Fingolimod: a potential disease modifier in a mouse model of amyotrophic lateral sclerosis

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Amyotrophic lateral sclerosis (ALS) is actually considered a multi-factorial disease, partially justifying the failure to develop effective therapeutic approaches (Musarò 2013); in fact, despite the several phase II and phase III clinical trials performed so far, Riluzole remains the only available drug for the treatment of ALS with only modest effects. A growing body of evidence suggests that, besides many other mechanisms, malfunction of the immune system can actively influence disease progression in animal models and in ALS patients (Hovden et al. 2013).
Fingolimod (FTY720), the first approved oral therapy for multiple sclerosis, primarily acts as an immune-modulator preventing lymphocytes from moving out of the lymphoid organs and inhibiting autoreactive lymphocytes from infiltrating the central nervous system. Interestingly, FTY720 also exerts multiple effects in the CNS directly acting on resident cells, which indicates a potentially broader spectrum of activity in neurodegenerative diseases.
The present study was designed to determine whether chronic treatment with Fingolimod is able to extend the survival and/or to improve the phenotype of SOD1G93A ALS mice.
26 transgenic mice expressing high copy number of G93A mutant form of human SOD1 (B6.Cg-Tg(SOD1*G93A) and 10 age-matched non-transgenic (WT) littermates mice were used.
As the majority of ALS patients are sporadic, and thus treatment can only be started at the time of diagnosis, SOD1G93A mice were treated with Fingolimod (0.1 mg/kg i.p., 3 times a week) or vehicle starting from the very onset of symptoms (i.e. from around 17 weeks of age) until end-stage of disease. WT mice were treated with Fingolimod. A detailed assessment of motor behaviour, body weight and survival was performed in WT as well as in vehicle- and drug-treated SOD1G93A mice. Animals general conditions were evaluated weekly using ALS TDI neurological score system. Finally, the expression of genes related to neuroinflammation were analyzed by Real Time-PCR in homogenates from the cortex and spinal cords of end-stage animals.
As expected, SOD1G93A mice showed a gradual decrease in their body weight. A significant worsening of motor symptoms, tested using an acceleration rotarod device, was observed after 16 weeks of life in mutant mice. Although Fingolimod did not affect motor performance, it significantly improved the survival of SOD1G93A mice (Logrank Test $p<0.01$) and delayed neurological deficits of mutant mice compared to vehicle group throughout the trial. The neurological score was significantly affected by treatment [two-way ANOVA, $P<0.001$], time [$P<0.001$] but not by their interaction; at 19 and 20 weeks of age, the number of disease-free animals was significantly higher in Fingolimod than in vehicle-treated animals (Fisher's $p=0.001$).
In SNC tissue of moribund animal chronically treated whit Fingolimod, we found a significant increase in the expression of FoxP3 (a marker of regulatory T cell, Treg) and a reduction in beta-integrin CD11b (marker of activated microglia) with respect to veh-treated mice; these data, supported by molecular analysis that showed in the same tissue an increased level of arginase-1 and IL-10 (markers of the protective M2 microglia phenotype) and a decrease of iNOS and IL-1beta (M1 inflammatory phenotype), suggest that Fingolimod could modulate pathogenic activated microglia to produce relevant pro-inflammatory cytokines and, at the same time, recover the homeostasis of cells with regulatory function (Treg) contributing to restore a balance between T cell populations affected in ALS pathology.
In conclusion, our data strongly support Fingolimod as new therapeutic approach for ALS, since it has the potential to impact simultaneously on different pathogenic mechanisms of the disease, such as microglial activation and innate immunity.

Musarò A (2013), FEBS J. 280:4315-22.
Hovden et al. (2013), Acta Neurol Scand. 128:287–296.