**Table S1. ddPCR results of postive control and negative control of MYD88 L265P**

| Sample | ddPCR for MYD88 L265P | | |
|---|---|---|---|
| | Mutant droplets | Total filled droplets | VAF |
| **Series dilution 1** | | | |
| Expected VAF 10% | 516 | 3756 | 0.1374 |
| Expected VAF 5% | 292 | 3777 | 0.0773 |
| Expected VAF 1% | 54 | 3316 | 0.0163 |
| Expected VAF 0.5% | 37 | 3707 | 0.0100 |
| Expected VAF 0.3% | 26 | 3527 | 0.0074 |
| Expected VAF 0.1% | 5 | 3596 | 0.0014 |
| **Series dilution 2** | | | |
| Expected VAF 10% | 470 | 3496 | 0.1344 |
| Expected VAF 5% | 262 | 3858 | 0.0679 |
| Expected VAF 1% | 56 | 3687 | 0.0152 |
| Expected VAF 0.5% | 29 | 3180 | 0.0091 |
| Expected VAF 0.3% | 19 | 3823 | 0.0050 |
| Expected VAF 0.1% | 6 | 4065 | 0.0015 |
| **Series dilution 3** | | | |
| Expected VAF 10% | 587 | 3913 | 0.1500 |
| Expected VAF 5% | 243 | 3586 | 0.0678 |
| Expected VAF 1% | | | FAIL |
| Expected VAF 0.5% | 34 | 3897 | 0.0087 |
| Expected VAF 0.3% | 20 | 3611 | 0.0055 |
| Expected VAF 0.1% | 9 | 3345 | 0.0027 |

**Table S2. ddPCR results of postive control and negative control of CD79B Y196**

| Sample | ddPCR for CD79B Y196 | | |
|---|---|---|---|
| | Mutant droplets | Total filled droplets | VAF |
| **series dilution1** | | | |
| Expected VAF 10% | 102 | 1120 | 0.0911 |
| Expected VAF 5% | 116 | 1888 | 0.0614 |
| Expected VAF 1% | 25 | 2456 | 0.0102 |
| Expected VAF 0.5% | 23 | 3048 | 0.0075 |
| Expected VAF 0.3% | 5 | 2458 | 0.0020 |
| Expected VAF 0.1% | 6 | 2918 | 0.0021 |
| **Series dilution 2** | | | |
| Expected VAF 10% | 147 | 1280 | 0.1148 |
| Expected VAF 5% | 117 | 1892 | 0.0618 |
| Expected VAF 1% | 27 | 2697 | 0.0100 |
| Expected VAF 0.5% | 23 | 3126 | 0.0074 |
| Expected VAF 0.3% | 6 | 2593 | 0.0023 |
| Expected VAF 0.1% | 4 | 3187 | 0.0013 |
| **Series dilution 3** | | | |
| Expected VAF 10% | 138 | 1414 | 0.0976 |
| Expected VAF 5% | 99 | 1861 | 0.0532 |
| Expected VAF 1% | 35 | 2995 | 0.0117 |
| Expected VAF 0.5% | 13 | 2426 | 0.0054 |
| Expected VAF 0.3% | 15 | 3221 | 0.0047 |
| Expected VAF 0.1% | 4 | 2644 | 0.0015 |

**Table S3. ddPCR results of healthy controls and patient samples**

| Case | Plasma sampling | Histology | Plasma collecting Tube | cfDNA yield/ml | ddPCR for MYD88 L265P | | | | | ddPCR for CD79B Y196 | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | Tissue | Plasma | | | | Tissue | Plasma | | | |
| | | | | | | Mutant droplets | Total filled droplets | VAF | Result cfDNA | | Mutant droplets | Total filled droplets | VAF | Result cfDNA |
| #1 | Diagnosis | PCNSL | EDTA | 4.2 | negative | 1 | 729 | 0.0014 | negative | negative | 0 | 372 | 0.0000 | inconclusive |
| #2 | Diagnosis | PCNSL | STRECK | 14 | positive | 1 | 685 | 0.0015 | negative | positive | 1 | 779 | 0.0013 | negative |
| #3 | Diagnosis | PCNSL | STRECK | 15.5 | negative | 0 | 1221 | 0.0000 | negative | negative | 0 | 1095 | 0.0000 | negative |
| #4 | Diagnosis | PCNSL | STRECK | 12 | positive | 2 | 2312 | 0.0009 | negative | negative | 2 | 931 | 0.0021 | negative |
| #5 | Diagnosis | PCNSL | STRECK | 34.5 | positive | 4 | 2423 | 0.0017 | negative | negative | 1 | 1595 | 0.0006 | negative |
| #6 | Diagnosis | PCNSL | STRECK | 14.1 | positive | 2 | 1329 | 0.0015 | negative | negative | 2 | 902 | 0.0022 | negative |
| #7 | Diagnosis | PCNSL | STRECK | 14 | positive | 1 | 1404 | 0.0007 | negative | positive | 1 | 650 | 0.0015 | negative |
| #8 | Diagnosis | PCNSL | STRECK | 8.9 | positive | 18 | 1774 | 0.0101 | positive | negative | 0 | 654 | 0.0000 | negative |
| #9 | Diagnosis | PCNSL | EDTA | 26 | | 3 | 1254 | 0.0024 | negative | | 1 | 1267 | 0.0008 | negative |
| #10 | Diagnosis | PCNSL | STRECK | 14.9 | | 0 | 1349 | 0.0000 | negative | | 0 | 525 | 0.0000 | negative |
| #11 | Diagnosis | PCNSL | STRECK | 18.9 | | 0 | 2780 | 0.0000 | negative | | 1 | 1284 | 0.0008 | negative |
| #12 | Diagnosis | PCNSL | STRECK | 10.8 | | 1 | 1494 | 0.0007 | negative | | 0 | 740 | 0.0000 | negative |
| #13 | Diagnosis | PCNSL | STRECK | 10.8 | | 0 | 1266 | 0.0000 | negative | | 0 | 628 | 0.0000 | negative |
| #14 | Diagnosis | PTLD-PCNSL | EDTA | 19.9 | | 0 | 1232 | 0.0000 | negative | | 0 | 963 | 0.0000 | negative |
| #15 | PD | PCNSL | EDTA | 65 | | 7 | 1949 | 0.0036 | positive | | 2 | 1266 | 0.0016 | negative |
| #16 | PD | PCNSL | STRECK | 8 | negative | 0 | 462 | 0.0000 | inconclusive | | 0 | 620 | 0.0000 | negative |
| #17 | PR | PCNSL | EDTA | 6.6 | positive | 0 | 1134 | 0.0000 | negative | negative | 0 | 258 | 0.0000 | inconclusive |
| #18 | PR | PCNSL | EDTA | 7 | positive | 7 | 1278 | 0.0055 | positive | negative | | | | not available |
| #19 | PR | PCNSL | EDTA | 10.1 | | 0 | 1095 | 0.0000 | negative | | 0 | 733 | 0.0000 | negative |
| #20 | PR | PCNSL | STRECK | 10.9 | | 2 | 2085 | 0.0010 | negative | | 1 | 886 | 0.0011 | negative |
| #21 | PR | PCNSL | STRECK | 16 | positive | 4 | 2470 | 0.0016 | negative | negative | 0 | 384 | 0.0000 | inconclusive |
| #22 | PR | PCNSL | STRECK | 8 | negative | 0 | 878 | 0.0000 | negative | negative | 0 | 406 | 0.0000 | inconclusive |
| #23 | PR | PTLD-PCNSL | STRECK | 44.5 | | 0 | 1831 | 0.0000 | negative | | 0 | 1085 | 0.0000 | negative |
| #24 | PR | PTLD-PCNSL | STRECK | 16.1 | negative | 0 | 1479 | 0.0000 | negative | negative | 0 | 815 | 0.0000 | negative |
| **WBC** | | | | | | | | | | | | | | |
| 1 | WBC | | | | | 0 | 3313 | 0.0000 | negative | | 4 | 3073 | 0.0013 | negative |
| 2 | WBC | | | | | 0 | 1569 | 0.0000 | negative | | 3 | 1587 | 0.0019 | negative |
| 3 | WBC | | | | | 0 | 917 | 0.0000 | negative | | 0 | 836 | 0.0000 | negative |
| 4 | WBC | | | | | 1 | 4218 | 0.0002 | negative | | 0 | 4058 | 0.0000 | negative |
| 5 | WBC | | | | | 0 | 1378 | 0.0000 | negative | | 0 | 1276 | 0.0000 | negative |
| 6 | WBC | | | | | 0 | 710 | 0.0000 | negative | | 1 | 816 | 0.0012 | negative |
| 7 | WBC | | | | | 2 | 5765 | 0.0003 | negative | | 1 | 5318 | 0.0002 | negative |
| 8 | WBC | | | | | 1 | 1292 | 0.0008 | negative | | 3 | 2493 | 0.0012 | negative |
| 9 | WBC | | | | | 1 | 2723 | 0.0004 | negative | | 0 | 1024 | 0.0000 | negative |
| 10 | WBC | | | | | 2 | 4465 | 0.0004 | negative | | 2 | 3717 | 0.0005 | negative |
| 11 | WBC | | | | | 2 | 1383 | 0.0014 | negative | | 0 | 1292 | 0.0000 | negative |
| 12 | WBC | | | | | 0 | 661 | 0.0000 | negative | | 2 | 584 | 0.0034 | negative |
| 13 | WBC | | | | | 0 | 2381 | 0.0000 | negative | | 2 | 2085 | 0.0010 | negative |
| 14 | WBC | | | | | 1 | 500 | 0.0020 | negative | | 0 | 511 | 0.0000 | negative |
| 15 | WBC | | | | | 0 | 324 | 0.0000 | inconclusive | | 1 | 275 | 0.0036 | inconclusive |
| 16 | WBC | | | | | 1 | 4622 | 0.0002 | negative | | 1 | 4422 | 0.0002 | negative |
| 17 | WBC | | | | | 0 | 1581 | 0.0000 | negative | | 0 | 1427 | 0.0000 | negative |
| 18 | WBC | | | | | 1 | 847 | 0.0012 | negative | | 0 | 797 | 0.0000 | negative |
| 19 | WBC | | | | | 1 | 2891 | 0.0003 | negative | | 3 | 3211 | 0.0009 | negative |
| 20 | WBC | | | | | 1 | 1592 | 0.0006 | negative | | 0 | 1578 | 0.0000 | negative |
| 21 | WBC | | | | | 0 | 768 | 0.0000 | negative | | 1 | 871 | 0.0011 | negative |
| 22 | WBC | | | | | 1 | 3907 | 0.0003 | negative | | 1 | 3179 | 0.0003 | negative |
| 23 | WBC | | | | | 1 | 2114 | 0.0005 | negative | | 0 | 2021 | 0.0000 | negative |
| 24 | WBC | | | | | 1 | 987 | 0.0010 | negative | | 0 | 960 | 0.0000 | negative |
| **Controls** | | | | | | | | | | | | | | |
| 1 | Positive control_1 | | | | | 3050 | 3051 | 0.9997 | positive | | 348 | 1129 | 0.3082 | positive |
| 2 | Positive control_2 | | | | | 1682 | 1682 | 1.0000 | positive | | 231 | 706 | 0.3272 | positive |
| 3 | Negative control_1 | | | | | 0 | 3566 | 0.0000 | negative | | 3 | 3253 | 0.0009 | negative |
| 4 | Negative control_2 | | | | | 0 | 2087 | 0.0000 | negative | | 1 | 1654 | 0.0006 | negative |
| 5 | No template control_1 | | | | | 0 | 0 | | | | 0 | 0 | | |
| 6 | No template control_2 | | | | | 0 | 0 | | | | 0 | 0 | | |

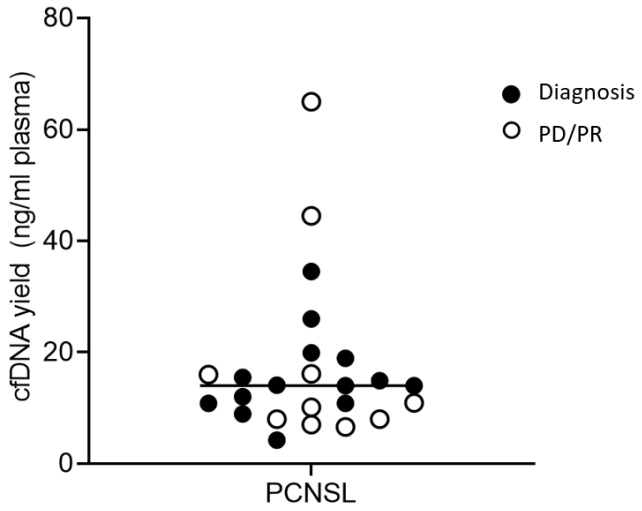


**Figure S1. cfDNA yields of plasma samples from PCNSL patients**. DNA yields ranged from 4.4 to 65 ng per ml of plasma. The median DNA yield in PCNSL cases was 14 ng/ml (range 4.4 to 65 ng/ml). Partial response (PR); Progressive disease (PD).

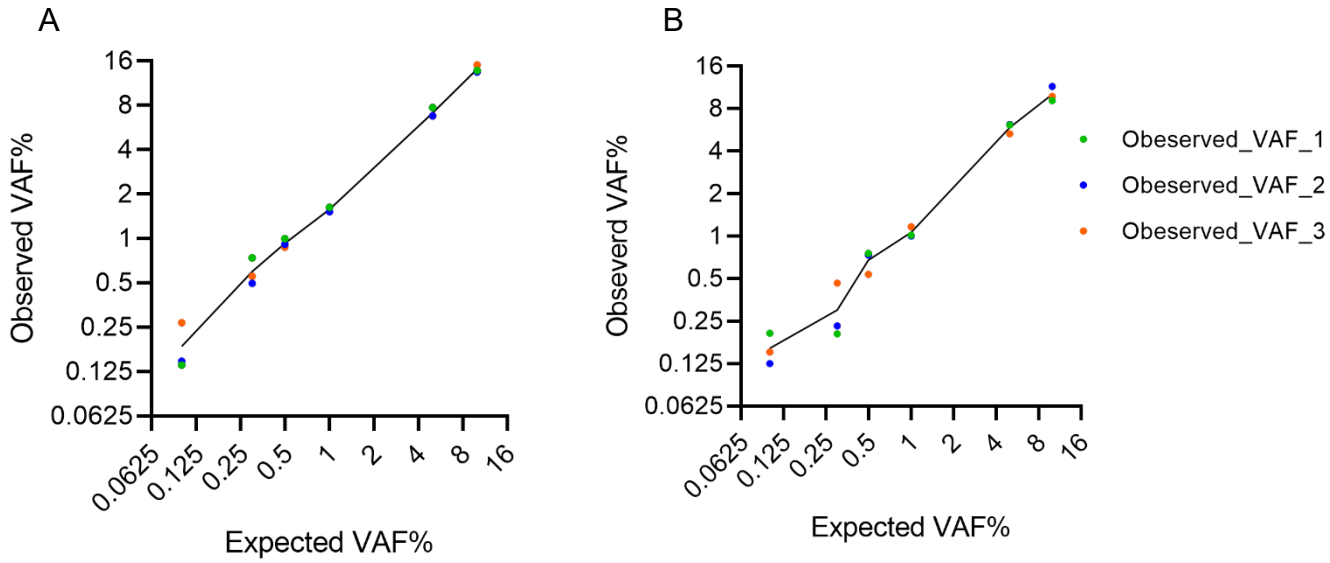


**Figure S2. Stimulate VAF of MYD88 L265P and CD79B Y196**. A. ddPCR test of MYD88 L265P on three independent series dilutions, expected VAFs is 10%, 5%, 1%, 0.5%, 0.3%, 0.1%. Observed results show a clear linear pattern ranging from 14% to 0.25%. B. ddPCR test of CD79B Y196 on three independent series dilutions, expected VAFs is 10%, 5%, 1%, 0.5%, 0.3%, 0.1%. Observed results show a clear linear pattern ranging from 10% to 0.5%. The black line shows the average VAF of the three independent experiments.