# BN-3P0: OPERATIONAL AND TECHNICAL SPECIFICATIONS

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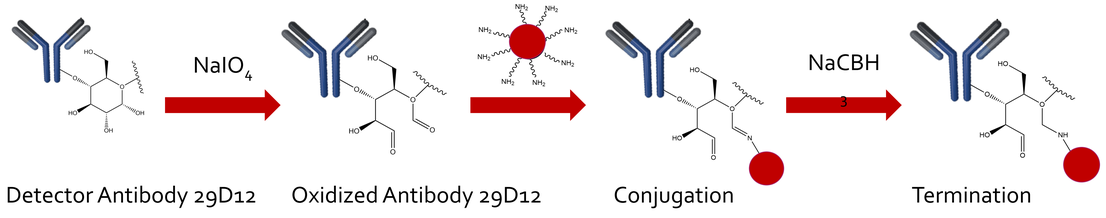
## sensing motif and molecular recognition

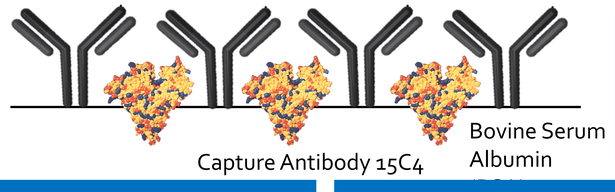
### sensor design

- Detection by fluorescent **particle-based immunoassay**.
- Antibodies selected based on kinetic assay.
  - MAb# 15C4 **Monoclonal mouse anti-human N-terminal proBNP (NT-proBNP)** (Hytest)
  - MAb# 29D12 **Monoclonal mouse anti-human N-terminal proBNP** (NT-proBNP) (Hytest)
- 15C4 acts as the capture antibody, cross linked to disc using **Glutaraldehyde Crosslinking**
- 29D12 acts as the detection antibody, conjugated to Latex Beads by **Reductive Amination**

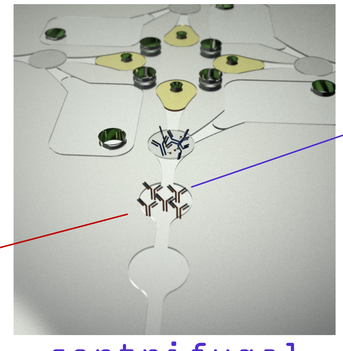
### detection antibody conjugated to latex beads




### capture antibody crosslinked to disc



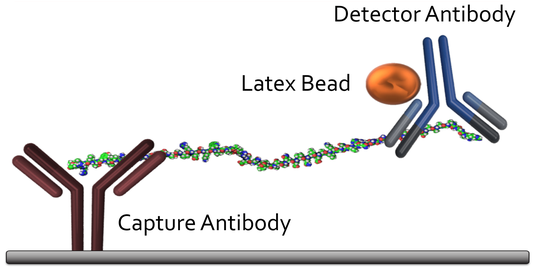

**glutaraldehyde crosslinking:** Relative aldehyde groups form Schiff base linkages between proteins – forming a layer on the surface of the PMMA. This facilitates detector capture via sandwich immunoassay in the detection well.



centrifugal microfluidic disk

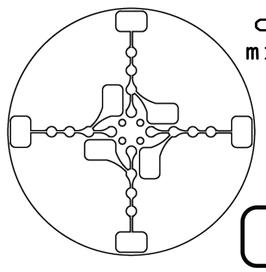
### sandwich immunoassay

**NT-proBNP capture**
NT-proBNP with ***regioselective*** capture and detection antibodies.

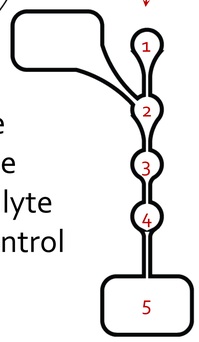



## microfluidics

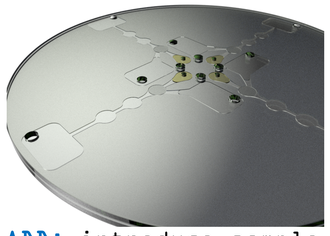
Our device relies on novel **sample processing**, **analyte capture, optical detection**, **and signal processing** to measure BNP levels in blood plasma in less than 10 minutes.



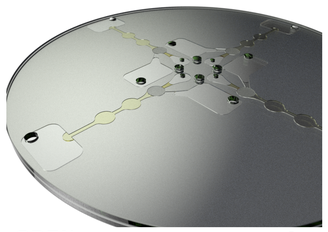
centrifugal microfluidic disk



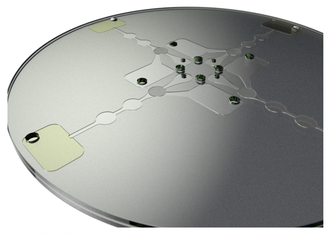

(1) load sample
(2) label analyte
(3) capture analyte
(4) measure control
(5) waste



**ADD:** introduce sample



**SPIN:** process sample

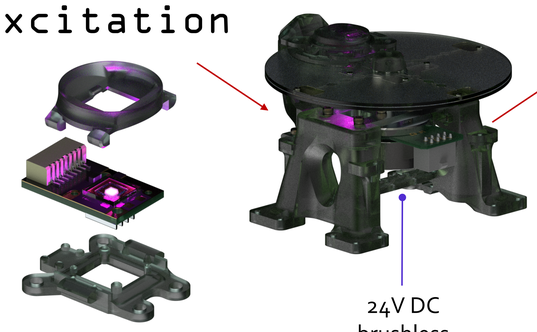


**READ:** analyze sample

**Device Specifications:**
- LOD: 40 pg/mL
- Working Range: 0-20,000 pg/mL
- Sample Volume: 5 µL
- Time to Results: 3-4 minutes
- Power Needed: <11 Watts

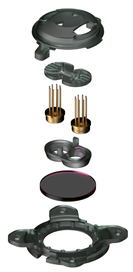
## fluorometric detection

excitation



24V DC brushless drive motor

emission



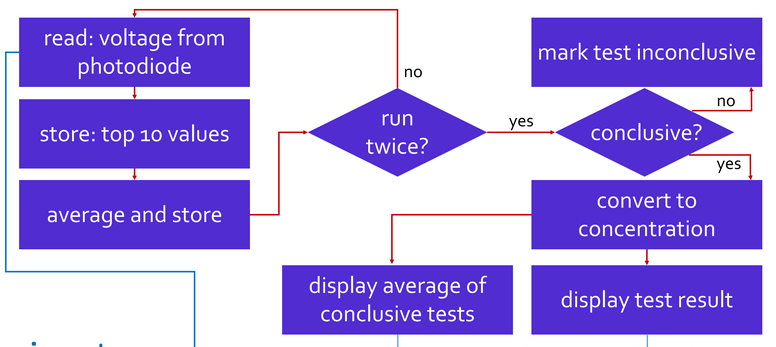
**LED** mounted under **lowpass filter** to block emission from photodiode.

bead emission passes above lowpass filter to allow **data collection using a photodiode**.

### multiplexed assay disc

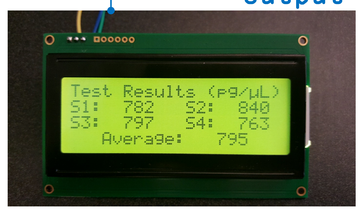

| | |
|---|---|
| Base Material: | Poly(methyl methacrylate, Oxygen-Plasma treated |
| Replicates per disc: | 4 (1 sample per channel) |
| Volume of sample: | 5 µL per well |
| Cost of production | Est. $4-10 per disc |
| Shelf life of lyophilized antibodies (4°C): | Multiple Years |

## signal processing




input


output




- The algorithm spins the disk and begins to sample the output voltage from the photodiode at a rate of **100,000 samples per second.**
- When a threshold value of voltage is reached, the **top ten values are saved locally**. This occurs for each well.
- The data is transformed into **a single point (max or average) of the readings**.
- The test is **performed twice** and data is checked from both runs.
- Wells with too much variation between test runs are **flagged as inconclusive**.
- The voltage average of both tests are converted to concentration using a pre-loaded **standard curve** and displayed to the user on a digital display.