durations in the animals subjected to multiple THEM transplantation were smaller (0.416s ± 0.058s) compare to that of the control animals (0.450s ± 0.077s). The healthy animals had a statistically shorter electromyographic anterior tibial muscle activation duration with respect to all the other experimental groups (0.161s ± 0.020s) (Figure 6). These data indicated that the treatment with THEM restores the normal excitability of the flexor motor neurons, although it has no effect in terms of re-innervation.

THEM promote the regeneration of severe SCI mice spinal cord tissue

Traumatic events which occur at the level of the spinal cord cause mechanical damage and consequent tissue degeneration. These events include: degradation of the cell membrane of axons, degeneration of the blood-brain barrier, demyelination, migration of immune cells, and death of spinal parenchymal cells [2, 5]. To study how THEM transplantation contributes to tissue regeneration of the spinal cord, the distribution of cells in the spinal parenchyma at 31 dpi and the histological features related to cyst formation, glial scar, number of neurons and in particular of cholinergic neurons (motor neurons), myelin content, activation of the immune response, extracellular matrix and tissue vascularization were first studied.

Transplanted THEM distribution in spinal cord tissue

In order to assess the persistence of the transplanted cells, their viability and location, the distribution of the transplanted cells at the end of the experiment (31 dpi) was analyzed. The number of THEMs and M2s was quantified in spinal cord cross-sections following severe spinal cord injury and multiple transplantation representing a 0.6 cm long spinal region (9 spinal cord/animal cross-sections) centred at the site of the spinal injury. For the analysis, the absolute number of fluorescent red cells (dT-Tomato; THEM or M2) co-localizing with the nuclear marker 4',6-diamino-2-phenylindole (DAPI) was considered. The distribution of the THEMs was analyzed based on their different location in the spinal tissue (parenchyma, cyst area, dorsal/ventral meninges). As shown in Figure 7A-B, the THEMs and M2 macrophages persist in the spinal parenchyma even in the days following transplantation, and their presence is detected not only in the section of the spinal lesion but also in the neighboring sections. Therefore, the diffusion of the transplanted cells (THEM and M2 macrophages) is not only radial with respect to the transplantation site, but also longitudinal in the rostral and caudal direction. The percentage values were normalized on the total THEM or M2 cells present in the spinal cord cross-sections representing a marrow region equal to 0.6 cm in length (9 spinal cord/animal cross-sections). In the section corresponding to spinal trauma, although they are concentrated in the cyst area (53.54% ± 10.40% THEM, n=5; 53.10% ± 10.95% M2, n=3), a significant percentage is widespread in the surrounding parenchyma (46.46% ± 10.40% THEM, n=5; 46.90% ± 10.95% M2, n=3) (Figure 7 C). The percentage values were normalized over the total THEM or M2 cells in the spinal trauma