

Supplementary Results

Effects of gene therapy and enzyme replacement therapy on the behavioral abnormalities of mucopolysaccharidosis VI mice

Both gene therapy and enzyme replacement therapy (ERT) similarly improved the abnormalities observed in (1) the rotarod task (Supplementary Fig. S2a); (2) the hanging wire task (Supplementary Fig. S2b); and (3) the first 5 min testing of the open field (Supplementary Fig. S2d). Indeed in each of these instances, MPS VI animals treated with adeno-associated viral (AAV) vectors did not significantly differ from normal controls. To test whether the two treatments were associated with animal distress, we analyzed also the exploratory patterns (Supplementary Fig. S2c and e). An altered exploratory pattern/behavior in the open field is commonly reported in the literature as a sign of anxiety or distress (Li *et al.*, 2012). A qualitative analysis of the exploratory patterns

clearly shows that affected (AF) mice, differently from normal (NR) controls, tend to spend most of their exploratory time in the periphery of the arena (Supplementary Fig. S2c and e). AAV treatment ameliorates not only the path length, but also the exploratory pattern, while ERT-treated mice, even if more active than AF controls, tend to spend most of their time in the periphery of the arena (Supplementary Fig. S2d). The exploratory behavior was quantified as percentage of time spent in the central part of the arena that indicates an almost significant effect of groups (Supplementary Fig. S2e). Although this effect was not fully significant, we performed a pairwise comparison to assess whether there were statistical significant differences between the different groups. We found that only AF and ERT significantly differed from the NR group. These results suggest that although both therapies are comparable in rescuing the motor deficits in mucopolysaccharidosis VI mice, gene therapy might be additionally associated with reduced signs of distress compared with ERT.