



**Technical Report**

**A test of the performance of cfDNA extraction by using Maelstrom 9600**

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**1. Purpose**

Liquid Biopsy has been widely used in clinical testing in recent years. Blood is a specimen that is relatively easy to obtain clinically. Therefore, if disease information can be obtained from blood, it is bound to be faster in future medical diagnosis. In this experiment, cfDNA will be extracted from human serum using TANBead Maelstrom 9600 automated nucleic acid extractor and TANBead cfDNA Extraction Kit, which will be compared in parallel with Applied Biosystems-MagMax Cell-Free Isolation Kit.

**2. Materials**

2.1 Instruments

- Maelstrom 9600 (TANBead®)
- Qubit 4 Fluorometer (Invitrogen™)
- Qsep 100 (BiOptic Inc.)

2.2 Sample

- Human Serum, Pooled, 500 mL, Frozen (zenbio, HSER-P500ML)  
Lot: SER092519E-BL

**3. Method**

3.1 Reagent plate format

| Plate       | 1   | 2   | 3   | 4   | 5  | 6   | 8   |
|-------------|-----|-----|-----|-----|----|-----|-----|
| Buffer      | LB  | LB  | WB  | WB  | MB | EB  |     |
| Volume (μL) | 800 | 800 | 500 | 500 | 60 | 440 | 100 |

- 500 μL pre-treated serum was added to plate 1, 2 and add 20 μL Proteinase K respectively.

3.2 Serum pretreatment

- Place the frozen serum at 4°C until it is completely thawed.
- Transfer 1.0 mL thawed serum into 1.5 mL centrifuge tube.
- Centrifuge the serum at 16000 rcf for 10 minutes at 4°C.
- Carefully transfer the supernatant into a new centrifuge tube.

3.3 Applied Biosystems - MagMax Cell-Free DNA Isolation Kit

- Manually operate according to kit's manual and adjust Elution steps to 100 μL ◦

3.4 Use Qubit 4 Fluorometer to quantify the amount of dsDNA

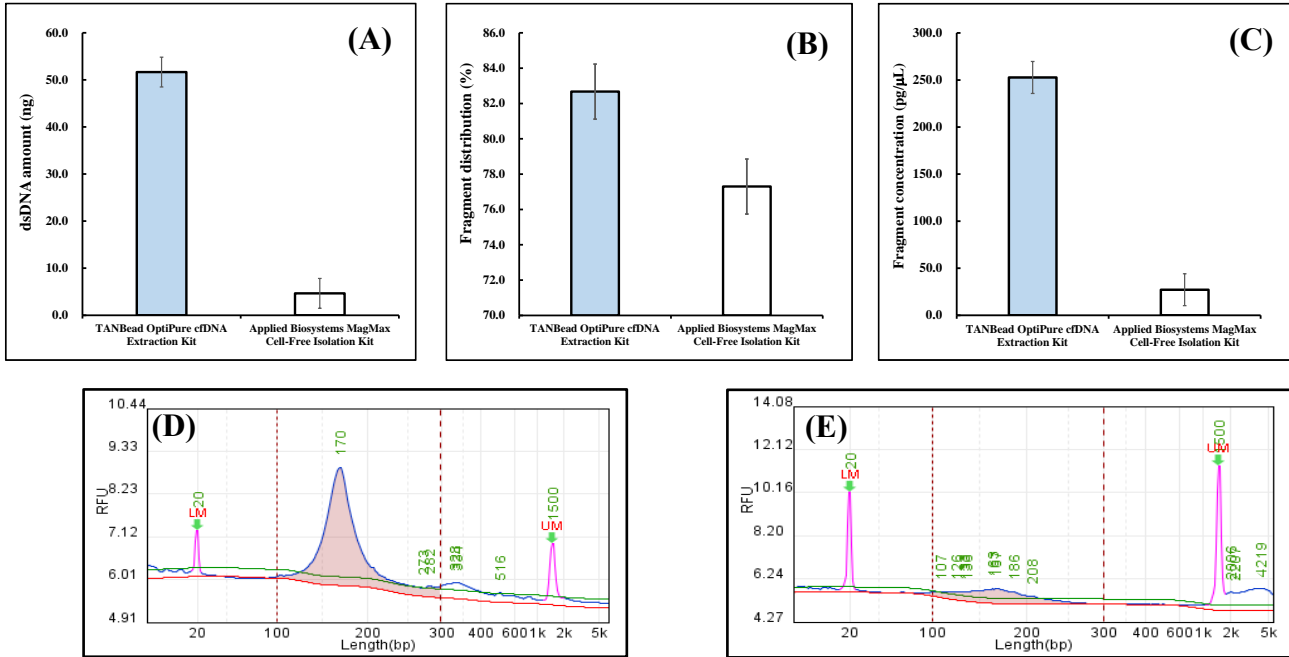
- Operate Qubit 4 Fluorometer by adding 2μL extracted DNA into 198 μL Qubit working solution for testing.

3.5 Use Qsep 100 with N1 high sensitivity cartridge to analyze the quality of extracted cfDNA



- Calibrate N1 high sensitivity cartridge according to the manual of Qsep 100.
- Take 10  $\mu$ L extracted sample for testing.

#### 4. Results



**Fig 1.** (A) Result of quantity of extracted dsDNA amount by using Qubit 4 Fluorometer. TANBead OptiPure cfDNA Extraction kit has higher amount of dsDNA yield compared to Applied Biosystems MagMax Cell-Free Isolation Kit; (B, C) Results of fragment analysis (100 ~300 bp) of extracted cfDNA by using Qsep 100 capillary electrophoresis. TANBead OptiPure cfDNA Extraction kit has higher fragment distribution (B) and concentration (C) compared to Applied Biosystems MagMax Cell-Free Isolation Kit; (D, E) TANBead OptiPure cfDNA Extraction kit (D) can obtain a major peak at 170 bp compared to Applied Biosystems MagMax Cell-Free Isolation Kit (E) suggesting that TANBead extraction system can obtain higher quality of cfDNA.

#### 5. Conclusion

According to the experiment results, you only need 1 mL sample to extract high quality cfDNA using TANBead OptiPure cfDNA Extraction Kit and Maelstrom 9600 automated nucleic acid extractor, which is very important for downstream applications such as establishment of NGS library and other clinical diagnosis application.

#### 6. Reference

- 6.1 <https://www.biopic.com.tw/index.php?action=application&cid=6&id=11&lang=EN> (BiOptic Inc.)
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