Effects of the Immunostimulant GcMAF in Cerebral Ischemia

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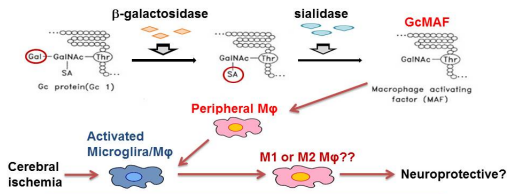
Background

Post-ischemic inflammation is an essential step in the evolution of ischemic brain damage, and it is also associated with the repairment of injured brain tissue. Macrophages are the main inflammatory effector among the various immune cells infiltrated from blood. The group-specific component (Gc) protein, known as vitamin D-binding protein or Gc globulin, is a 53-Kda glycoprotein.

Inflammation results in the hydrolysis of terminal galactose and sialic acid and this is mediated by β-galactosidase and sialidase to produce the Gc protein-derived macrophage activating factor (GcMAF). GcMAF is an unique molecule and stimulates macrophage phagocytic activity but dose not stimulate the release of possibly harmful cytokines such as TNFα.

Macrophages have diverse phenotypes and engage different functional programs. M1 macrophages typically release destructive inflammatory mediators. In contrast, M2 macrophages possess neuroprotective properties. In cerebral ischemia, the relationship between GcMAF, M1, and M2 macrophages remains to be elucidated.



Purposes

To verify the role of GcMAF in the early- and the delayed treatment after cerebral ischemia

Hypothesis

Treatment with GcMAF in acute ischemic period exacerbates brain damage, while it may facilitate recovery in the late phase.

Methods

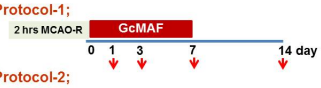
Animals;

7-week-old Wistar male rat

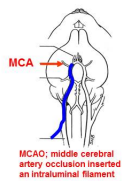
Groups

1. MCAO-R/vehicle control (VC)
2. MCAO-R/40ng/kg/day, ip GcMAF (GcMAF)

Protocol-1;



Protocol-2;

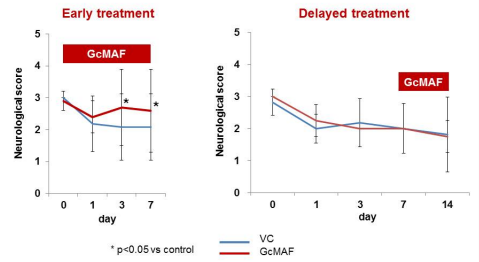


Assessment;

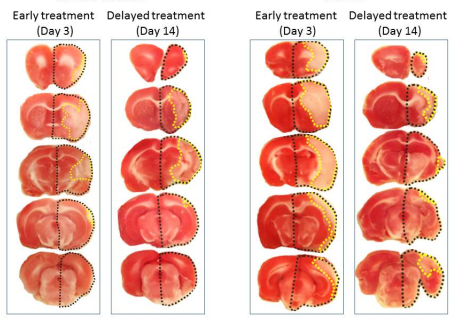
- neurological score
- infarct size
- Immunohistochemistry
- Quntitative real time PCR

Results

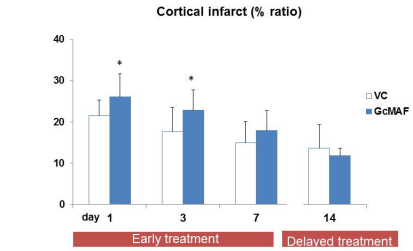
Postischemic neurological deficit is exacerbated by the early- but not the delayed treatment with GcMAF after ischemia



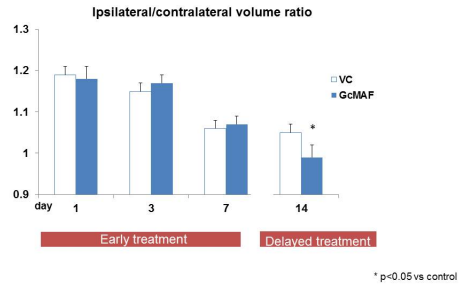
Representative infarct area in the early- and the delayed treatment with GcMAF and the vehicle control



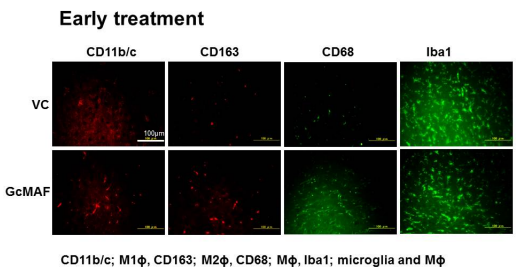
Cortical Infarct area is larger in the early treatment and smaller in the delayed treatment with GcMAF than vehicle control



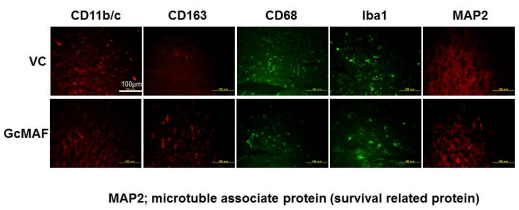
Ratio of ipsilateral/contralateral volume is reduced in the delayed treatment with GcMAF



GcMAF in the early treatment after ischemia increases M1 but not M2 macrophages in the cortex with infiltration of peripheral macrophages



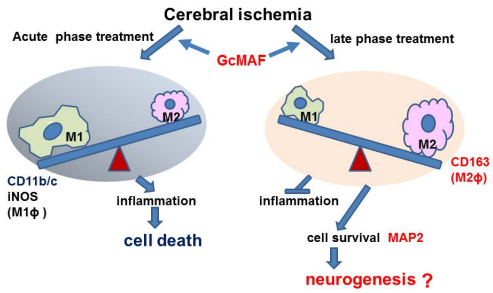
GcMAF in the delayed treatment after ischemia increases the cortical M2 but not M1 macrophages and MAP2 positive cells



Discussion

We newly demonstrated that GcMAF in the early treatment after ischemia increases M1- more than M2 macrophages and exacerbated brain damage. Interestingly, in the delayed treatment GcMAF reduces M1 macrophages and improved imbalance of M1/M2 macrophages. This resulted in the reduction of the brain damage, suggesting that the improvement of the imbalance of M1/M2 macrophages by GcMAF may contribute to the postischemic neuroprotection. Microglia/macrophages respond dynamically to ischemic injury. Experiencing an early "healthy M2 phenotype", followed by a transition to a "sick" M1phenotype are activated in the ischemic brain. It is suggested that adjusting the balance between beneficial and detrimental microglia/macrophage responses is important. To verify whether the improvement of the imbalance of M1/M2 macrophages in the late phase are associated with the promotion of neurogenesis, further studies are on going.

Schematic diagram of the role of GcMAF in cerebral ischemia



Conclusion

GcMAF treatment may exert bidirectional effects in a time-dependent manner after cerebral ischemia.

Further studies are required to confirm whether GcMAF treatment in the late phase plays an essential role to promote regeneration of central neurovascular unit after stroke.

References

1. Hu X, Li P, Guo Y, Wang H, Leak RK, Chen S, Gao Y, Chen J. Microglia/macrophage polarization dynamics reveal novel mechanism of injury expansion after focal cerebral ischemia. Stroke. 2012;43:3063-70
2. Inácio AR, Ruscher K, Leng L, Bucala R, Deierborg T. Macrophage migration inhibitory factor promotes cell death and aggravates neurologic deficits after experimental stroke. J Cereb Blood Flow Metab. 2011;31:1093-106