**SUPPLEMENTAL INFORMATION**

## Contents

**Supplemental Methods**

*Patient Descriptions – MS Patients*

Patient **MS-1** has relapsing-remitting MS (RRMS) with 3 attacks in the 12 months prior to lumbar puncture (LP); brain and cervical spinal cord MRIs were consistent with MS and showed an increase in lesion load in the 7 months prior to LP; she received a 3-day course of 1000 mg intravenous methylprednisolone 2 weeks prior to LP; EDSS was 1.0.

Patient **MS-2** experienced an attack of multifocal neurological symptoms 2 years prior to LP associated with multiple T2 hyperintense foci on MRI suggestive of demyelinating disease, consistent with the diagnosis of a clinically isolated syndrome (CIS). She was treated with a course of glucocorticoids, and gradually improved. LP revealed OCB. This patient developed a clinical relapse while this manuscript was under preparation. Therefore, this patient now carries a diagnosis of MS; EDSS at the time of her LP was 2.0.

Patient **MS-3** has RRMS for >3 years with typical MRI and CSF findings; treatment was with glatiramer acetate (GA) continuously for 15 months prior to LP; EDSS was 2.0. This patient participated in our studies exclusively for research purposes; no WBC, WBC differentiation, and OCB analysis was obtained. Her previous LP revealed CSF-restricted OCB.

Patient **MS-4** has RRMS with 2 attacks within the two years prior to LP; MRI and CSF findings showed changes typical of MS; EDSS was 1.5.

Patient **MS-5** had an initial episode of neurological symptoms 2 months prior to LP; brain MRI performed 1 week prior to LP revealed a single contrast-enhancing lesion; a diagnosis of clinically definite MS was reached by McDonald-criteria (1); EDSS was 1.5.

Patient **MS-6** presented with left face numbness and diplopia 1 month prior to LP; her MS diagnosis was confirmed by repeat brain MRI showing numerous new T2 hyperintense lesions after 6 months; her EDSS was 2.0.

*Patient Descriptions OND-Patients (see MRI images below)*

Patient **OND-1** has a single asymptomatic non-enhancing cerebellar lesion; her CSF is "negative" with respect to OCB and other inflammatory markers (IgG-Index, white cell count), and a clinical and imaging follow-up after 14-months revealed an unchanged clinical and imaging situation. This patient may have radiologically isolated syndrome (RIS) (2) but cannot be classified as having either CIS, or MS (1).

Patient **OND-2's** most likely diagnosis is a low-grade cervical spinal cord glioma. In particular, her lesion extends beyond the spinal cord circumference and thus appears tumor-like, has grown very slowly over the course of 2 years, does not show pathological contrast enhancement, and is a solitary lesion of considerable size. If this lesion were indeed of a demyelinating nature, a significant clinical neurological deficit would be expected; however, the patient displays only very little neurological symptoms. In this clinical constellation a biopsy of the cervical cord lesion to confirm the diagnosis of glioma is contraindicated given the very high risk of complications.

Patient **OND-3** had a single episode with C3-C4 level myelitis 2 years prior to LP; MRI findings improved over time; CSF was normal; NMO-IgG antibodies were negative. He had been treated with IFN-$\beta$ 1a for a presumed CIS, but treatment was discontinued shortly after the LP because the patient did not meet diagnostic criteria for MS (1) and normal CSF findings placed him in a lower risk group to develop MS in the future.

Patient **OND-4** had a single episode of a high cervical cord transverse myelitis 5 months prior to LP. CSF findings and brain MRI were normal. The patient did not meet diagnostic criteria for MS (1).

Patient **OND-5** presented with generalized body pain and on brain MRI had innumerable small foci of increased T2 signal without enhancement; these were largely atypical for demyelinating

disease and were thought to more likely represent microvascular events. CSF was normal. The patient did not meet diagnostic criteria for MS (1).

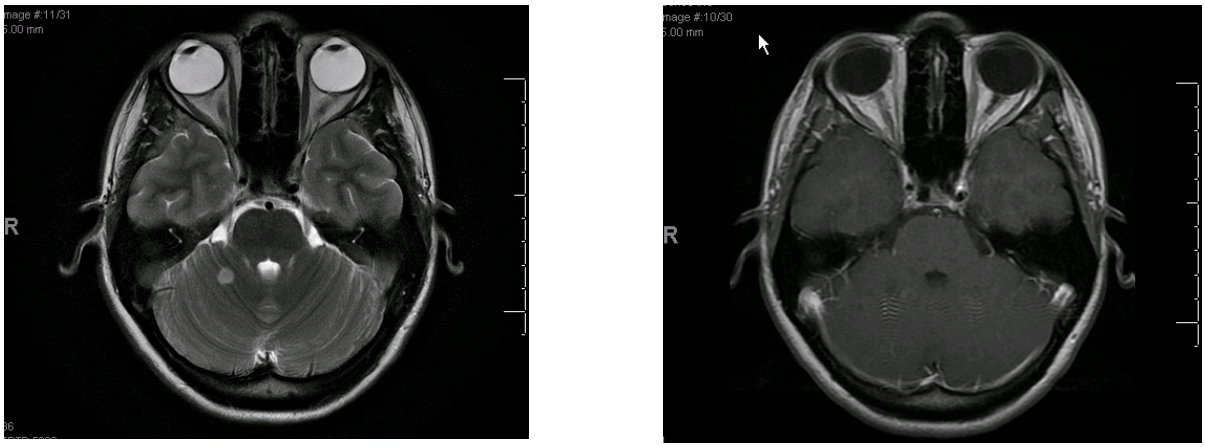
Patient **OND-6** had a slowly progressive clinical myelopathy, and underwent an anterior cervical discectomy and fusion C5/6 in 2009 for spinal canal stenosis. In 2005 cervical spine MRI revealed spinal stenosis from C4 to C6; spinal cord signal abnormality was not noted. In 2008 cervical spine MRI revealed progressive spinal stenosis at C5/6 and a newly described abnormal T2 signal hyperintensity at C4-5, and to a lesser degree at level C5-7; pathological contrast enhancement was not noted. In 2012 cervical spine MRI revealed persistent cervical spinal cord T2 signal abnormality at level C5-6. Her brain MRI revealed no abnormal findings in 2008 and 2012. There is no evidence for a clinical or radiographic dissemination in time or space. Her CSF analysis in 2011 revealed 5 OCB, a normal IgG-Index of 0.6 and normal cell count. The patient did not meet diagnostic criteria for MS (1).

Patient **OND-7** has chronic migraine; in 2009 she developed right shoulder and arm weakness lasting a few weeks. Brain MRI revealed non-specific foci of increased T2 and FLAIR signal; cervical spine MRI was unremarkable. CSF was normal, and an extensive diagnostic workup was unrevealing. The patient did not meet diagnostic criteria for MS (1).

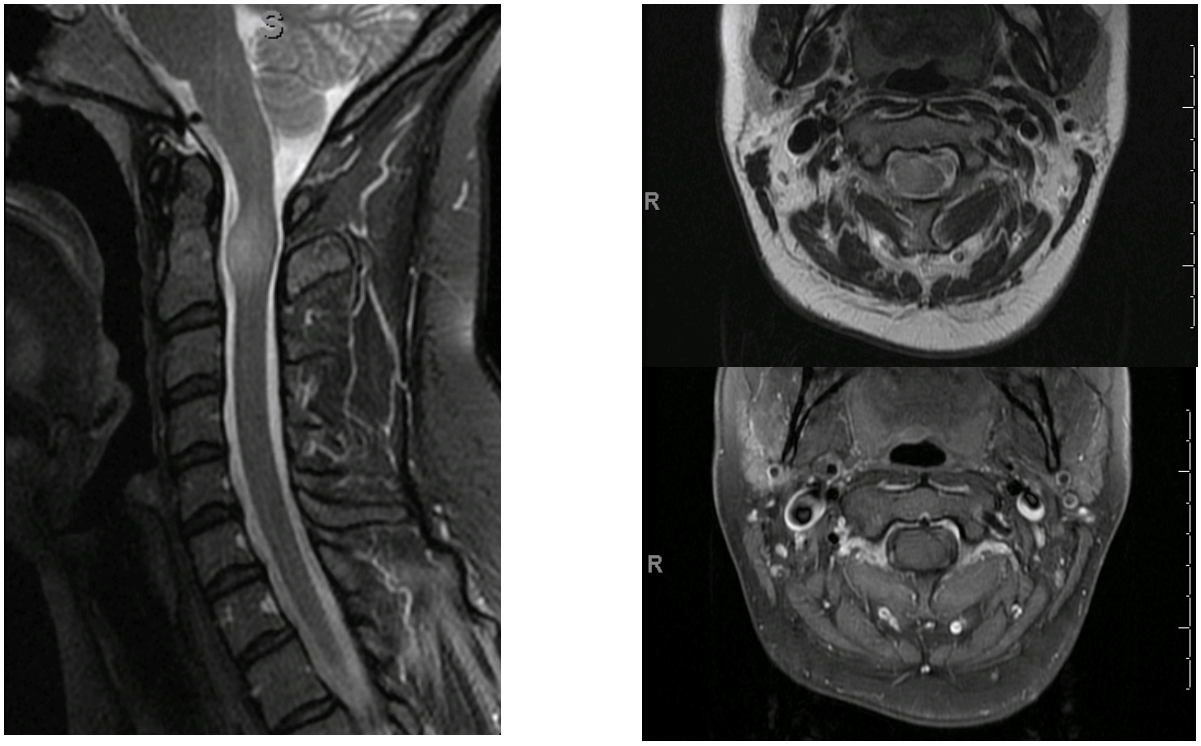
**References**

1. Polman, C.H., Reingold, S.C., Banwell, B., Clanet, M., Cohen, J.A., Filippi, M., Fujihara, K., Havrdova, E., Hutchinson, M., Kappos, L., et al. 2011. Diagnostic criteria for multiple sclerosis: 2010 revisions to the McDonald criteria. *Ann Neurol* 69:292-302.
2. Okuda, D.T., Mowry, E.M., Beheshtian, A., Waubant, E., Baranzini, S.E., Goodin, D.S., Hauser, S.L., and Pelletier, D. 2009. Incidental MRI anomalies suggestive of multiple sclerosis: the radiologically isolated syndrome. *Neurology* 72:800-805.

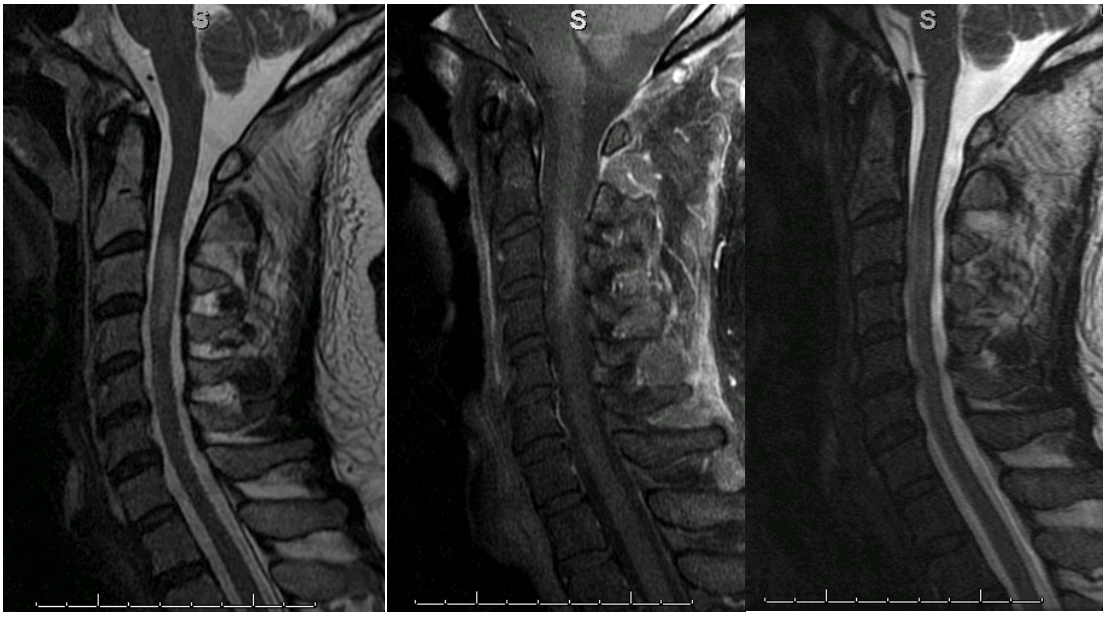
***MRI Images of OND Patients***



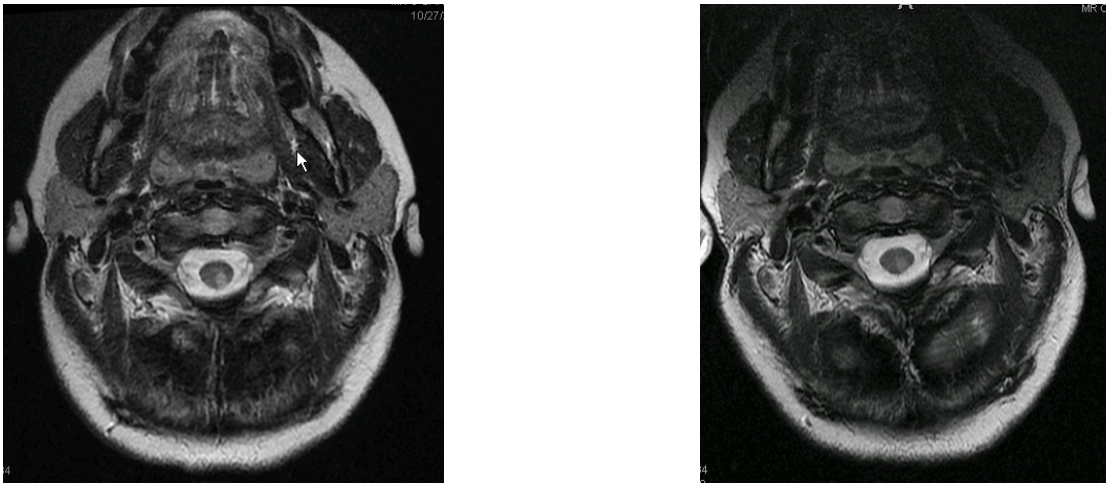
Patient **OND-1**: Brain MRI 09/2010. Left panel shows axial T2 with a single focus of increased T2 signal in the left cerebellar hemisphere. This lesion did not show pathological contrast enhancement. 14 months later there was no change to the patient's MRI findings.



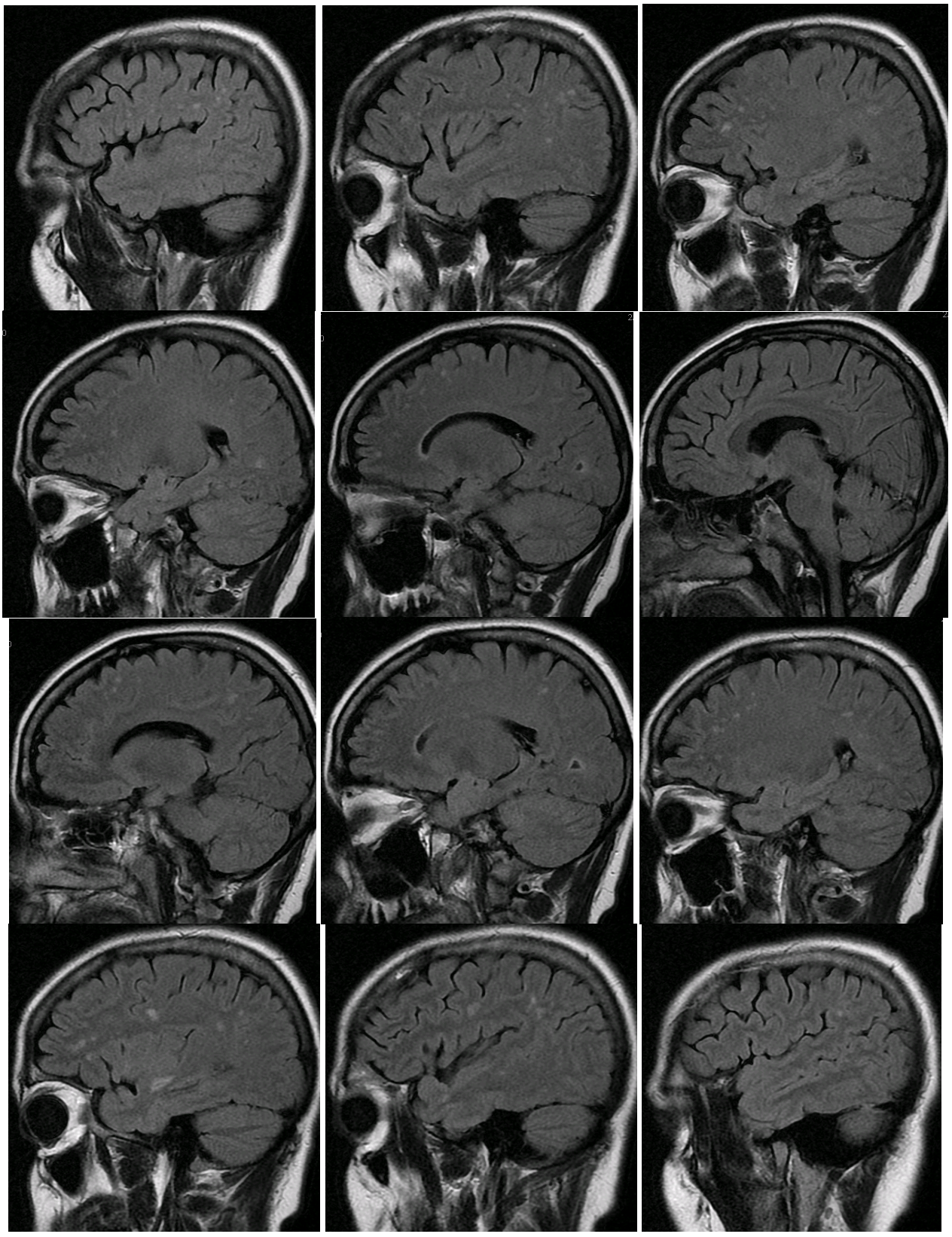
Cervical spinal cord MRI (04/2012) of patient **OND-2**. Left panel, sagittal T2; right upper panel axial T2 at level C2; right lower panel, axial T1 post gadolinium at level C2. The C2 level lesion has increased slightly in size since 2009.



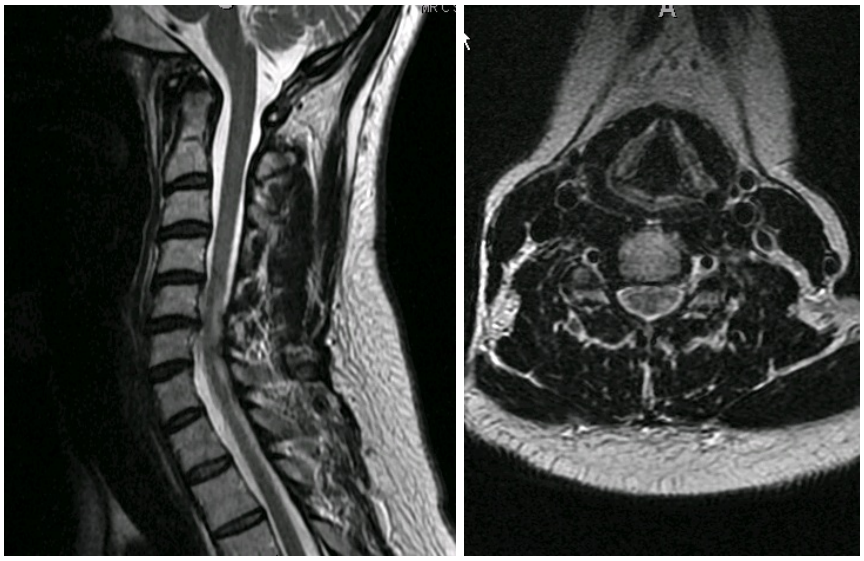
Patient **OND-3**: Left panel: Sagittal T2 C cord 07/2009. Middle panel: Sagittal T1+gadolinium 08/2009. Right panel: Sagittal T2 C cord 09/2009.



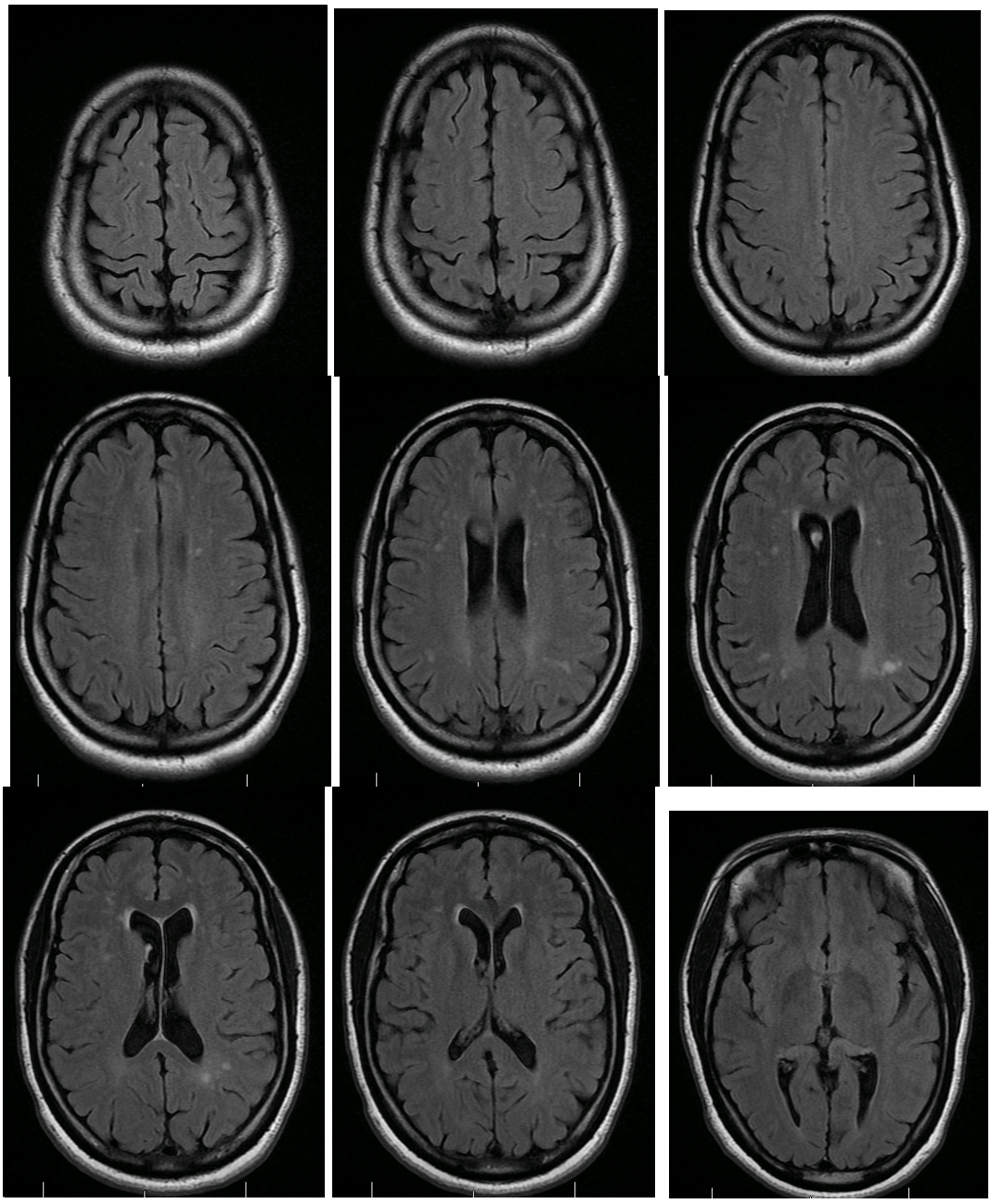
Patient **OND-4**: Left panel: Axial T2 C cord 10/2010 showing a left posterior focus of increased T2 signal at level C1; there was very subtle gadolinium enhancement (not shown). Right panel: Axial T2 C cord 02/2011 showing the same lesion with less T2 hyperintensity 4 months later. Brain MRI was normal (not shown).



Patient **OND-5**: Sagittal FLAIR 02/2011: Top left panel to bottom right, left to right sections, respiectively. There was no pathological contrast enhancement after gadolinium (not shown).



Patient **OND-6**: Left panel: Sagittal T2 C cord 07/2009. Right panel: Sagittal T1+gadolinium 08/2009.



Patient **OND- 7**: Axial FLAIR sequences showing numerous foci of abnormal T2 signal which prompted further MS diagnostic work-up in this patient. As described above, a MS diagnosis could not be established in this patient.

## Supplemental Tables

| ID | CSF (all reads) | PBMC (all reads) | CSF (V,J,CDR3) | PBMC (V,J,CDR3) | CSF (non-red) | PBMC (non-red) |
|---|---|---|---|---|---|---|
| MS-1 | 6,660 | 735,584 | 5,661 | 507,411 | 270 | 17,151 |
| MS-2 | 27,130 | 307,641 | 20,752 | 235,515 | 383 | 9,657 |
| MS-3 | 34,813 | 397,723 | 25,658 | 243,301 | 625 | 10,986 |
| MS-4 | 11,981 | 423,533 | 6,164 | 347,616 | 170 | 10,123 |
| MS-5 | 30,907 | 296,397 | 26,226 | 256,809 | 494 | 9,348 |
| MS-6 | 34,550 | 438,860 | 30,286 | 344,805 | 432 | 13,260 |
| OND-1 | 21,854 | 415,570 | 6,759 | 341,955 | 74 | 11,034 |
| OND-2 | 11,073 | 380,129 | 3,933 | 308,411 | 107 | 17,750 |
| OND-3 | 18,021 | 393,717 | 4,059 | 324,620 | 89 | 9,484 |
| OND-6 | 26,965 | 435,258 | 14,262 | 339,707 | 81 | 5,620 |
| OND-4 | -* | 317,020 | -* | 277,157 | -* | 10,015 |
| OND-5 | -* | 350,603 | -* | 273,079 | -* | 7,549 |
| OND-7 | -* | 307,517 | -* | 272,077 | -* | 7,498 |

Supplemental Table 1: Numbers of sequences obtained by deep repertoire sequencing ("all reads"). "V, J, CDR3" are numbers of sequences with identifiable IGHV, IGHJ and H-CDR3 sequences. Clonally non-redundant datasets ("non-red") for IGHV usage analyses were generated by considering only one representative from a set of reads bearing the same IGHV-segment, IGHVJ-segment, H-CDR3 length, and H-CDR3 amino acid composition that differed by less than two residues from another member in the clonal family. *No IgG-VH sequences could be amplified from these patients' mRNA. Presence of viable mRNA was confirmed by RT-PCR of GAPDH.

| Patient ID | Figure | CDR3 | IGHV | mut | IGHJ |
|---|---|---|---|---|---|
| MS-1 | | CARVYYKWNDEWSWFDPW | IGHV1-3 | 28 | IGHJ5 |
| | | CAREPGTSLHCSGGNCYSKDASNFW | IGHV1-69 | 30 | IGHJ3 |
| | | CARGRYSRSSRYYFDLW | IGHV1-69 | 21 | IGHJ4 |
| | 2A | CARIEPSSSRGSLFFFDYW | IGHV2-70 | 18 | IGHJ4 |
| | | CTTDEGGNSGYYFEYW | IGHV3-15 | 34 | IGHJ4 |
| | 5A | CAKAGGYSGSSGRRNFYSFDSW | IGHV3-23 | 10 | IGHJ4 |
| | | CARGTGGGWYYRIHFDYW | IGHV3-23 | 23 | IGHJ4 |
| | | CAKVRFVEWLHLFDYW | IGHV3-23 | 12 | IGHJ4 |
| | 5C | CARVVNYDILTGYFRDAFDIW | IGHV3-23 | 16 | IGHJ3 |
| | | CARDQGGYYGNFFLDYW | IGHV3-48 | 30 | IGHJ4 |
| | | CARQKSTRTTRYYYMYGLDVW | IGHV3-48 | 23 | IGHJ6 |
| | | CAGRRYFESSGPPEFPW | IGHV3-48 | 16 | IGHJ5 |
| | 3A | **CARHGTSLTPSYSRFYYW** | **IGHV4-39** | **18** | **IGHJ4** |
| | | **CARSGTKKVMGHFDYW** | **IGHV4-39** | **8** | **IGHJ4** |
| | | **CARSGPGGEPVYYFDPW** | **IGHV4-59/61** | **34** | **IGHJ5** |
| | | **CARVRRAGGRSWFDPW** | **IGHV4-59/61** | **14** | **IGHJ5** |
| | | CASHRGQWLVTSW | IGHV5-a | 27 | IGHJ5 |
| OND-1 | | CAREDCSAASCYWAYLHHW | IGHV1-18 | 19 | IGHJ1 |
| | | CARGEAPTKLQLLGTFDFW | IGHV1-46 | 26 | IGHJ4 |
| | | CAKDSAGPVLWVKPNWFDPW | IGHV3-23 | 10 | IGHJ5 |
| | | CARDSMPDKDFADYFPYW | IGHV3-30/33rn | 32 | IGHJ1 |
| | | CTRAGPPELWDYFYGLDVW | IGHV3-49 | 8 | IGHJ6 |
| | | CVRDRDWNYGDCW | IGHV3-7 | 18 | IGHJ4 |
| | | CVKSPTYSSGQGYFDYW | IGHV3-9 | 11 | IGHJ4 |
| | | **CVRDRAVAAHHDAFDIW** | **IGHV4-59/61** | **33** | **IGHJ3** |
| OND-2 | | CARTFYDSSEMGGSW | IGHV1-18 | 10 | IGHJ5 |
| | | CAHCSGGTCAFDYW | IGHV1-2 | 3 | IGHJ4 |
| | | CAREFDSW | IGHV1-2 | 9 | IGHJ4 |
| | | CANSTSSETIWFDPW | IGHV1-2 | 23 | IGHJ5 |
| | | CGRGERKATPMEVW | IGHV1-8 | 33 | IGHJ6 |
| | | CARRRTLGHCSSSSCGRAFDIW | IGHV2-5 | 17 | IGHJ3 |
| | | CARDLKGREVMGPHPPDFDYW | IGHV3-30/33rn | 15 | IGHJ4 |
| | | CARDPTQTIPTGFSGYFDYW | IGHV3-30/33rn | 20 | IGHJ4 |
| | | CARARDYYDSSGYGYW | IGHV3-48 | 13 | IGHJ4 |
| | | CARDGPTGALDYW | IGHV3-48 | 12 | IGHJ4 |
| | | CAKWYNSGYRAFDIW | IGHV3-53/66 | 17 | IGHJ3 |
| | | CARGVDSWSGGPGDW | IGHV3-7 | 23 | IGHJ4 |
| | | CVLRNSGTLYFASW | IGHV3-9 | 21 | IGHJ4 |
| | | CARWDNYYDSGYFDYW | IGHV4-30-4/31 | 4 | IGHJ4 |
| | | CARAYDFWSGYEYYAMDVW | IGHV4-34 | 8 | IGHJ6 |
| | | CARGRGTMASASSGWDYW | IGHV4-34 | 36 | IGHJ4 |
| OND-3 | | CAKAKRHCSNAGCPVPMSASDYW | IGHV1-18 | 16 | IGHJ4 |
| | | CARGSYDKPFDYW | IGHV1-2 | 32 | IGHJ4 |
| | | CARSLKQLVRTFGYW | IGHV1-2 | 14 | IGHJ4 |

| | | | | | |
|---|---|---|---|---|---|
| | | CASAQYPDDGLSTYYYYGIDVW | IGHV1-69 | 13 | IGHJ6 |
| | | CARGTQIGDLDGYFGLDVW | IGHV1-69 | 21 | IGHJ6 |
| | | CARGYGDYGRVFDFW | IGHV1-69 | 23 | IGHJ4 |
| | | CARDAPKLVAGNKHWDYW | IGHV3-21 | 14 | IGHJ4 |
| | | CAKVFRGAVAGSFDYW | IGHV3-23 | 14 | IGHJ4 |
| | | CATDLGSSLTGYYYFW | IGHV3-30/33rn | 22 | IGHJ5 |
| | | CMVGNYHGSGRHDYW | IGHV3-49 | 40 | IGHJ4 |
| | | CARGNPLTNLRRGTGFDPW | IGHV4-34 | 24 | IGHJ5 |
| | | **CARQGFSYGLRTMAHFDFW** | **IGHV4-39** | **30** | **IGHJ2** |
| | | **CARQGYYDHWSGYSSWFDPW** | **IGHV4-39** | **13** | **IGHJ5** |
| MS-2 | | CARGPYYYDISGDSYAHYGMDVW | IGHV3-13 | 12 | IGHJ6 |
| | | CTRERSYSYLRSGYFDYW | IGHV3-15 | 17 | IGHJ4 |
| | | CVPIKGAEANYW | IGHV3-15 | 17 | IGHJ4 |
| | | CTRGLLWLDGDYRDYW | IGHV3-30/33rn | 20 | IGHJ4 |
| | | CAGGQTGYCTGGNSQRCYGMDVW | IGHV3-53/66 | 34 | IGHJ6 |
| | | CARVDATSWYYFDSW | IGHV3-53/66 | 22 | IGHJ4 |
| | | CAKVKGRQLVHYYYGMDVW | IGHV3-53/66 | 21 | IGHJ6 |
| | | CARDEPFDYDTSSYYSPFDSW | IGHV3-7 | 3 | IGHJ4 |
| | | CAREGVYDNLLDETDAFDVW | IGHV3-7 | 19 | IGHJ3 |
| | | CARVPRRFSAYDV | IGHV3-7 | 6 | IGHJ5 |
| | 3C | CTTESWYTFYSW | IGHV3-72 | 32 | IGHJ5 |
| | | CVRAFTSSWDKGGSVDWFDPW | IGHV4-30-4/31 | 12 | IGHJ5 |
| | | CARDERITLIPGAFDVW | IGHV4-30-4/31 | 7 | IGHJ3 |
| | | CGRDGRGRELLPFPGFDSW | IGHV4-30-4/31 | 16 | IGHJ4 |
| | | CARDLDYGSGNLAYW | IGHV4-30-4/31 | 16 | IGHJ4 |
| | | CARDLSGSGSYFPVDAW | IGHV4-30-4/31 | 18 | IGHJ5 |
| | | CARDPAESGYFSGTFDIW | IGHV4-30-4/31 | 16 | IGHJ3 |
| | | CARDRVGWVRGVPFGAW | IGHV4-30-4/31 | 8 | IGHJ5 |
| | | CARETGLFKSFYFDYW | IGHV4-30-4/31 | 16 | IGHJ4 |
| | | CARGLTGWYPDNW | IGHV4-30-4/31 | 29 | IGHJ4 |
| | | CARGRGYSSNWYPLRFDSW | IGHV4-30-4/31 | 8 | IGHJ4 |
| | | CARRASPHHYDGSGEDYW | IGHV4-30-4/31 | 29 | IGHJ4 |
| | | CAHTKDAGMLTWLEHW | IGHV4-30-4/31 | 34 | IGHJ4 |
| | | CARVDRTAGYYFDNW | IGHV4-30-4/31 | 14 | IGHJ4 |
| | | CARVPRGGPNPFIRGTFDYW | IGHV4-30-4/31 | 14 | IGHJ4 |
| | | CARVVDYFDHW | IGHV4-30-4/31 | 17 | IGHJ4 |
| | | CARGRGFYDGTGHQNYFDPW | IGHV4-34 | 14 | IGHJ5 |
| | | **CAGDGSGSYYTRFDQW** | **IGHV4-39** | **7** | **IGHJ4** |
| | | **CARGLPRLLSPHSDW** | **IGHV4-39** | **31** | **IGHJ4** |
| | | **CARHGLPPPYNWNDVGSTHQYFDSW** | **IGHV4-39** | **19** | **IGHJ5** |
| | | **CARHGTGDSW** | **IGHV4-39** | **30** | **IGHJ4** |
| | | **CARHGTGDSWYRGIRVGDW** | **IGHV4-39** | **29** | **IGHJ4** |
| | | **CARHPAYSSNWYLPIYYFHHW** | **IGHV4-39** | **18** | **IGHJ1** |
| | | **CARHPDSSDNTGRAFFNPFDYW** | **IGHV4-39** | **19** | **IGHJ4** |
| | | **CARHPPPGSSGFYGHDAFDVW** | **IGHV4-39** | **8** | **IGHJ3** |
| | | **CARHPTPFIAGPGTFDYW** | **IGHV4-39** | **11** | **IGHJ4** |

| | | | | |
|---|---|---|---|---|
| **MS-2** | **CARHRDYYDSSGFYYRRAFAMW** | **IGHV4-39** | **12** | **IGHJ3** |
| | **CARHRGIAVAVQLNPSRNYFDYW** | **IGHV4-39** | **13** | **IGHJ4** |
| | **CVRHSGGHYYDDSVYKWMSYYFDYW** | **IGHV4-39** | **16** | **IGHJ4** |
| | **CTRHSHYQLQGVMPINWFDPW** | **IGHV4-39** | **20** | **IGHJ5** |
| | **CARHSPHYFYGSGSYQGWFGPW** | **IGHV4-39** | **23** | **IGHJ5** |
| | **CAAIRGRMAAAYHW** | **IGHV4-39** | **26** | **IGHJ4** |
| | **CVRLGPDYGDFNFDYW** | **IGHV4-39** | **13** | **IGHJ4** |
| | **CARLGYCSSISCYADYW** | **IGHV4-39** | **11** | **IGHJ4** |
| | **CARLGYCSSISCYAESW** | **IGHV4-39** | **15** | **IGHJ5** |
| | **CARLMKGSKDIAVAGTFDYW** | **IGHV4-39** | **16** | **IGHJ4** |
| | **CARLNDYDSGDYYWNFDYW** | **IGHV4-39** | **18** | **IGHJ4** |
| | **CARLQYGSGSYPYYYYGMDVW** | **IGHV4-39** | **13** | **IGHJ6** |
| | **CVALRYFDWSVGKLFDYW** | **IGHV4-39** | **15** | **IGHJ4** |
| | **CARPGGSDGYNPFDFW** | **IGHV4-39** | **15** | **IGHJ4** |
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| | **CARQGYYYDSSGLFDYW** | **IGHV4-39** | **16** | **IGHJ4** |
| | **CARQPNRFLSDRSGGDYW** | **IGHV4-39** | **13** | **IGHJ5** |
| | **CARQPNRFYYDSSGGDYW** | **IGHV4-39** | **10** | **IGHJ5** |
| | **CTKRSSWYGEHYYGMDVW** | **IGHV4-39** | **20** | **IGHJ6** |
| | **CAKSPPLGNSGAFDIW** | **IGHV4-39** | **15** | **IGHJ3** |
| | **CARVEMSTIRGVGLLDYW** | **IGHV4-39** | **13** | **IGHJ4** |
| | **CARVGPTTSDAVPGGLFYWYFDLW** | **IGHV4-39** | **25** | **IGHJ2** |
| | **CVAVRYFAWSVGKLFDYW** | **IGHV4-39** | **16** | **IGHJ4** |
| | CARDTFRQTTMVTRWFDPW | IGHV4-4 | 18 | IGHJ5 |
| | CARTVRYVDWRNKLLYHFDHW | IGHV4-4 | 29 | IGHJ4 |
| | CARVPLLRDGGIYYGLDVW | IGHV4-4 | 14 | IGHJ6 |
| | **CARAYDTNSQGPFDFW** | **IGHV4-59/61** | **22** | **IGHJ4** |
| | **CARGSDILTGYSLGGWFDPW** | **IGHV4-59/61** | **13** | **IGHJ5** |
| | **CARLRRDGPQEYFHHW** | **IGHV4-59/61** | **22** | **IGHJ1** |
| | **CAKRAAYCDGKRCSRAFDYW** | **IGHV4-59/61** | **16** | **IGHJ4** |
| | **CASRMGGQQLVWGYW** | **IGHV4-59/61** | **17** | **IGHJ4** |
| | **CARSVGASGTLTGYFHHW** | **IGHV4-59/61** | **19** | **IGHJ1** |
| | **CARYFAPGHYGSGDGAFDIW** | **IGHV4-59/61** | **19** | **IGHJ3** |
| | CARRIAPLRGTYSMFAFDIW | IGHV5-51 | 16 | IGHJ3 |
| | CARPQRSSSSTFDHW | IGHV5-a | 25 | IGHJ4 |
| MS-3 | CARGGYCTGGSCYEPEFDYW | IGHV1-18 | 13 | IGHJ4 |
| | CAHWESGYSYSRGGGSRRRYFDSW | IGHV2-5 | 11 | IGHJ4 |
| | CTTVKLGWRSSYYDTAFDYW | IGHV3-15 | 16 | IGHJ4 |
| | CVKDQGDYIWGTYPSTFDYW | IGHV3-23 | 20 | IGHJ4 |
| | CANRYLVDSSGSYRDDPFDIW | IGHV3-23 | 5 | IGHJ3 |
| | CAKTRYADYYVVGNYFNYW | IGHV3-23 | 20 | IGHJ4 |
| | CTRARVLAARSRYADYNYGMDVW | IGHV3-30/33rn | 24 | IGHJ6 |
| | CARKPDRGYNLDDYGEYKPTSPFDYW | IGHV3-30/33rn | 21 | IGHJ4 |
| | CARDAPDCGGDCYSLPTVRFDYW | IGHV3-7 | 10 | IGHJ4 |
| | CARDEETFVWGSYRDQTNYYYYGMDVW | IGHV3-7 | 8 | IGHJ6 |
| | CVRDHQWLVLGRRCDSW | IGHV3-7 | 17 | IGHJ5 |

| | | | | | |
|---|---|---|---|---|---|
| MS-3 | | CAKDWTDAGAVTNVFDYW | IGHV3-7 | 24 | IGHJ4 |
| | | CARDYGSATYYASYYHGMGVW | IGHV3-7 | 13 | IGHJ6 |
| | | CARGIAAYSGGRYDSHFDSW | IGHV3-7 | 21 | IGHJ4 |
| | | CARGQGSGRYYRLRFDYW | IGHV3-7 | 11 | IGHJ4 |
| | | CARRGATTPRGRTLDSW | IGHV3-7 | 9 | IGHJ4 |
| | | CARVLTVRGVSSQGFDSW | IGHV3-7 | 18 | IGHJ4 |
| | | CARVPPWVGTITPLFDYW | IGHV3-7 | 6 | IGHJ4 |
| | | CARGYHSFDMW | IGHV3-72 | 15 | IGHJ3 |
| | | CARDAKWFGESNYYDMDVW | IGHV4-30-4/31 | 12 | IGHJ6 |
| | | CARDKTWLGELGIEHNAFDVW | IGHV4-30-4/31 | 13 | IGHJ3 |
| | | CARDLHDYGDYFDYW | IGHV4-30-4/31 | 10 | IGHJ4 |
| | | CARDYDYGDKWFDPW | IGHV4-30-4/31 | 14 | IGHJ5 |
| | | CAREGGAVAGILVW | IGHV4-30-4/31 | 14 | IGHJ5 |
| | 2B | CAREGIPDYDYYGMDVW | IGHV4-30-4/31 | 22 | IGHJ6 |
| | | CARESAHDSSYFYFDSW | IGHV4-30-4/31 | 10 | IGHJ4 |
| | | CARGPAVLRYFDRLLHFDYW | IGHV4-30-4/31 | 34 | IGHJ4 |
| | | CASNVYDVLTAYYSGGSYFDHW | IGHV4-30-4/31 | 12 | IGHJ4 |
| | | CASQYYDILTAYYNVGSWFDPW | IGHV4-30-4/31 | 13 | IGHJ5 |
| | | CASRSGYTNNNNWFDPW | IGHV4-30-4/31 | 11 | IGHJ5 |
| | | CVRSKYDILTGYYDKGHAFHIW | IGHV4-30-4/31 | 10 | IGHJ3 |
| | | CARTGMRNWFDLW | IGHV4-30-4/31 | 17 | IGHJ5 |
| | | CAKTSGWFGVDSW | IGHV4-30-4/31 | 19 | IGHJ4 |
| | | CARVPAAIDTLLKFLFDSW | IGHV4-30-4/31 | 6 | IGHJ4 |
| | | CARVPDYESDPVIWGSYFDAW | IGHV4-30-4/31 | 25 | IGHJ5 |
| | | CRWGGDMDVW | IGHV4-34 | 20 | IGHJ6 |
| | | CARGQYSEWDLTRPHYYYYAMDVW | IGHV4-34 | 14 | IGHJ6 |
| | | **CASEEYDIMNGRYGLSNWFGAW** | **IGHV4-39** | **12** | **IGHJ5** |
| | | **CARFPAQILPGYYVGRAGDYW** | **IGHV4-39** | **12** | **IGHJ4** |
| | | **CAKGEEGYYDTSGIFQQW** | **IGHV4-39** | **31** | **IGHJ1** |
| | | **CAGGGHWPQVFDFW** | **IGHV4-39** | **23** | **IGHJ4** |
| | | **CARHEIKQTPTTSKFDPW** | **IGHV4-39** | **17** | **IGHJ5** |
| | | **CARHELLQTPTTSKFGPW** | **IGHV4-39** | **30** | **IGHJ5** |
| | | **CARHIALPARAMYYHDTIGTPGPLDFW** | **IGHV4-39** | **17** | **IGHJ5** |
| | | **CARHNYDILTGYYSRPHYSDSW** | **IGHV4-39** | **6** | **IGHJ4** |
| | | **CARHNYDIL** | **IGHV4-39** | **6** | **IGHJ4** |
| | | **CARHQTGDDDYFDYW** | **IGHV4-39** | **16** | **IGHJ4** |
| | | **CARHVLAAGGTLPWGPKFPSPKYFDPW** | **IGHV4-39** | **21** | **IGHJ5** |
| | | **CARLNNFSTSTSYYDPPYYFYAMDVW** | **IGHV4-39** | **11** | **IGHJ6** |
| | | **CARLSSDYYINWFDFW** | **IGHV4-39** | **12** | **IGHJ5** |
| | | **CARLYYDVLTAYYGLPSYLDYW** | **IGHV4-39** | **10** | **IGHJ4** |
| | | **CARPHYDILTAYYNVASWFDPW** | **IGHV4-39** | **13** | **IGHJ5** |
| | | **CARPHYDILTGRYNVAHWFDPW** | **IGHV4-39** | **8** | **IGHJ5** |
| | | **CASPPEQGIW** | **IGHV4-39** | **20** | **IGHJ3** |
| | | **CARPPGTSVSWYFDLW** | **IGHV4-39** | **18** | **IGHJ2** |
| | | **CASPRAIGATGPFDYW** | **IGHV4-39** | **21** | **IGHJ4** |
| | | **CVRPSINYYDSSGYYFHDAFDIW** | **IGHV4-39** | **13** | **IGHJ3** |

| | | | | | |
|---|---|---|---|---|---|
| **MS-3** | | **CVRQGFYGSGTYYIPEDW** | **IGHV4-39** | **32** | **IGHJ4** |
| | | **CARQGYDILNAYYGRPHYFDYW** | **IGHV4-39** | **18** | **IGHJ4** |
| | | **CARQPGDYYDSTEYYFDYW** | **IGHV4-39** | **14** | **IGHJ4** |
| | | **CARQYYDEYSPNWFDSW** | **IGHV4-39** | **16** | **IGHJ5** |
| | | **CASQYYDILTGRYSAGQYFDYW** | **IGHV4-39** | **7** | **IGHJ4** |
| | | **CARRGYDMLTAYYGEGNWFDPW** | **IGHV4-39** | **20** | **IGHJ5** |
| | | **CARRPTYYPGSESAYRVVSW** | **IGHV4-39** | **14** | **IGHJ5** |
| | | **CASSSSWKGHFQHW** | **IGHV4-39** | **18** | **IGHJ1** |
| | | **CVGSSSWKGWFDPW** | **IGHV4-39** | **15** | **IGHJ5** |
| | | **CASSTGTTLRRSYFDSW** | **IGHV4-39** | **35** | **IGHJ4** |
| | | **CASSTGTTLRRSHFADW** | **IGHV4-39** | **18** | **IGHJ4** |
| | | **CARTSLLVPDTGPW** | **IGHV4-39** | **15** | **IGHJ5** |
| | | **CATVVPGNYYVSSGYFPDYW** | **IGHV4-39** | **10** | **IGHJ4** |
| | | CARADCDISGYSSWYFDYW | IGHV4-4 | 21 | IGHJ4 |
| | | CARAYYDISGYSSWYFDYW | IGHV4-4 | 23 | IGHJ4 |
| | | **CAKANVRHGFRILVGNYHPMDAW** | **IGHV4-59/61** | **19** | **IGHJ6** |
| | | **CARDLGNFDFW** | **IGHV4-59/61** | **13** | **IGHJ4** |
| | | **CARDLGQFDSW** | **IGHV4-59/61** | **18** | **IGHJ4** |
| | | **CAREMEGSGLW** | **IGHV4-59/61** | **25** | **IGHJ5** |
| | | **CARGKEGWDLRDSFYFDYW** | **IGHV4-59/61** | **8** | **IGHJ4** |
| | | **CARHFSDLVGTISYWRENYYYYGMDVW** | **IGHV4-59/61** | **11** | **IGHJ6** |
| | | **CARHRANGGMDVW** | **IGHV4-59/61** | **28** | **IGHJ6** |
| | | **CAGNRLGYDDNGHPHGMDLW** | **IGHV4-59/61** | **23** | **IGHJ6** |
| | | **CARRPGTGPIPYLYYGMDVW** | **IGHV4-59/61** | **16** | **IGHJ6** |
| | | **CARTEYNWFDPW** | **IGHV4-59/61** | **26** | **IGHJ5** |
| | | **CARTYGSGTYSRDYYYGMDVW** | **IGHV4-59/61** | **10** | **IGHJ6** |
| | | **CARVKVNFDSSGYTRSFDYW** | **IGHV4-59/61** | **7** | **IGHJ4** |
| | | **CARVRSAMDVW** | **IGHV4-59/61** | **9** | **IGHJ6** |
| MS-4 | | CARRERYSDSWYDYW | IGHV1-3 | 10 | IGHJ4 |
| | 2C | CAKSDDYDFHNIDSW | IGHV3-23 | 21 | IGHJ4 |
| | | CARDVFDAWYDHRFDFW | IGHV3-53/66 | 15 | IGHJ4 |
| | | CARDFYYTSGRYALDLW | IGHV3-7 | 23 | IGHJ3 |
| | | CARDQYSDGWPGTLGPRRLYYYGVAVW | IGHV3-7 | 13 | IGHJ6 |
| | | **CARDLGRGGYDYVWGTYRSRVFDYW** | **IGHV4-39** | **8** | **IGHJ4** |
| | | **CARDWYNTGWSPFYFDYW** | **IGHV4-39** | **15** | **IGHJ4** |
| | | **CARSWGFGEWYFDYW** | **IGHV4-39** | **20** | **IGHJ4** |
| | | CARDSDSGSWYARYFDVW | IGHV4-4 | 16 | IGHJ2 |
| | | **CAREIRFCNTASCHKWIDPW** | **IGHV4-59/61** | **20** | **IGHJ5** |
| | | **CAVGEMTTIVGSYYYFGMDVW** | **IGHV4-59/61** | **14** | **IGHJ6** |
| | | **CAQGEVVGTPSHFYYYPMDVW** | **IGHV4-59/61** | **12** | **IGHJ6** |
| MS-5 | | CARDLGGGAIYYYSYYMDVW | IGHV1-18 | 13 | IGHJ6 |
| | | CARERSRNAVVSEGAFDVW | IGHV1-18 | 42 | IGHJ3 |
| | | CAREPYTAMAASFDYW | IGHV1-2 | 1 | IGHJ4 |
| | | CARGLDGYNWNYVGYW | IGHV1-2 | 1 | IGHJ4 |
| | | CARVSIKSGSYLHDYW | IGHV1-2 | 7 | IGHJ4 |
| | | CARDGVYGARNYPWYFDLW | IGHV1-3 | 17 | IGHJ2 |

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|---|---|---|---|---|---|
| MS-5 | | CIRELSGGQFDNW | IGHV1-46 | 26 | IGHJ4 |
| | | CGSGSTPDFMDVW | IGHV1-46 | 24 | IGHJ6 |
| | | CARGDYGDYGYYYYYMDVW | IGHV1-69 | 12 | IGHJ6 |
| | | CARGIYIAGAGGKTYFDYW | IGHV1-69 | 27 | IGHJ4 |
| | | CATVTFPGYMDVW | IGHV1-69 | 25 | IGHJ6 |
| | | CALACRNAVCPQRDFDFNYW | IGHV1-8 | 19 | IGHJ4 |
| | | CTTDRTVGATRDFGYW | IGHV3-15 | 4 | IGHJ4 |
| | | CAKDGWELHQDAFDVW | IGHV3-23 | 17 | IGHJ3 |
| | | CAKDRAVGVGSGDSFESW | IGHV3-23 | 18 | IGHJ3 |
| | | CAMHRYSFAYSVIVDYW | IGHV3-23 | 8 | IGHJ4 |
| | | CARSPARLLDVW | IGHV3-23 | 15 | IGHJ6 |
| | | CVRGSRALGGTGDYW | IGHV3-30/33rn | 24 | IGHJ4 |
| | | CARLFYSKQGYDMDVW | IGHV3-30/33rn | 0 | IGHJ6 |
| | | CARDRQYQLLPAPTWFDPW | IGHV3-48 | 16 | IGHJ5 |
| | | CAREPGDFWSGDYFFDCW | IGHV3-48 | 20 | IGHJ4 |
| | | CARGGSLLYGYCSDGNCYLDYW | IGHV3-53/66 | 8 | IGHJ4 |
| | | CASSSLR | IGHV3-7 | 14 | IGHJ5 |
| | 2E | CVGFNPPIDYW | IGHV3-7 | 13 | IGHJ4 |
| | | CARGSAVAGNYW | IGHV3-7 | 18 | IGHJ4 |
| | | CARNADYDILTGYYRPGNFDFW | IGHV3-7 | 21 | IGHJ4 |
| | | CARDSSRWSFDVW | IGHV3-72 | 31 | IGHJ6 |
| | 4A | CSRMYNWNFDYW | IGHV3-72 | 19 | IGHJ4 |
| | | CTRRGCTSVSCSSIW | IGHV3-73 | 14 | IGHJ5 |
| | | CARDPHDYGGNRFDYW | IGHV3-74 | 3 | IGHJ4 |
| | | CSRDQHNFWTGSPYYMDVW | IGHV3-74 | 12 | IGHJ6 |
| | | CAEFGRHGDYW | IGHV3-74 | 19 | IGHJ4 |
| | | CAKEILRSKYDLWSGYYKPFDIW | IGHV3-9 | 17 | IGHJ3 |
| | | CAKEPDSSGWSTGGFDPW | IGHV3-9 | 13 | IGHJ5 |
| | 3B | CASQSGDIVVSSWYMDVW | IGHV4-30-2 | 1 | IGHJ6 |
| | | CARCHYGSGSKFFDYW | IGHV4-30-4/31 | 18 | IGHJ4 |
| | | CARGCTGGNCYFDYW | IGHV4-30-4/31 | 15 | IGHJ4 |
| | | CASPRTNYDPGNPMPFDIW | IGHV4-30-4/31 | 12 | IGHJ3 |
| | | CARRTSRREGVNWFDPW | IGHV4-30-4/31 | 17 | IGHJ5 |
| | | CARSSGPGRFDPW | IGHV4-30-4/31 | 30 | IGHJ5 |
| | | CARAFSTDLWGGYYKGNWFDPW | IGHV4-34 | 11 | IGHJ5 |
| | | CASALSTDLWSGYYKGNWFDPW | IGHV4-34 | 3 | IGHJ5 |
| | | **CARDGNDFWSGHHAGYFDLW** | **IGHV4-39** | **22** | **IGHJ4** |
| | 4C | **CAREGPMVRGVTRTFDCW** | **IGHV4-39** | **21** | **IGHJ4** |
| | 5B | **CARHHPSWATTGPDSW** | **IGHV4-39** | **28** | **IGHJ4** |
| | | **CVRPSGGRNWYFDVW** | **IGHV4-39** | **24** | **IGHJ2** |
| | | **CARQLIARSQVDIVVVVTGTFFDYW** | **IGHV4-39** | **13** | **IGHJ4** |
| | | **CARQYPSWSTTGPDYW** | **IGHV4-39** | **4** | **IGHJ4** |
| | | **CARRRHSGVTDSDYIWGSEREDAFDIW** | **IGHV4-39** | **10** | **IGHJ3** |
| | | CARDQGSSGWGDAFELW | IGHV4-4 | 18 | IGHJ3 |
| | | CATGGPRMDYW | IGHV4-4 | 22 | IGHJ4 |
| | | **CARARWFGELRPPAPYMDVW** | **IGHV4-59/61** | **14** | **IGHJ6** |

| | | | | | |
|---|---|---|---|---|---|
| MS-5 | | **CARFGARYCDSPRCQGYYYYYYMDVW** | **IGHV4-59/61** | **25** | **IGHJ6** |
| | | **CARFIDSSIWGSFPDYW** | **IGHV4-59/61** | **5** | **IGHJ4** |
| | | **CAGRGFWSPYYYYMDVW** | **IGHV4-59/61** | **2** | **IGHJ6** |
| | | **CARVGDDFWSGFWPLW** | **IGHV4-59/61** | **30** | **IGHJ5** |
| | | **CARVGTTSYYYYYMDVW** | **IGHV4-59/61** | **5** | **IGHJ6** |
| | 3D | **CARVKEDFWSGYTFDYW** | **IGHV4-59/61** | **13** | **IGHJ4** |
| | | **CARVNVETRDSTSAPYYGAVDFW** | **IGHV4-59/61** | **20** | **IGHJ4** |
| OND-6 | | CARDAAGTYDFWSGYYSDW | IGHV3-7 | 9 | IGHJ4 |
| | | CARGGDGYNFYYYHYGMDVW | IGHV4-30-4/31 | 11 | IGHJ6 |
| MS-6 | | CAREHRYGSFATGDYFDYW | IGHV1-2 | 13 | IGHJ4 |
| | | CARERVVSSSSNWGYW | IGHV1-2 | 19 | IGHJ4 |
| | | CARDDGWWSGFHGDYRWFESW | IGHV1-69 | 25 | IGHJ5 |
| | | CASGDLGHCTRTHCYEDQYYYYYYMDVW | IGHV1-69 | 23 | IGHJ6 |
| | | CARPVTGTRGAFDIW | IGHV1-69 | 15 | IGHJ3 |
| | | CASSAYCSGGSCLNWFDPW | IGHV1-69 | 21 | IGHJ5 |
| | 2D | CASGDLGHCTTSNCYEDQYYYYYYMDVW | IGHV1-69 | 14 | IGHJ6 |
| | | CARRRSPPYRSGMDVW | IGHV3-13 | 9 | IGHJ6 |
| | | CVREYNWNDGNWIDPW | IGHV3-21 | 26 | IGHJ5 |
| | 3E | CAKRWGAGGQFDYW | IGHV3-23 | 20 | IGHJ4 |
| | 4B | CAREGFGVFDYW | IGHV3-30/33rn | 20 | IGHJ4 |
| | 5D | CAKGIGRDYDFWSGYFYYW | IGHV3-30/33rn | 10 | IGHJ4 |
| | | CAKKDVNYYGIDVW | IGHV3-30/33rn | 15 | IGHJ6 |
| | | CATEGVDCRAGNCYWGFYYW | IGHV3-48 | 22 | IGHJ4 |
| | | CATEGVDCRAGNCYW | IGHV3-48 | 21 | IGHJ6 |
| | | CARDRAVVTDYYDSSGYQNDVDVFDLW | IGHV3-49 | 24 | IGHJ3 |
| | | **CARDRELDGYNSPFFDYW** | **IGHV4-59/61** | **19** | **IGHJ4** |
| | | **CARTYNWNYWSGLDYW** | **IGHV4-59/61** | **13** | **IGHJ4** |

Supplemental Table 2: Characteristics of IgG-VH clusters containing clonally related sequences present either exclusively in the CSF or in both, CSF, and PB. IgG-VH sequences were clustered based on usage of identical IGHV and IGHJ germline segments and H-CDR3 aminoacid sequences using a defined distance metric (see Methods). "CDR3" is the most frequent H-CDR3 aminoacid sequence present per cluster. IGHV and IGHJ are the closest germline segments per network and "Mut" are average numbers of nucleotide mutations present in IGHV ranging from CDR1 to FR3 among all sequences present in the cluster. IgG-VH using IGHV4-39 and IGHV4-59/61 which appear to be overrepresented in MS CSF are in bold letters. Per cluster all IgG-VH sequences that contained high-quality sequence information ranging from the 5' end of H-CDR1 to the 3' end of H-CDR3 were selected from the database to generate IgG-VH lineage trees (Figures 2 to 5); column "Figure" indicates respective trees in Figures 2 to 5 and clusters in Figure 1.

| IGHV | MS-1 | MS-2 | MS-3 | MS-4 | MS-5 | MS-6 | OND-1 | OND-2 | OND-3 | OND-6 | p |
|---|---|---|---|---|---|---|---|---|---|---|---|
| IGHV1-18 | 0 | 0.26 | 3.04 | 0 | 1.01 | 0 | 17.57 | 0 | 5.62 | 0 | n.s. |
| IGHV1-2 | 0 | 0.52 | 0 | 0 | 1.01 | 12.96 | 0 | 6.54 | 2.25 | 0 | n.s. |
| IGHV1-24 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 1.87 | 0 | 0 | n.s. |
| IGHV1-3 | 8.15 | 0 | 0 | 5.88 | 0.4 | 0 | 0 | 0.93 | 0 | 0 | n.s. |
| IGHV1-46 | 0 | 0 | 0 | 0 | 1.62 | 0 | 50 | 0.93 | 0 | 0 | n.s. |
| IGHV1-69 | 14.07 | 0.26 | 0 | 0 | 1.82 | 64.81 | 0 | 0.93 | 25.84 | 1.23 | n.s. |
| IGHV1-8 | 0 | 0 | 0 | 0 | 0.61 | 0 | 0 | 0.93 | 0 | 0 | n.s. |
| IGHV2-26 | 0 | 0.26 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV2-5 | 0 | 0.26 | 1.6 | 0 | 0 | 0 | 0 | 1.87 | 0 | 0 | n.s. |
| IGHV2-70 | 10.37 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-11 | 0 | 0 | 0 | 0 | 0.2 | 0.46 | 0 | 1.87 | 4.49 | 0 | n.s. |
| IGHV3-13 | 0 | 4.44 | 0 | 0 | 0.2 | 0.46 | 0 | 0.93 | 0 | 0 | n.s. |
| IGHV3-15 | 2.96 | 1.04 | 0.16 | 0 | 0.4 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-20 | 0 | 0 | 0 | 0 | 0.2 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-21 | 0 | 0 | 0 | 0 | 1.01 | 2.08 | 0 | 0.93 | 5.62 | 0 | n.s. |
| IGHV3-23 | 21.85 | 0.78 | 4.8 | 17.06 | 2.83 | 0.23 | 1.35 | 0.93 | 4.49 | 0 | n.s. |
| IGHV3- | 0.37 | 0.78 | 0.48 | 0 | 1.62 | 14.12 | 2.7 | 22.43 | 0 | 4.94 | n.s. |
| IGHV3-48 | 20 | 0 | 0 | 0 | 1.42 | 0 | 0 | 17.76 | 0 | 0 | n.s. |
| IGHV3-49 | 0 | 0 | 0 | 0 | 0.2 | 0.69 | 10.81 | 0.93 | 5.62 | 0 | n.s. |
| IGHV3-53/66 | 0.37 | 2.61 | 0 | 8.82 | 2.43 | 0 | 0 | 10.28 | 0 | 0 | n.s. |
| IGHV3-7 | 0 | 2.87 | 10.24 | 18.82 | 9.51 | 0.23 | 1.35 | 0.93 | 0 | 0 | n.s. |
| IGHV3-72 | 0 | 0 | 0.16 | 0 | 7.49 | 0 | 0 | 0.93 | 0 | 0 | n.s. |
| IGHV3-74 | 0 | 0 | 0 | 0 | 0.81 | 0.23 | 0 | 1.87 | 0 | 0 | n.s. |
| IGHV3-9 | 0 | 0 | 0 | 0 | 1.62 | 0 | 9.46 | 14.95 | 0 | 0 | n.s. |
| IGHV3-h | 2.22 | 0 | 0 | 0 | 0 | 0 | 0 | 2.8 | 0 | 0 | n.s. |
| IGHV4-30-2 | 0 | 0.78 | 0.16 | 0 | 0.4 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV4-30- | 0 | 19.84 | 16 | 0 | 20.04 | 0 | 0 | 1.87 | 1.12 | 93.83 | n.s. |
| IGHV4-34 | 0 | 1.04 | 1.6 | 0 | 0.81 | 0 | 0 | 3.74 | 29.21 | 0 | n.s. |
| **IGHV4-39** | **8.89** | **42.56** | **52.48** | **18.24** | **23.68** | **0** | **0** | **0** | **14.61** | **0** | **0.04** |
| IGHV4-4 | 0 | 3.13 | 0.64 | 10.59 | 0.81 | 0 | 0 | 0 | 0 | 0 | n.s. |
| **IGHV4-59/61** | **10.74** | **13.32** | **8.32** | **20.59** | **17.21** | **3.7** | **6.76** | **1.87** | **1.12** | **0** | **0.01** |
| IGHV4-b | 0 | 0 | 0.32 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV5-51 | 0 | 4.96 | 0 | 0 | 0.61 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV5-a | 0 | 0.26 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV7-4-1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0.93 | 0 | 0 | n.s. |

Supplemental Table 3: Percent usage per IGHV germline segment in the **CSF** of MS and OND patients. Comparison was made between MS and OND and p-values were calculated using a resampling-based permutation test. Only germline segments are represented which were found in at least one CSF sample. IGHV4-39 and IGHV4-59/61 appear significantly overrepresented in MS CSF; n.s., not significant.

| IGHV | MS-1 | MS-2 | MS-3 | MS-4 | MS-5 | MS-6 | OND-1 | OND-2 | OND-3 | OND-6 | p |
|---|---|---|---|---|---|---|---|---|---|---|---|
| IGHV1-18 | 0 | 0 | 1.18 | 0 | 3.28 | 0 | 12.50 | 6.25 | 7.69 | 0 | n.s. |
| IGHV1-2 | 0 | 0 | 0 | 0 | 4.92 | 11.11 | 0 | 18.75 | 15.38 | 0 | n.s. |
| IGHV1-3 | 5.88 | 0 | 0 | 8.33 | 1.64 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV1-46 | 0 | 0 | 0 | 0 | 3.28 | 0 | 12.50 | 0 | 0 | 0 | n.s. |
| IGHV1-69 | 11.76 | 0 | 0 | 0 | 4.92 | 27.78 | 0 | 0 | 23.08 | 0 | n.s. |
| IGHV1-8 | 0 | 0 | 0 | 0 | 1.64 | 0 | 0 | 6.25 | 0 | 0 | n.s. |
| IGHV2-5 | 0 | 0 | 1.18 | 0 | 0 | 0 | 0 | 6.25 | 0 | 0 | n.s. |
| IGHV2-70 | 5.88 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-13 | 0 | 1.41 | 0 | 0 | 0 | 5.56 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-15 | 5.88 | 2.82 | 1.18 | 0 | 1.64 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-21 | 0 | 0 | 0 | 0 | 0 | 5.56 | 0 | 0 | 7.69 | 0 | n.s. |
| IGHV3-23 | 23.53 | 0 | 3.53 | 8.33 | 6.56 | 5.56 | 12.50 | 0 | 7.69 | 0 | n.s. |
| IGHV3-30/33rn | 0 | 1.41 | 2.35 | 0 | 3.28 | 16.67 | 12.50 | 12.50 | 7.69 | 0 | n.s. |
| IGHV3-48 | 17.65 | 0 | 0 | 0 | 3.28 | 11.11 | 0 | 12.50 | 0 | 0 | n.s. |
| IGHV3-49 | 0 | 0 | 0 | 0 | 0 | 5.56 | 12.50 | 0 | 7.69 | 0 | n.s. |
| IGHV3-53/66 | 0 | 4.23 | 0 | 8.33 | 1.64 | 0 | 0 | 6.25 | 0 | 0 | n.s. |
| IGHV3-7 | 0 | 4.23 | 11.76 | 16.67 | 6.56 | 0 | 12.50 | 6.25 | 0 | 50.00 | n.s. |
| IGHV3-72 | 0 | 1.41 | 1.18 | 0 | 3.28 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-73 | 0 | 0 | 0 | 0 | 1.64 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-74 | 0 | 0 | 0 | 0 | 4.92 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-9 | 0 | 0 | 0 | 0 | 3.28 | 0 | 12.50 | 6.25 | 0 | 0 | n.s. |
| IGHV4-30-2 | 0 | 0 | 0 | 0 | 1.64 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV4-30-4/31 | 0 | 21.13 | 18.82 | 0 | 8.20 | 0 | 0 | 6.25 | 0 | 50.00 | n.s. |
| IGHV4-34 | 0 | 1.41 | 2.35 | 0 | 3.28 | 0 | 0 | 12.50 | 7.69 | 0 | n.s. |
| IGHV4-39 | 11.76 | 45.07 | 38.82 | 25.00 | 14.75 | 0 | 0 | 0 | 15.38 | 0 | 0.06 |
| IGHV4-4 | 0 | 4.23 | 2.35 | 8.33 | 3.28 | 0 | 0 | 0 | 0 | 0 | 0.07 |
| **IGHV4-59/61** | **11.76** | **9.86** | **15.29** | **25.00** | **13.11** | **11.11** | **12.50** | **0** | **0** | **0** | **0.02** |
| IGHV5-51 | 0 | 1.41 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV5-a | 5.88 | 1.41 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | n.s. |

Supplemental Table 4: Percent usage per IGHV germline segment amongst **IgG-VH clusters** (Figure 1, Table S2) of closely related or identical IgG-VH with at least one CSF clone in MS and OND patients. Comparison was made between MS and OND and p-values were calculated using a resampling-based permutation test. Only germline segments are represented which were found in at least one B cell cluster. IGHV4-59/61 is significantly overrepresented in MS B cell clusters, IGHV4-39 and IGHV4-4 reach marginal statistical significance; n.s., not significant ($p$>0.05).

| IGHV | MS-1 | MS-2 | MS-3 | MS4 | MS-5 | MS-6 | OND-1 | OND-2 | OND-3 | OND-4 | OND-5 | OND-6 | OND-7 | p |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| IGHV1-14 | 0 | 0 | 0 | 0 | 0 | 0.01 | 0 | 0.01 | 0.01 | 0 | 0 | 0 | 0 | n.s. |
| IGHV1-17 | 0 | 0 | 0.01 | 0 | 0 | 0 | 0 | 0 | 0.01 | 0 | 0 | 0 | 0 | n.s. |
| IGHV1-18 | 5.77 | 4.79 | 5.52 | 5.28 | 5.57 | 4.30 | 4.22 | 5.52 | 5.62 | 7.72 | 4.58 | 3.54 | 3.31 | n.s. |
| IGHV1-2 | 2.76 | 2.34 | 2.37 | 4.56 | 2.11 | 3.58 | 2.83 | 5.30 | 6.66 | 0.63 | 0.64 | 5.30 | 5.01 | n.s. |
| IGHV1-24 | 1.59 | 0.86 | 1.51 | 1.08 | 0.80 | 1.13 | 1.25 | 1.93 | 1.19 | 1.79 | 0.15 | 0.87 | 1.09 | n.s. |
| IGHV1-3 | 2.05 | 3.55 | 2.93 | 1.51 | 2.47 | 1.24 | 2.59 | 1.68 | 0 | 2.30 | 2.92 | 0 | 0.02 | n.s. |
| IGHV1-45 | 0 | 0 | 0 | 0.13 | 0 | 0 | 0 | 0 | 0 | 0 | 0.11 | 0 | 0 | n.s. |
| IGHV1-46 | 1.96 | 1.68 | 3.90 | 2.03 | 2.01 | 1.62 | 5.87 | 1.70 | 1.21 | 4.62 | 2.23 | 2.10 | 0.56 | n.s. |
| IGHV1-58 | 0.14 | 0.04 | 0.04 | 0.30 | 0.03 | 0.14 | 0.13 | 0.11 | 0.02 | 0.11 | 0.07 | 0 | 0 | n.s. |
| IGHV1-67 | 0 | 0 | 0 | 0 | 0 | 0 | 0.01 | 0 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV1-68 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0.01 | 0 | 0 | 0 | 0 | n.s. |
| IGHV1-69 | 11.80 | 4.73 | 1.55 | 4.72 | 4.93 | 6.50 | 5.88 | 3.91 | 11.20 | 4.48 | 4.67 | 4.27 | 7.53 | n.s. |
| IGHV1-8 | 0.07 | 0 | 1.56 | 0.64 | 1.85 | 2.05 | 0.35 | 0.65 | 1.37 | 0 | 1.44 | 2.46 | 1.41 | n.s. |
| IGHV1-c | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0.02 | 0.16 | n.s. |
| IGHV1-f | 0.43 | 0.80 | 0 | 0 | 0 | 1.05 | 0.01 | 0 | 0.50 | 0 | 0 | 0.02 | 2.50 | n.s. |
| IGHV2-10 | 0 | 0 | 0 | 0 | 0 | 0.01 | 0 | 0 | 0 | 0 | 0 | 0.20 | 0 | n.s. |
| IGHV2-26 | 0.17 | 0.12 | 0.07 | 0.16 | 0.16 | 0.23 | 0.16 | 0.22 | 0.12 | 1.17 | 0.29 | 0.02 | 0.35 | n.s. |
| IGHV2-5 | 2.58 | 1.97 | 4.11 | 1.89 | 2.15 | 2.35 | 3.34 | 3.31 | 1.60 | 3.50 | 1.39 | 1.42 | 0.10 | n.s. |
| IGHV2-70 | 0.75 | 0.56 | 0.35 | 0.63 | 0.18 | 0.43 | 0.39 | 0.78 | 0.59 | 0.70 | 0.11 | 0.16 | 0.45 | n.s. |
| IGHV3-11 | 1.11 | 1.43 | 1.98 | 1.99 | 1.39 | 2.23 | 2.37 | 2.06 | 2.16 | 0.15 | 0.11 | 1.23 | 2.25 | n.s. |
| IGHV3-13 | 0.24 | 0.40 | 0.44 | 0.77 | 0.15 | 0.44 | 0.29 | 0.41 | 0.24 | 0.13 | 0.11 | 1.09 | 0 | n.s. |
| IGHV3-15 | 2.15 | 3.32 | 1.86 | 1.62 | 1.87 | 1.79 | 3.43 | 1.50 | 1.44 | 1.40 | 1.42 | 2.86 | 2.24 | n.s. |
| IGHV3-20 | 0.45 | 0.58 | 0.64 | 0.28 | 0.18 | 0.26 | 0.47 | 0.30 | 0.03 | 0 | 0.73 | 0.04 | 0.12 | n.s. |
| IGHV3-21 | 1.54 | 3.48 | 3.59 | 2.61 | 4.49 | 2.33 | 2.50 | 2.92 | 2.56 | 3.28 | 2.33 | 2.79 | 2.12 | n.s. |
| **IGHV3-23** | **6.92** | **11.05** | **11.78** | **10.89** | **9.14** | **9.98** | **7.59** | **5.37** | **5.70** | **4.06** | **9.18** | **10.30** | **5.15** | **0.04** |
| IGHV3- | 12.21 | 13.51 | 4.18 | 10.14 | 9.45 | 9.65 | 8.66 | 12.97 | 11.33 | 15.72 | 8.10 | 14.13 | 9.89 | n.s. |
| IGHV3-35 | 0.01 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-41 | 0 | 0.01 | 0 | 0 | 0 | 0 | 0 | 0 | 0.05 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-43 | 0.41 | 0.14 | 0.66 | 0.60 | 0.33 | 0.27 | 0.30 | 0.15 | 0.36 | 0.52 | 0.67 | 0.20 | 0.87 | n.s. |
| IGHV3-47 | 0 | 0 | 0.02 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-48 | 2.62 | 1.82 | 2.84 | 1.89 | 4.50 | 4.56 | 2.59 | 3.49 | 1.33 | 2.42 | 2.79 | 3.90 | 3.47 | n.s. |
| IGHV3-49 | 1.14 | 1.11 | 1.10 | 0.74 | 0.44 | 1.93 | 0.98 | 1.03 | 0.45 | 0.92 | 1.62 | 1.16 | 1.02 | n.s. |
| IGHV3-53/66 | 4.47 | 2.59 | 3.24 | 3.23 | 2.35 | 0.79 | 1.79 | 2.92 | 1.92 | 2.25 | 3.98 | 2.95 | 2.10 | n.s. |
| IGHV3-64 | 0.50 | 1.53 | 0.54 | 0.58 | 0 | 0.38 | 2.23 | 0.84 | 0 | 0.40 | 0.49 | 0.27 | 1.06 | n.s. |
| IGHV3-65 | 0 | 0 | 0.02 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0.04 | 0 | 0 | n.s. |
| IGHV3-7 | 2.68 | 4.63 | 4.15 | 5.71 | 6.42 | 6.06 | 6.18 | 3.78 | 7.89 | 1.72 | 4.56 | 7.74 | 5.00 | n.s. |
| IGHV3-71 | 0 | 0.01 | 0 | 0 | 0 | 0 | 0 | 0.01 | 0 | 0 | 0 | 0 | 0 | n.s. |
| IGHV3-72 | 1.10 | 0.84 | 0.66 | 0.34 | 0.81 | 0.69 | 1.76 | 0.77 | 0.06 | 0.03 | 0.55 | 1.03 | 2.24 | n.s. |
| IGHV3-73 | 1.21 | 0.58 | 0.63 | 0.31 | 0.17 | 0.72 | 0.70 | 0.34 | 1.50 | 0.75 | 1.02 | 0.30 | 0.57 | n.s. |
| **IGHV3-74** | **3.51** | **3.31** | **5.02** | **5.01** | **3.40** | **3.24** | **3.43** | **2.65** | **1.23** | **3.07** | **2.10** | **1.12** | **2.32** | **0.02** |
| IGHV3-9 | 0.08 | 0 | 1.80 | 2.29 | 4.69 | 2.87 | 2.88 | 2.06 | 6.24 | 0 | 0.42 | 3.67 | 0.52 | n.s. |
| IGHV3-h | 0.20 | 0.13 | 0.04 | 0.14 | 0.04 | 0.41 | 0.29 | 0.41 | 0.02 | 0.21 | 0.38 | 0.16 | 0.09 | n.s. |
| IGHV4-28 | 0.02 | 0 | 0.07 | 0.05 | 0 | 0.01 | 0 | 0.01 | 0.01 | 0 | 0.07 | 0 | 0.09 | n.s. |
| IGHV4-30-2 | 0.75 | 0.29 | 0.01 | 0 | 1.26 | 0.59 | 0.04 | 0.06 | 0.62 | 1.98 | 1.71 | 0.30 | 0.12 | n.s. |
| IGHV4-30- | 4.89 | 3.07 | 1.15 | 1.69 | 2.85 | 3.78 | 1.31 | 2.34 | 4.85 | 2.72 | 0.49 | 5.07 | 0.40 | n.s. |
| IGHV4-34 | 4.34 | 2.71 | 5.45 | 5.78 | 2.94 | 2.06 | 2.43 | 6.33 | 3.25 | 3.60 | 7.81 | 3.59 | 3.19 | n.s. |
| IGHV4-39 | 3.31 | 8.19 | 6.38 | 6.90 | 6.05 | 6.58 | 3.12 | 3.84 | 6.74 | 5.37 | 8.61 | 4.48 | 8.34 | n.s. |
| IGHV4-4 | 1.43 | 2.18 | 1.85 | 2.09 | 3.05 | 1.75 | 1.73 | 2.26 | 0.83 | 2.00 | 1.24 | 1.71 | 1.35 | n.s. |
| IGHV4-55 | 0 | 0 | 0 | 0 | 0 | 0.02 | 0 | 0.01 | 0.02 | 0.01 | 0.02 | 0 | 0.02 | n.s. |
| IGHV4-59/61 | 6.01 | 5.97 | 7.04 | 6.79 | 8.65 | 4.96 | 6.21 | 8.33 | 5.80 | 10.14 | 10.96 | 3.22 | 5.92 | n.s. |
| IGHV4-b | 1.24 | | 0.01 | 0.01 | | 2.29 | 2.58 | 1.49 | 0.01 | 3.62 | 3.41 | 0.25 | 12.40 | n.s. |
| IGHV5-51 | 3.67 | 3.17 | 2.90 | 2.55 | 2.56 | 2.47 | 1.93 | 3.48 | 2.22 | 2.79 | 1.46 | 1.73 | 3.31 | n.s. |
| IGHV5-a | 0.69 | 1.50 | 1.98 | 0.57 | 0 | 0 | 0.34 | 0.83 | 0 | 2.15 | 1.00 | 0 | 0 | n.s. |
| IGHV6-1 | 1.02 | 0.97 | 1.77 | 1.53 | 0.53 | 0.79 | 0.51 | 0.47 | 0.99 | 1.56 | 1.88 | 4.34 | 1.35 | n.s. |
| IGHV7-4-1 | 0.03 | 0 | 2.30 | 0.01 | 0 | 1.46 | 4.31 | 1.48 | 0 | 0 | 2.13 | 0 | 0 | n.s. |

Supplemental Table 5: Percent usage per IGHV germline segment in the **PBMC** of MS and OND patients. Comparison was made between MS and OND and p-values were calculated using a resampling-based permutation test. Only germline segments are represented which were found in at least one PBMC sample. IGHV3-23 and IGHV3-74 appear to be significantly overrepresented among PBMC IgG-VH in MS; n.s., not significant ($p$>0.05).

**Supplemental Figures**

*Legends*

Supplemental Figure 1: **Comparison of sequence counts** shown in Table S1. Shown are numbers of IgG-VH sequences per disease group (MS or OND) after sequencing and initial quality control ("all reads"), numbers of sequences with identifiable IGHV, IHGJ, and CDR3 ("V, J, CDR3"), and numbers of non-redundant IgG-VH datasets ("non-red"; see Methods). Numbers of non-redundant CSF IgG-VH sequences were significantly higher in MS patients compared to OND (unpaired 2-tailed t-test).

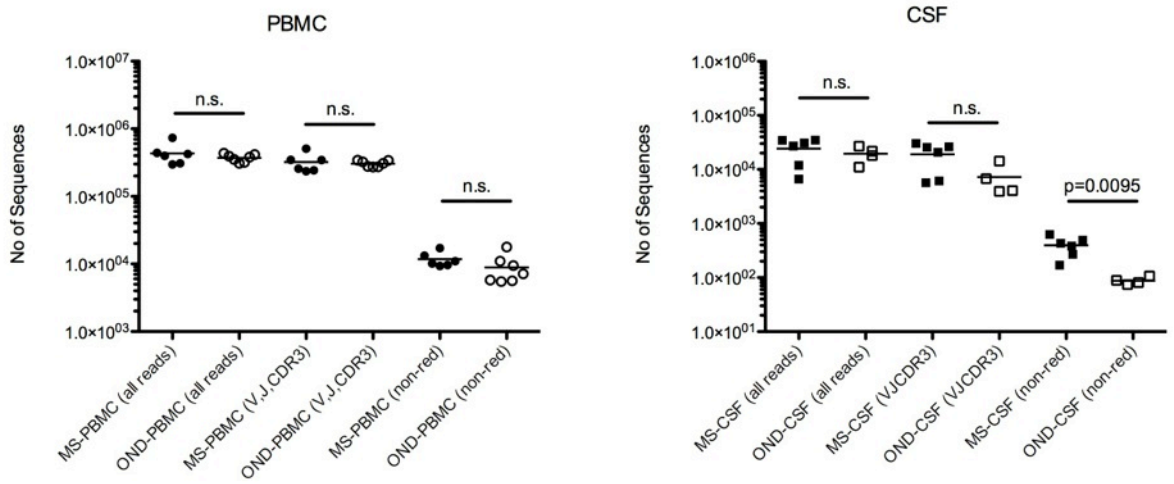
Supplemental Figure 2: **Representative multiple alignments of IgG-VH sequence clusters.** Shown are examples from patients MS-1 (Figure 1, cluster N), and MS-5 (Figure 1, cluster M). Alignments of translated amino acid sequences corresponding to the indicated clusters; sequences are labeled by unique identifier, sequencing ID, and compartment (i.e. CSF or PBMC). Closest IGHV germline segments and average similarity to germline are: MS-1 IGHV3-23, 94%; MS-5, IGHV4-39 90%.

Supplemental Figure 3: **Comparisons of IGHV usage profiles** in PBMC of MS and OND patients. Only IGHV are shown that were represented at >0.5% in the respective PBMC sample; in PB IGHV3-23 and IGHV3-74 were over-represented ($p$=0.04 and $p$=0.02, respectively) in MS patients compared with OND. Shown values are mean + SEM.

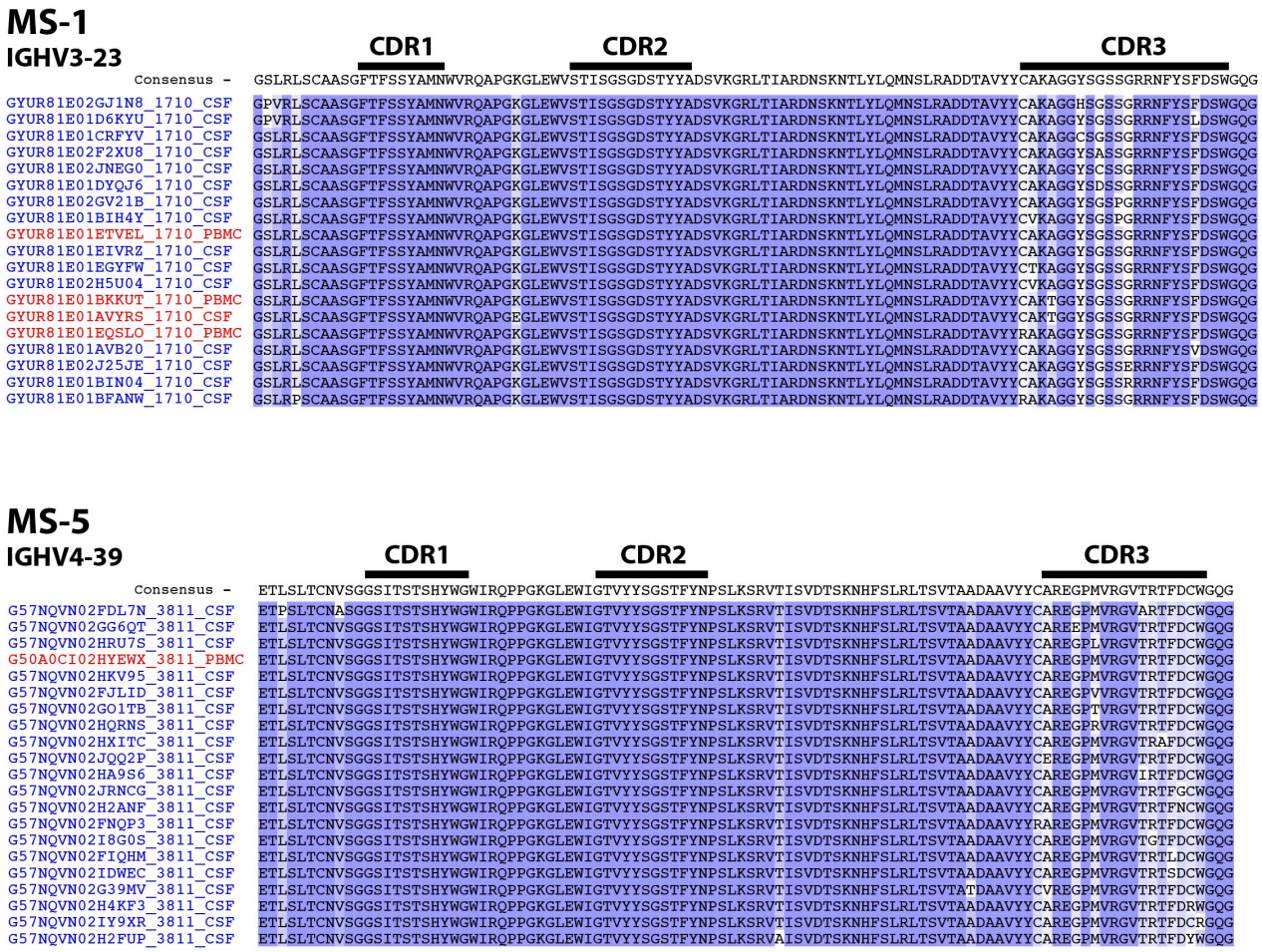
Supplemental Figure 4: **Distribution of CDR3 length (A) and IGHV mutations (B) in MS and OND IgG-VH networks**. IGHV mutations shown are nucleotide mutations. No significant differences could be observed between MS and OND (unpaired 2-tailed t-test).

Supplemental Figure 5: **Strategy of clustering of related B cells based on a Levenshtein distance of 1 in the H-CDR3.** Identical H-CDR3 are placed in individual nodes unless found in both compartments which is when they are placed in overlapping, differently colored nodes (blue=CSF, red=PB); H-CDR3 differing by a single aminoacid are directly connected by edges. Shown on the left is a resulting network and on the right the alignment of H-CDR3 sequences represented in this IgG-VH cluster.

*Figures*



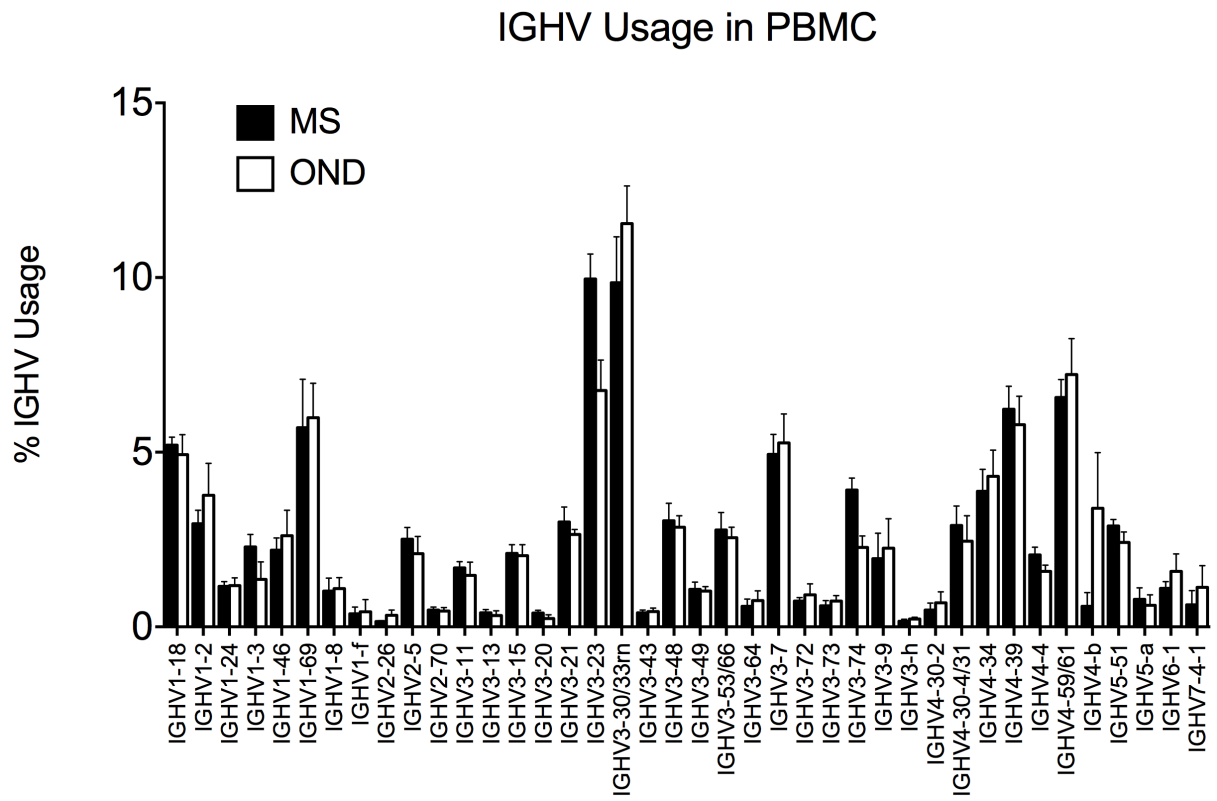

**Supplemental Figure 1**



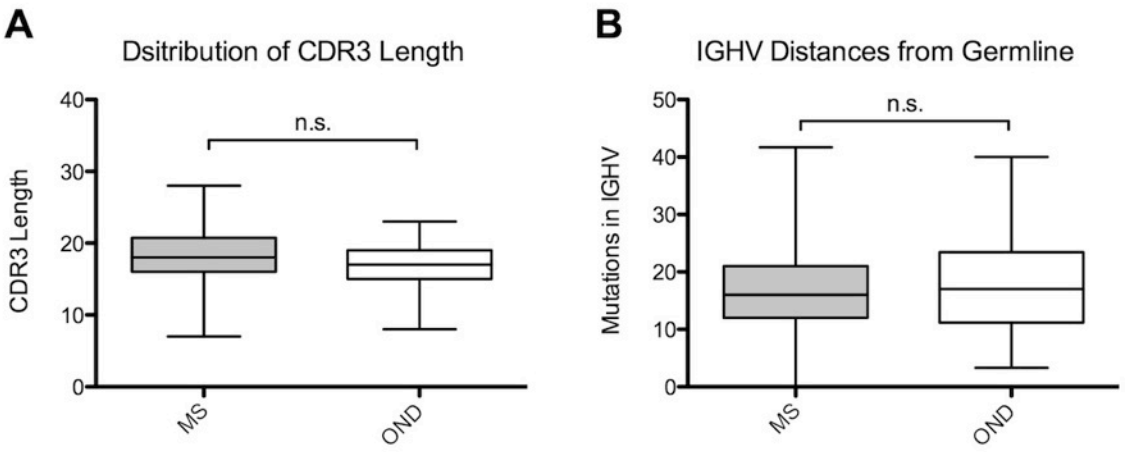




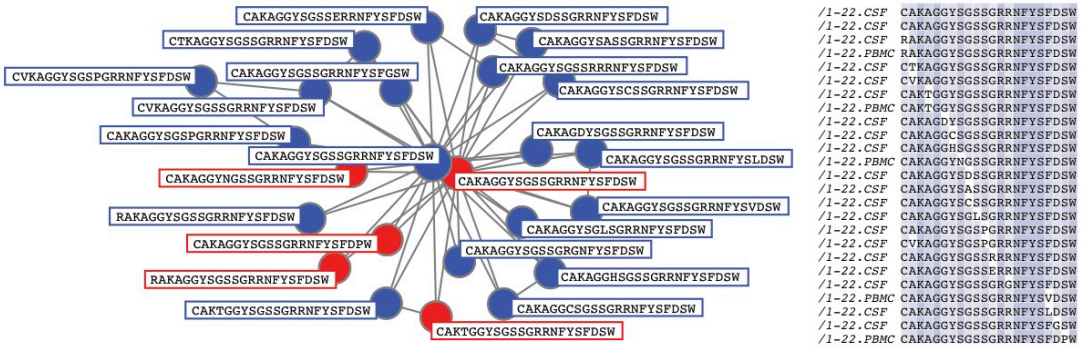
**Supplemental Figure 2**




**Supplemental Figure 3**




**Supplemental Figure 4**




**Supplemental Figure 5**