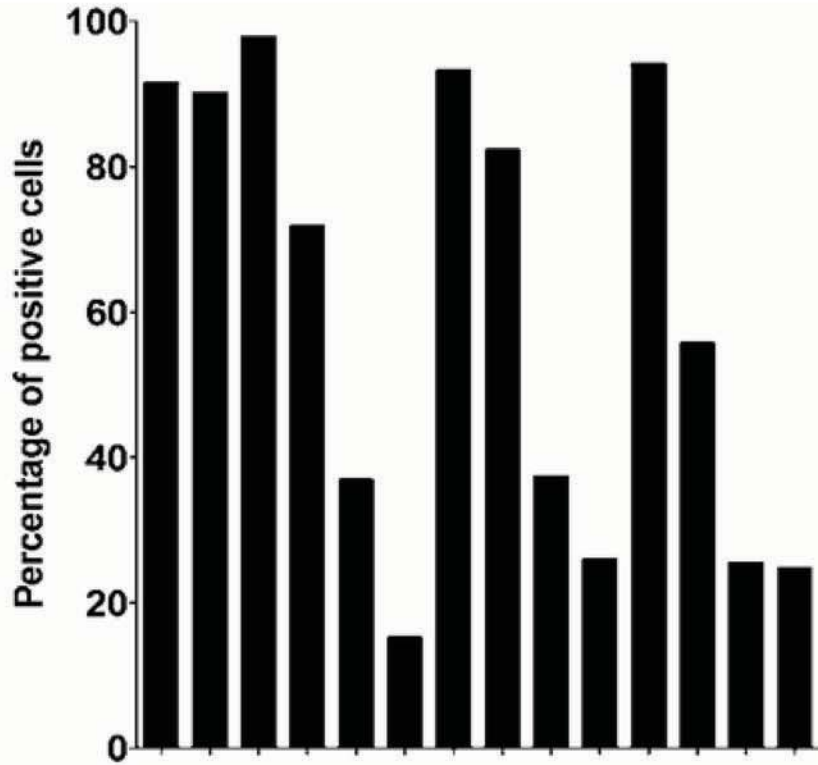


A

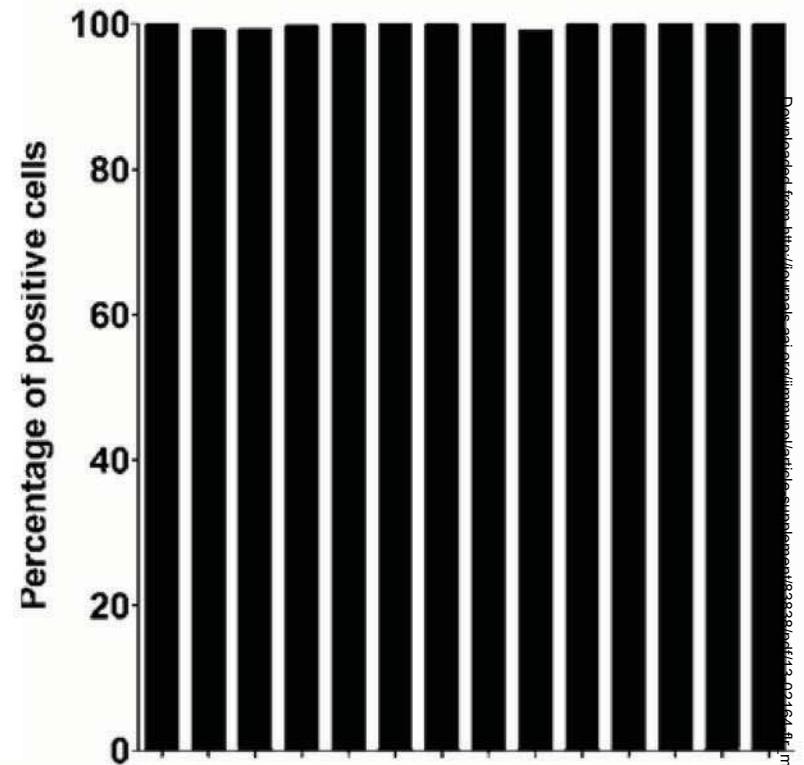
Cell membrane TGFβ2



IFNγ+TNFα	-	+	+	+	+	+	+	+	+	+	+	+	+	+
TGFβ1	-	-	0.1	1	10	100	-	-	-	-	-	-	-	-
TGFβ2	-	-	-	-	-	-	0.1	1	10	100	-	-	-	-
TGFβ3	-	-	-	-	-	-	-	-	-	-	0.1	1	10	100

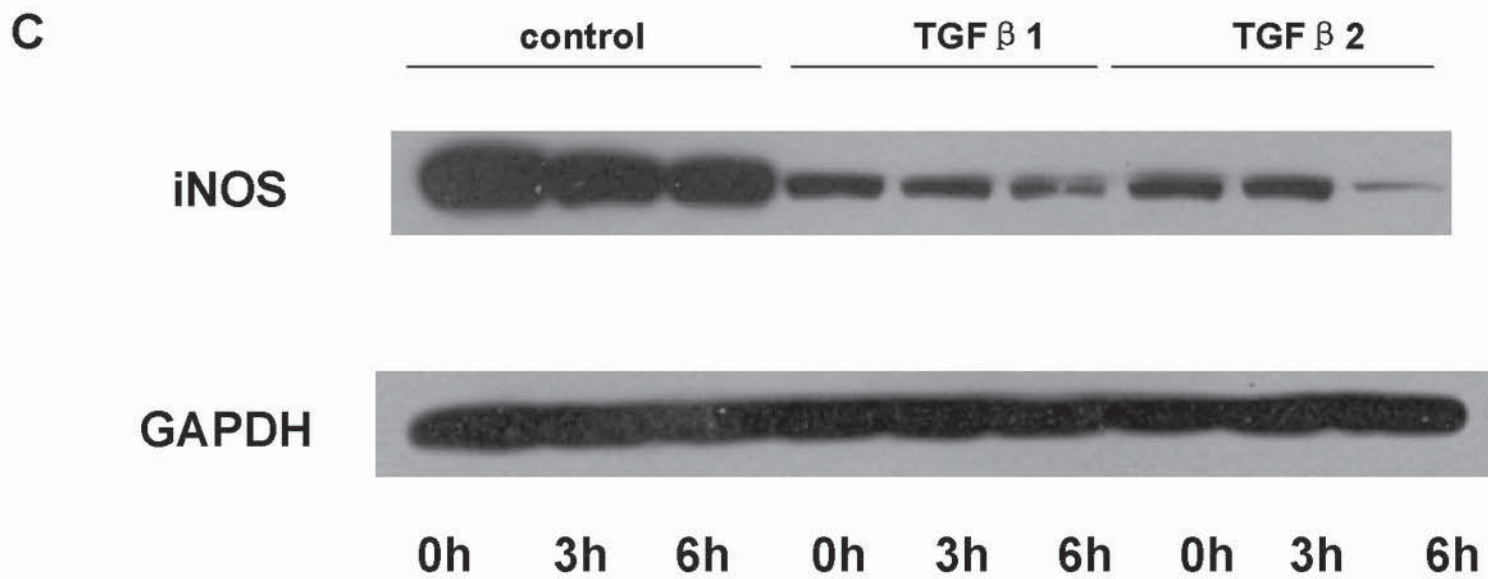
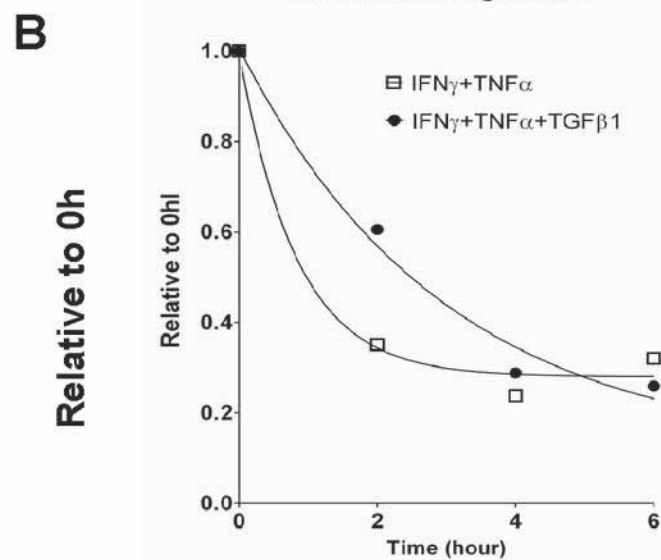
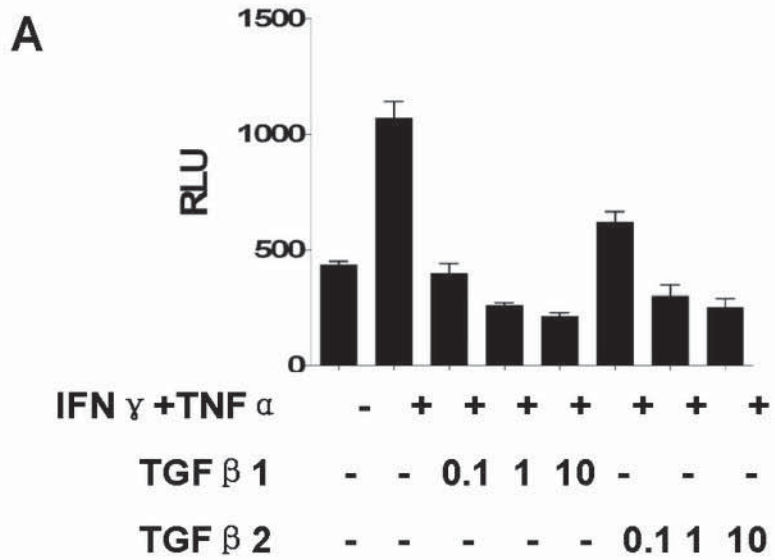
B

Intracellular TGFβ2



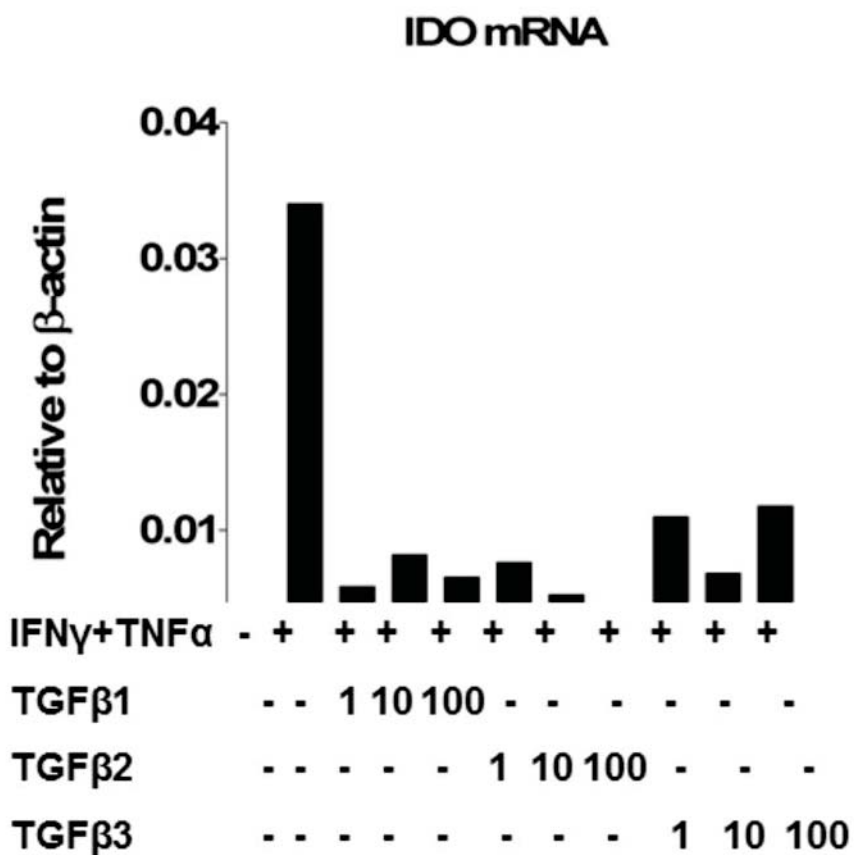
IFNγ+TNFα	-	+	+	+	+	+	+	+	+	+	+	+	+	+		
TGFβ1	-	-	0.1	1	10	100	-	-	-	-	-	-	-	-		
TGFβ2	-	-	-	-	-	-	0.1	1	10	100	-	-	-	-		
TGFβ3	-	-	-	-	-	-	-	-	-	-	-	-	0.1	1	10	100

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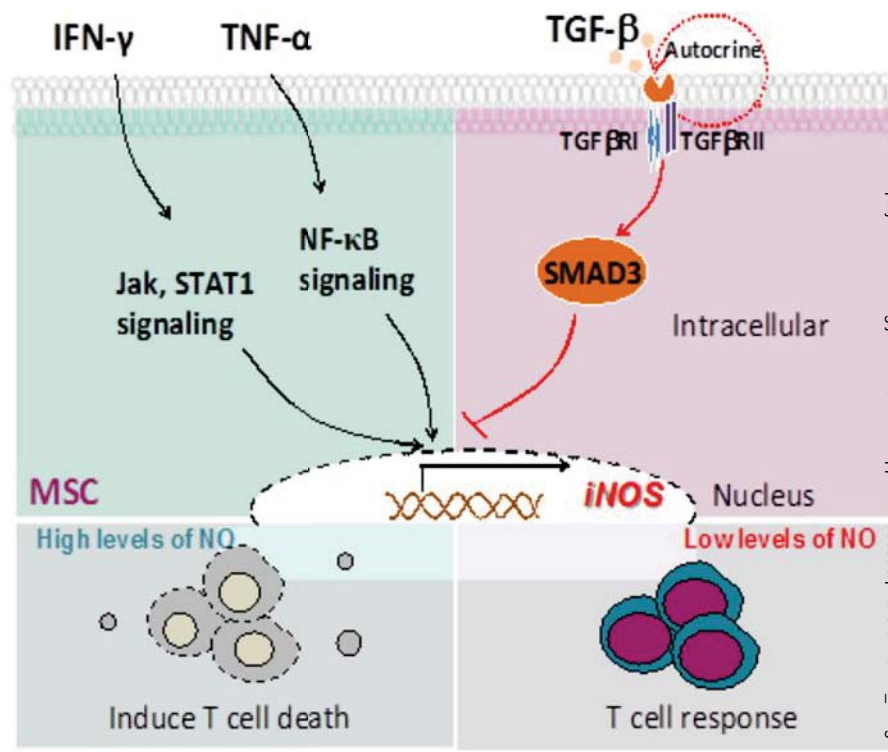


Supplementary Figure 3

A



B



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Supplementary Figure 4

Supplementary Figure 1. Three TGF- β family members had similar inhibitory effect on iNOS expression. MSCs were cultured at 5×10^5 /ml and stimulated with 10 ng/ml IFN γ and 10 ng/ml TNF α in the presence of different concentrations of TGF β 1, TGF β 2, TGF β 3 (0ng/ml, 1ng/ml, 10ng/ml). Nitrate concentration was determined by Griess test.

Supplementary Figure 2. High-dose of three TGF- β family members decreased the expression of extracellular membrane-bound TGF β RII. MSCs were cultured at 5×10^5 /ml and stimulated with 10 ng/ml IFN γ and 10 ng/ml TNF α in the presence of different concentrations of TGF β 1, TGF β 2, TGF β 3 (0 ng/ml, 0.1 ng/ml, 1 ng/ml, 10ng/ml and 100 ng/ml) for 24 hours. MSCs were then stained with polyclonal anti-mouse TGF- β RII-phycoerythrin and analyzed for the expression of both membrane-bound and intracellular TGF- β RII on flow cytometry using a FACS Calibur.

Supplementary Figure 3. TGF β had no effect on the degradation of iNOS mRNA and protein. (A) 2×10^6 MSCs were transfected by iNOS promoter reporter plasmid with Amaxa Nucleofector and seeded into 96-well plate at 1×10^5 cells/well. Twenty-four hours later, 10 ng/ml IFN γ , 10 ng/ml TNF α , and different concentrations of TGF β 1 and TGF β 2 (0.1 ng/ml, 1 ng/ml and 10 ng/ml) were added to the indicated wells. Forty-eight hours later, the cells were collected and the reporter activity was measured with luciferase

assay kit. **(B)** MSCs were activated with 10 ng/ml IFN γ and 10 ng/ml TNF α with or without 10 ng/ml TGF β 1. After 12 hours, 5 μ g/ml actinomycin D (ActD) was added to the culture media. Cells were collected into TRizol reagent after 0 hour, 2 hours, 4 hours, and 6 hours, and iNOS mRNA level was determined by real-time PCR. **(C)** MSCs were activated with 10 ng/ml IFN γ and 10 ng/ml TNF α with or without 10 ng/ml TGF β 1 or TGF β 2. After 12 hours, 1 mg/ml cycloheximide (CHX) was added to the culture media and cells were collected after 0 hour, 3 hours, and 6 hours for the determination of the iNOS protein level by western blotting analysis.

Supplementary Figure 4. **TGF β inhibited inflammatory cytokine induced IDO expression in human MSCs.** **(A)** Human umbilical cord derived MSCs were activated with 10 ng/ml IFN γ and 10 ng/ml TNF α with different concentrations of TGF β 1, TGF β 2, and TGF β 3 (1 ng/ml, 10ng /ml and 100 ng/ml) for 24 hours. IDO mRNA level was determined by real-time PCR. **(B)** TGF- β reverses the immune responses in the presence of MSCs. TGF- β reverses the immunosuppressive effect of MSCs by inhibiting iNOS expression triggered by inflammatory cytokines. This process is relied on the intracellular signaling of TGF- β , SMAD3-mediated pathway. Thus, TGF- β can exert immune enhancing roles in the presence of MSCs. Also, TGF- β secreted by MSCs plays a pivotal role in balancing the immunosuppressive function of MSCs.