

**Table S1. Details of single NGS library preparation steps in both canonical and optimized workflows**

|                                                                                  | Canonical                                                                          | Optimized                                    |
|----------------------------------------------------------------------------------|------------------------------------------------------------------------------------|----------------------------------------------|
| <b><i>1<sup>st</sup> amplification step (Molecule tagging) components:</i></b>   |                                                                                    |                                              |
| Breast cfDNA Panel v2                                                            | 2µl                                                                                | 0.6µl                                        |
| cfDNA Library PCR Master Mix                                                     | 15µl                                                                               | 5µl                                          |
| Nuclease-free water                                                              | 0µl                                                                                | 1.4µl                                        |
| DNA input                                                                        | 10µl normal genomic DNA (5ng) plus<br>3µl DNA from MDA sorted cells on<br>DEPArray | 3µl DNA from MDA sorted cells on<br>DEPArray |
| <b>Total volume</b>                                                              | <b>30µl</b>                                                                        | <b>10µl</b>                                  |
| <b><i>1<sup>st</sup> purification and elution steps:</i></b>                     |                                                                                    |                                              |
| Agencourt Ampure XP Reagent beads                                                | 45µl                                                                               | 15µl                                         |
| 80% ethanol                                                                      | 150µl                                                                              | 100µl                                        |
| Low TE                                                                           | 24µl                                                                               | 8µl                                          |
| <b><i>2<sup>nd</sup> amplification step (Library generation) components:</i></b> |                                                                                    |                                              |
| DNA from 1 <sup>st</sup> elution step                                            | 23µl                                                                               | 8µl                                          |
| Tag Sequencing BC (1-24)                                                         | 1µl                                                                                | 0.4µl                                        |
| cfDNA Library Primer P1                                                          | 1µl                                                                                | 0.4µl                                        |
| cfDNA Library PCR Master Mix                                                     | 25µl                                                                               | 8µl                                          |
| <b>Total volume</b>                                                              | <b>50µl</b>                                                                        | <b>16.8µl</b>                                |
| <b><i>2<sup>nd</sup> purification and elution steps:</i></b>                     |                                                                                    |                                              |
| Agencourt Ampure XP Reagent beads                                                | 57.5µl                                                                             | 19.3µl                                       |
| 80% ethanol                                                                      | 150µl                                                                              | 100µl                                        |
| Low TE                                                                           | 50µl                                                                               | 16.8µl                                       |
| <b><i>Size selection step:</i></b>                                               |                                                                                    |                                              |
| DNA from 2 <sup>nd</sup> elution step                                            | 50µl                                                                               | 16.8µl                                       |
| Agencourt Ampure XP Reagent beads                                                | 50µl                                                                               | 16.8µl                                       |
| 80% ethanol                                                                      | 150µl                                                                              | 100µl                                        |
| Low TE                                                                           | 30µl                                                                               | 22µl                                         |
| <b>Final transferred library volume</b>                                          | <b>28µl</b>                                                                        | <b>20µl</b>                                  |