# Unexpectedly, mometasone furoate demonstrates more therapeutic effects than methylprednisolone and dexamathasone in the chronic management of spinal cord injury in mice

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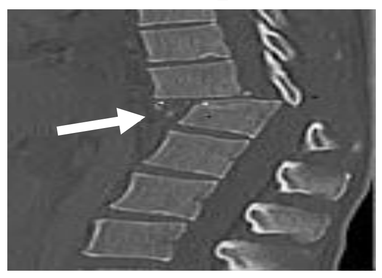
## *1.Introduction*

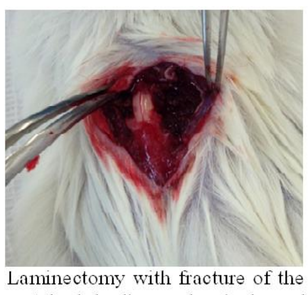
*Traumatic spinal cord injury (SCI) represents one of the most disabiling injuries of the human body causing temporary or permanent sensory and/or motor system deficit. Steroidal inflammatory drugs are the first line choice of treatment; however, SCI still remains a very complex medical and psychological challenge, with no curarative therapy available. The aim of the present study was to compare the current conventional therapy adopted in the management of SCI, represented by methylprednisolone sodium succinate (MPSS) in respect to other glucocorticoids (GCs), namely dexamethasone (Dex) and mometasone furoate (MF) in a mouse model of experimental spinal cord compression injury.*



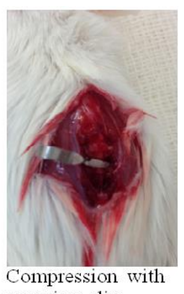
## *2.Materials and Methods*

*Animals: Male adult CD1 mice (25-30g weight) were housed in individually ventilated cages with food and water ad libitum.*

*SCI was induced in mice according to a standardized model firstly described by Rivlin and Tator (1). A longitudinal incision was made on the midline of the back, exposing the paravertebral muscles and T5-T8 vertebrae via laminectomy. SCI was produced by spinal cord compression at T6-T7 level using an aneurysm clip (see figure above left) with a closing force of 24g for 1 min. MPSS (6mg/Kg, i.p.), Dex (1mg/Kg, i.p.) and MF (0.1mg/Kg, i.p.) were administered 30 min after trauma induction and daily until sacrifice (eighth day). Spinal cord tissue was collected to perform immunohistochemical and western blot analyses.*



Laminectomy with fracture of the vertebral bodies and spinal cord exposition (T5-T8)



Compression with aneurism clip

## *3.Results*

*Achieved data demonstrate that SCI induces tissue damage, cellular infiltration (Fig.1), astrocyte activation (GFAP immuno staining [Fig.2]), iNOS expression (Fig.3), Antiapoptotic Bcl-2 expression (Fig.4), PARP-1 activation (Fig.5), and Proapoptotic Bax expression (Fig.6) in injured tissue. All the three GCs have demonstrated the capability to modulate inflammatory, oxidative and apoptotic pathways, but MF has demonstrated the best efficacy, while Dex and MPSS have shown alternative potency with a different protection degree.*

## *4.Figures*

***Fig.1***

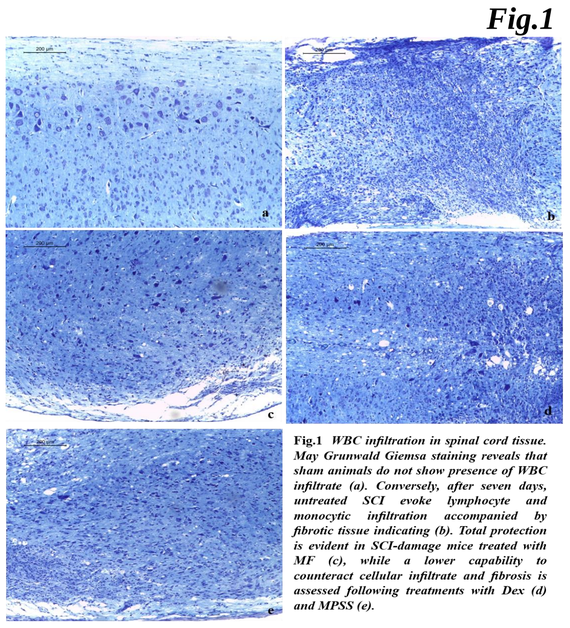


**Fig.1** *WBC infiltration in spinal cord tissue. May Grunwald Giemsa staining reveals that sham animals do not show presence of WBC infiltrate (a). Conversely, after seven days, untreated SCI evoke lymphocyte and monocytic infiltration accompanied by fibrotic tissue indicating (b). Total protection is evident in SCI-damage mice treated with MF (c), while a lower capability to counteract cellular infiltrate and fibrosis is assessed following treatments with Dex (d) and MPSS (e).*

***Fig.2***

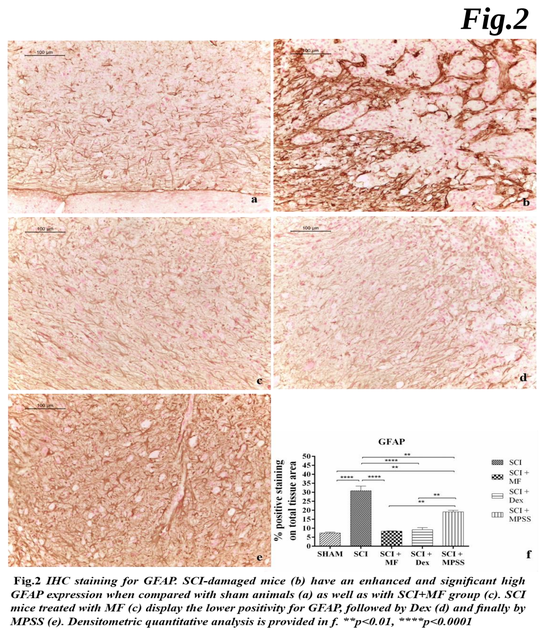



**Fig.2** *IHC staining for GFAP. SCI-damaged mice (b) have an enhanced and significant high GFAP expression when compared with sham animals (a) as well as with SCI+MF group (c). SCI mice treated with MF (c) display the lower positivity for GFAP, followed by Dex (d) and finally by MPSS (e). Densitometric quantitative analysis is provided in f. **p<0.01, ****p<0.0001*

***Fig.3***

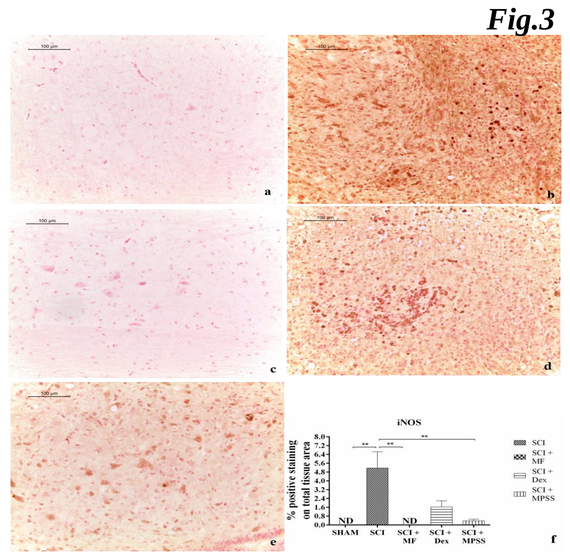



**Fig.3** *IHC staining for iNOS. Sham mice resulted negative for iNOS staining (a). Analyzing sections sampled by SCI mice, we demonstrated that untreated animals display a significant iNOS increasing (b), while there is a differentially modulated iNOS expression by GC treatments. In particular, the grading scale of protection vs untreated SCI mice is given by MF (c) >MPSS (e) >Dex (d). Densitometric quantitative analysis is provided in f. **p<0.01*

***Fig.4***

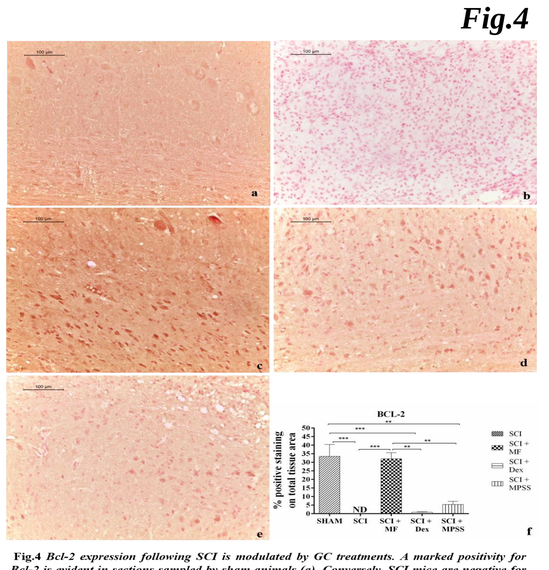



**Fig.4** *Bcl-2 expression following SCI is modulated by GC treatments. A marked positivity for Bcl-2 is evident in sections sampled by sham animals (a). Conversely, SCI mice are negative for Bcl-2(b),while MF treatment completely and significantly protect the tissue by Bcl2 downregulation (c). Dex(d) and MPSS(e) possess a lower,although still significant, power to protect against Bcl-2 degradation. Densitometric quantitative analysis is provided in f. **p<0.01, ***p<0.001, ND=Not Detectable*

***Fig.5***

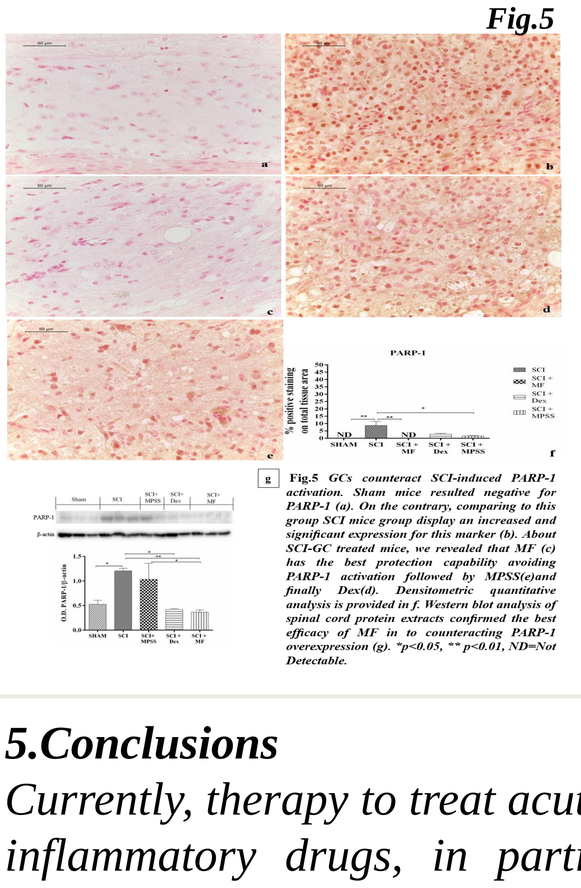



**Fig.5** *GCs counteract SCI-induced PARP-1 activation. Sham mice resulted negative for PARP-1 (a). On the contrary, comparing to this group SCI mice group display an increased and significant expression for this marker (b). About SCI-GC treated mice, we revealed that MF (c) has the best protection capability avoiding PARP-1 activation followed by MPSS(e)and finally Dex(d). Densitometric quantitative analysis is provided in f. Western blot analysis of spinal cord protein extracts confirmed the best efficacy of MF in to counteracting PARP-1 overexpression (g). *p<0.05, ** p<0.01, ND=Not Detectable.*

***Fig.6***

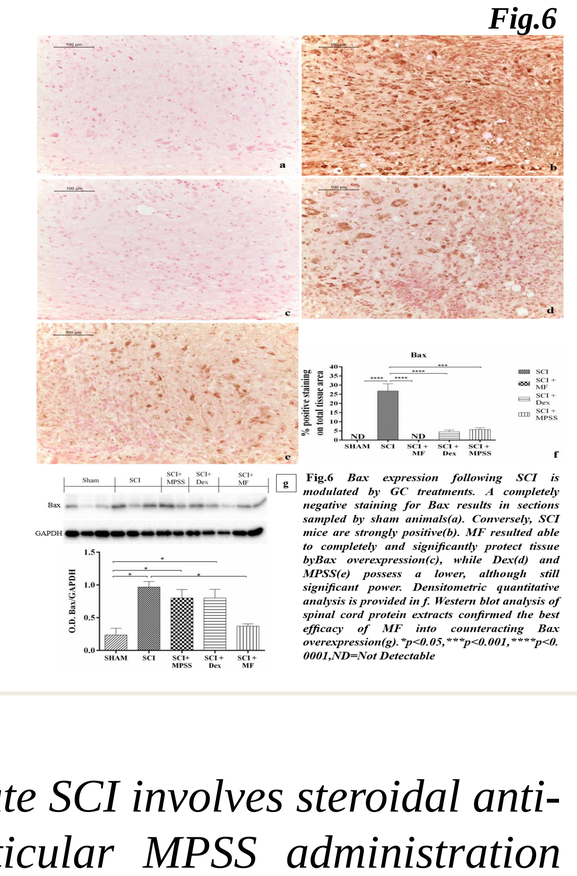



**Fig.6** *Bax expression following SCI is modulated by GC treatments. A completely negative staining for Bax results in sections sampled by sham animals(a). Conversely, SCI mice are strongly positive(b). MF resulted able to completely and significantly protect tissue byBax overexpression(c), while Dex(d) and MPSS(e) possess a lower, although still significant power. Densitometric quantitative analysis is provided in f. Western blot analysis of spinal cord protein extracts confirmed the best efficacy of MF into counteracting Bax overexpression(g).*p<0.05,***p<0.001,****p<0.0001,ND=Not Detectable*

## *5.Conclusions*

*Currently, therapy to treat acute SCI involves steroidal anti-inflammatory drugs, in particular MPSS administration within 3-8 hours from the trauma (2). Our results show that MF is the best candidate for post-traumatic chronic treatment since it counteracts the spreading of periwound secondary damage and ameliorates the spinal cord injury in a mice model with higher magnitude than Dex and MPSS.*

## *6.References*

*1.Rivlin AS, Tator CH. Effect of duration of acute spinal cord compression in a new acute cord injury model in the rat. Surgical neurology. 1978;10:38-43.*
*2.Bracken MB, Shepard MJ, Holford TR et al. Administration of methylprednisolone for 24 or 48 hours or tirilazad mesylate for 48 hours in the treatment of acute spinal cord injury. Results of the Third National Acute Spinal Cord Injury Randomized Controlled Trial. National Acute Spinal Cord Injury Study. JAMA 1997; 277:159.*

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