15 sec.
- 2.Add 20  $\mu L$  of Proteinase K to the sample and mix well by inverting the tube 4-5 times.
- 3.Incubate the mixture for 1 hour at 56°C.
- 4.Add 750  $\mu L$  of Binding Buffer (BB) to the mixture. Mix by inverting the tube a few times.
- 5.Transfer the lysate to a spin column and centrifuge the tube at 2000xg for 30 sec. Without removing the spin column, centrifuge again at the speed of 11,000xg for 5 sec.
- 6.Discard the flow-through and place the purification column back into the collection tube. Repeat this step until complete lysate has been transferred into the column and centrifuged.
- 7. Wash the spin column with 500  $\mu$ L Wash Buffer 1 (WB1) at 11,000xg for 30 sec and discard the flow through.
- 8.Wash the spin column with 250  $\mu L$  Wash Buffer 2 (WB2) at 11,000xg for 2 mins and discard the flow through.
- 9. Centrifuge the spin column again for 1 min at 11,000xg to remove any excess wash buffer.
- 10. Keep the purification column in a clean, sterile 1.5 mL microfuge tube and add 20  $\mu$ L- 30  $\mu$ L of Elution Buffer or DNase/RNase-free water to the center of the column.
- 11. Centrifuge the column for 11,000xg for 30 sec.
- 12. Discard the purification column and store the eluted cf-DNA at -20°C or -80°C until use.

## KIT COMPONENTS

Components	For 50 preps	For 25 preps
Lysis Buffer (LB)	4 mL	2 mL
Binding Buffer (BB)	40 mL	20 mL
Proteinase K	1.5 mL	1 mL
Wash Buffer I (WBI)	25 mL	15 mL
Wash Buffer 2 (WB2)	15 mL	8 mL
Elution Buffer(EB)	4 mL	2 mL
Spin Column	50 (Pouch Pack)	25 (Pouch Pack)

#### **CAUTION**

- Check the Binding Buffer and Lysis Buffer for any salt precipitation before every use.
- Re-dissolve any precipitate by warming the solution to 37°C, then cool it back to room temperature before use.
- During operation, always wear a lab coat, disposable gloves, protective goggles and mask.

#### KIT STORAGE AND STABILITY

- Store the kit at room temperature and Proteinase K at -20°C.
- Viable for 1 year if stored at appropriate conditions.

# **ORDERING INFORMATION**

Please call us at +91 8088747968 or mail at hello@azooka.life for any queries or assistance.

Additional information can be found online at www.azooka.life

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