Supplementary Figure 1. Analysis of the population of CD11b+, CD206+/CD11b+ and CD86+/CD11b+ cells in cultured bone marrow-derived mononuclear cells by method A, B or C. BM-MNCs were cultured by method A or B or C and collected. FACS analysis were performed to evaluate their population of CD11b+, CD206+/CD11b+ and CD86+/CD11b+ cells in each method. In all three patterns of CD antigen expression, significant differences were not shown among those methods. Error bars = means + SD. n.s.; not significant.
Supplementary Figure 2. Flow cytometry cell analysis of the population of CD45+ cells in BM-iMG cells induced by method C. The percentage of CD45+ cells in BM-iMG cells from wild type mouse is shown. Error bar = mean + SD.
Supplementary Figure 3. Basic evaluation of cell viability in NCS-34 cells by WST-8 assay, and of cell toxicity with NOC18 as NO-donor. (A) NCS-34 cells were seeded at the concentration of 0.6-3.0 x 10^4 cells per single well of 96-well plate, and their viability were measured by WST-8 assay. A450 optical density were measured by spectrometry for evaluation of their viability. Each A450 optical density was withdrawn the basal value in media only with no cell. Error bars = means ± SD. A450; Absorbance of 450 nm. WST; water soluble tetrazolium salts. Coefficient of determination : R²=0.9887. (B) Cell viability of NSC-34 cells after stimulation of NOC-18. NCS-34 cells were seeded and NOC-18 was added by 0-1600 µm concentration. After 24hr or 48hr, cell viability were evaluated by WST-8 assay. NOC-1 showed cell toxicity in a dose dependent manner. Bars = means ± SD.
Supplementary Figure 4. Analysis of the character of bone marrow-derived mononuclear cells (BM-MNCs) in wild type (WT) and SOD1-tg mouse (SOD1). (A) Isolated total number of nucleated bone marrow cells (BM cells) and BM-MNCs in each WT or SOD1 mouse are shown. (B) The percentage of BM-MNCs are shown in whole nucleated BM cells at WT and SOD1-tg groups. (C) The proliferation ratio to BM-iMG cells from BM-MNCs by method C are shown in WT and SOD1 mice. The ratio in day7 are standardised by that of day0 in each group. Error bars = means ± SD. n.s.; not significant. **p < 0.01.
Supplementary Figure 5. Flow cytometry cell analysis of the population of CD11b+, CD206+/CD11b+ and CD86+/CD11b+ cells in BM-iMG cells induced by method C. (A-C) The percentage of CD11b+ (A), CD206+/CD11b+ (B) and CD86+/CD11b+ (C) cells in BM-iMG cells from wild type (WT) and SOD1-tg mouse (SOD1) are shown. Error bars = means + SD. n.s.; not significant. **p<0.01.
Supplementary Figure 6. Analysis of the expression of mRNA in BM-iMG cells and their neuroprotective effects to cell viabilities of motor neurons. (A) The expression ratio of Arg1 and Nos2 mRNA in BM-iMG cells from wild type (WT) and SOD1-tg mouse (SOD1). (B) Cell viability of NSC-34 cells under cell injury by NOC-18 are shown with or without treatment of conditioned medium prepared by the supernatant in the culture of BM-iMG cells from WT or SOD1. Error bars = means + SD. n.s.; not significant. **p < 0.01.
Supplementary Figure 7. Time-course data plot of each individual mouse in body weight (A) and in rota-rod test (B) during in vivo experiments. Blue lines show the data of each mouse in control (CTL) group. And red lines show the data of each mouse in bone marrow-derived inducible microglia-like cells (BM-iMG) group.