**Supporting Information**

**Using clonal expansion of mononuclear bone marrow-derived stem cells to discover mosaicism in a patient with neurofibromatosis type 2.**

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**Supplemental Table**

Summary of sequencing primers for *NF2*

**Supplemental Methods**

**Isolation of mononuclear cells (MNCs)**

MNCs were isolated from peripheral blood using Ficoll–Paque PREMIUM (Cytiva, Tokyo, Japan) and Leucosep (Greiner Bio-One, Kremsmünster, Austria) according to the manufacturer’s recommendation.

**DNA isolation and Sanger sequencing**

DNA was obtained from MNCs and frozen tumor tissue using the AllPrep DNA/RNA Mini Kit (Qiagen, Tokyo, Japan). Genomic DNA was amplified by polymerase chain reaction (PCR) using Quick Taq ® HS DyeMix (TOYOBO, Osaka, Japan). The oligonucleotide primers used for PCR was listed in Supplementary Table. Cycle sequencing was performed using the BigDye Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems, Foster City, CA, USA) using forward and reverse PCR primers as sequencing primers. Sequencing data were obtained using the Genetic Analyzer 3130 Avant (Applied Biosystems, Foster City, CA, USA).

**Amplicon sequencing**

The adapter sequences for next-generation sequencing were ligated to PCR amplicon of exon 2 of *NF2* using NEBNext Ultra II DNA Library Prep Kit for Illumina (New England BioLabs, Ipswich, MA). Sequencing analysis was conducted using MiSeq System (Illumina, San Diego, CA). These procedures were performed by Hokkaido System Science INC (Sapporo, Japan).

**Quantitative reverse transcriptase-polymerase chain reaction (qRT-PCR)**

RNA extraction, cDNA synthesis, and qRTPCR were performed as previously described [Hou *et al*., 2019]. The oligonucleotide primers used for PCR were as follows: the forward primer for RICTOR [Vandamme *et al*., 2016], ACAGCTCACGGTTGTAGGTTGC; reverse primer for RICTOR [Vandamme *et al.,* 2016], TCTAAGTAGCCTTGCCCATCCTC; forward primer for RAPTOR [Vandamme *et al*., 2016], AGCTGGAGGATGAAGGATCGGATG; reverse primer for RAPTOR [Vandamme *et al*., 2016], AGGGTCCACACCAACATTCAGG; forward primer for Rac1, GTTGGAGAAACGTACGGTAAGG; reverse primer for Rac1, GCACCTCAGGATACCACTTTG; forward primer for PAK1 [Zhou *et al.,* 2013], AAGACATCCAACAGCCAGAA; reverse primer for PAK1 [Zhou *et al.,* 2013], TGTAGCCACGTCCCGAGT; forward primer for MST1 [Chang *et al.*, 2019], AGACCTCCAGGAGATAATCAAAGA; reverse primer for MST1 [Chang *et al*., 2019], AGATACAGAACCAGCCCCACA; forward primer for YAP1 [Chang *et al*., 2019], ACCCACAGCTCAGCATCTTCG; reverse primer for YAP1 [Chang *et al.*, 2019], TGGCTTGTTCCCATCCATCAG; forward primer for b-actin [Hou *et al.*, 2019], GTGAAGGTGACAGCAGTCGGTT; reverse primer for b-actin [Hou *et al.,* 2019], GAAGTGGGGTGGCTTTTAGGAT.

**Statistical analysis**

Statistical analysis with unpaired t-test was performed using Graph-Pad Prism 8 (GraphPad Software, San Diego, CA) to compare the two groups with NF2 wild-type and NF2 mutant iPSC. *p* < 0.05 was considered statistically significant.

**References**

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