Table S1. Index sequences of barcode.

|  |  |
| --- | --- |
| Index | Sequence |
| barcode1 | AGGCCAGAGCATTCGNNNNNNNN\* |
| barcode2 | CATCGGCGTACGACTNNNNNNNNATCCACGTGCTTGAG\* |
| barcode3 | NNNNNNNNGTGGCCGATGTTTCG\* |
| *index32* | GTGGCCGATGTTTCGCATCGGCGTACGACT |
| *index21* | ATCCACGTGCTTGAGAGGCCAGAGCATTCG |

\*NNNNNNNN referred to indicator sequences.

Table S2. The sequence of PrimerList.

|  |  |
| --- | --- |
| No. | Sequence |
| 1 | AGTCGTACGCCGATGCGAAACATCGGCCAC |
| 2 | CAGACGTGTGCTCTTCCGATCT |
| 3 | GTGGCCGATGTTTCGCATCGGCGTACGACT |
| 4 | CAAGCAGAAGACGGCATACGAGATGATCTGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT |
| 5 | CAAGCAGAAGACGGCATACGAGATTCAAGTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT |
| 6 | CAAGCAGAAGACGGCATACGAGATCTGATCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT |
| 7 | CAAGCAGAAGACGGCATACGAGATAAGCTAGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT |
| 8 | CAAGCAGAAGACGGCATACGAGATGTAGCCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT |
| 9 | CAAGCAGAAGACGGCATACGAGATTACAAGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT |
| 10 | CAAGCAGAAGACGGCATACGAGATTTGACTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT |
| 11 | CAAGCAGAAGACGGCATACGAGATGGAACTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT |
| 12 | AAGCAGTGGTATCAACGCAGAGT |
| 13 | AATGATACGGCGACCACCGAGATCTACACTAGATCGCTCGTCGGCAGCGTCAGATGTGTATAAGAGACAG |
| 14 | AAGCAGTGGTATCAACGCAGAGTGAATGGG |
| 15 | CGAATGCTCTGGCCTCTCAAGCACGTGGAT |
| 16 | ATCCACGTGCTTGAGAGGCCAGAGCATTCG |
| 17 | GTGGCCGATGTTTCGCATCGGCGTACGACT |
| 18 | AGATCGGAAGAGCACACGTCTG |
| 19 | AGTCGTACGCCGATGCGAAACATCGGCCAC |
| 20 | AGATCGGAAGAGCACACGTCTGAACTCCAGTCACCAGATCATCTCGTATGCCGTCTTCTGCTTG |
| 21 | AGATCGGAAGAGCACACGTCTGAACTCCAGTCACACTTGAATCTCGTATGCCGTCTTCTGCTTG |
| 22 | AGATCGGAAGAGCACACGTCTGAACTCCAGTCACGATCAGATCTCGTATGCCGTCTTCTGCTTG |
| 23 | AGATCGGAAGAGCACACGTCTGAACTCCAGTCACTAGCTTATCTCGTATGCCGTCTTCTGCTTG |
| 24 | AGATCGGAAGAGCACACGTCTGAACTCCAGTCACGGCTACATCTCGTATGCCGTCTTCTGCTTG |
| 25 | AGATCGGAAGAGCACACGTCTGAACTCCAGTCACCTTGTAATCTCGTATGCCGTCTTCTGCTTG |
| 26 | AGATCGGAAGAGCACACGTCTGAACTCCAGTCACAGTCAAATCTCGTATGCCGTCTTCTGCTTG |
| 27 | AGATCGGAAGAGCACACGTCTGAACTCCAGTCACAGTTCCATCTCGTATGCCGTCTTCTGCTTG |
| 28 | ACTCTGCGTTGATACCACTGCTT |
| 29 | CTGTCTCTTATACACATCTGACGCTGCCGACGAGCGATCTAGTGTAGATCTCGGTGGTCGCCGTATCATT |
| 30 | CCCATTCACTCTGCGTTGATACCACTGCTT |
| 31 | ATCCACGTGCTTGAGAGGCCAGAGCATTCG |
| 32 | CGAATGCTCTGGCCTCTCAAGCACGTGGAT |

Table S3. The uniquely identified and mapped reads information in SCSit.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Sample | Identified reads | | | |  | Mapped reads | | |
| Total no. | UMI and barcode identified | Abense barcode | Barcode with indel | Total no. | Consistent no.1 | Unique no.2 |
| SAMN08567263 | 7,142,680 | 6,529,228 | 37,968 | 575,484 | 5,924,198 | 5,551,723 | 372,475 |
| SAMN08567261 | 22,079,788 | 20,035,543 | 112,125 | 1,932,120 | 17,661,502 | 16,587,344 | 1,074,158 |
| SAMN08567259 | 20,440,162 | 19,084,797 | 104,712 | 1,250,653 | 16,127,622 | 15,014,861 | 1,112,761 |
| SAMN08567260 | 83,743,898 | 81,971,157 | 94,533 | 1,678,208 | 63,095,029 | 62,454,583 | 640,446 |
| SAMN08567264 | 86,777,528 | 84,999,601 | 99,277 | 1,678,650 | 62,799,705 | 62,037,223 | 762,482 |

1: Consistent was consistently identified reads using SCSit and original in uniquely mapped.

2: Unique was uniquely identified reads using SCSit in uniquely mapped.

Table S4. The identified reads number in SCSit with more than one mismatch and indel.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| BioSample | Identified reads No. | Increased reads No. | Increased ratio (%) | Runtime (h) | Increased time (%) |
| SAMN08567263 | 59,729,025 | 2,895,697 | 5.10 | 0.88 | 41.94 |
| SAMN08567261 | 170,496,465 | 6,715,843 | 4.10 | 2.48 | 30.53 |
| SAMN08567260 | 170,944,621 | 7,441,637 | 4.55 | 2.48 | 25.25 |
| SAMN08567259 | 154,181,681 | 5,758,512 | 3.88 | 2.47 | 32.09 |
| SAMN08567264 | 166,753,060 | 6,495,821 | 4.05 | 2.67 | 24.19 |