5 Summary

The enhanced expression of T cell immunoglobulin and mucin protein-3 (TIM-3) on TADCs attenuates antitumor effects of DNA vaccines. To identify potential target (or targets) for reducing TIM-3 expression on tumor-associated DCs, we explored the molecular mechanisms regulating TIM-3 expression. Here we have identified a novel signaling pathway [c-Src→Bruton's tyrosine kinase (Btk)→transcription factors Ets1, Ets2, USF1, USF2] necessary for TIM-3 up-regulation on DCs. Both IL-10 and TGF-β, which are produced in tumor microenvironment, up-regulated TIM-3 expression on DCs via this pathway. Suppressed expression of c-Src or downstream Btk, Ets1, Ets2, USF1 or USF2 blocked IL-10- and TGF-β-induced TIM-3 up-regulation on DCs. Notably, *in vivo* knockdown of c-Src in mice reduced TIM-3 expression on tumor-associated DCs. Furthermore, adoptive transfer of c-Src-silenced DCs in mouse tumors enhanced the *in vivo* antitumor effects of immunostimulatory CpG DNA; however, TIM-3 overexpression in c-Src-silenced DCs blocked this effect. Collectively, our data reveal the molecular mechanism regulating TIM-3 expression in DCs and identify c-Src as a target for improving the efficacy of nucleic acid-mediated anticancer therapy.

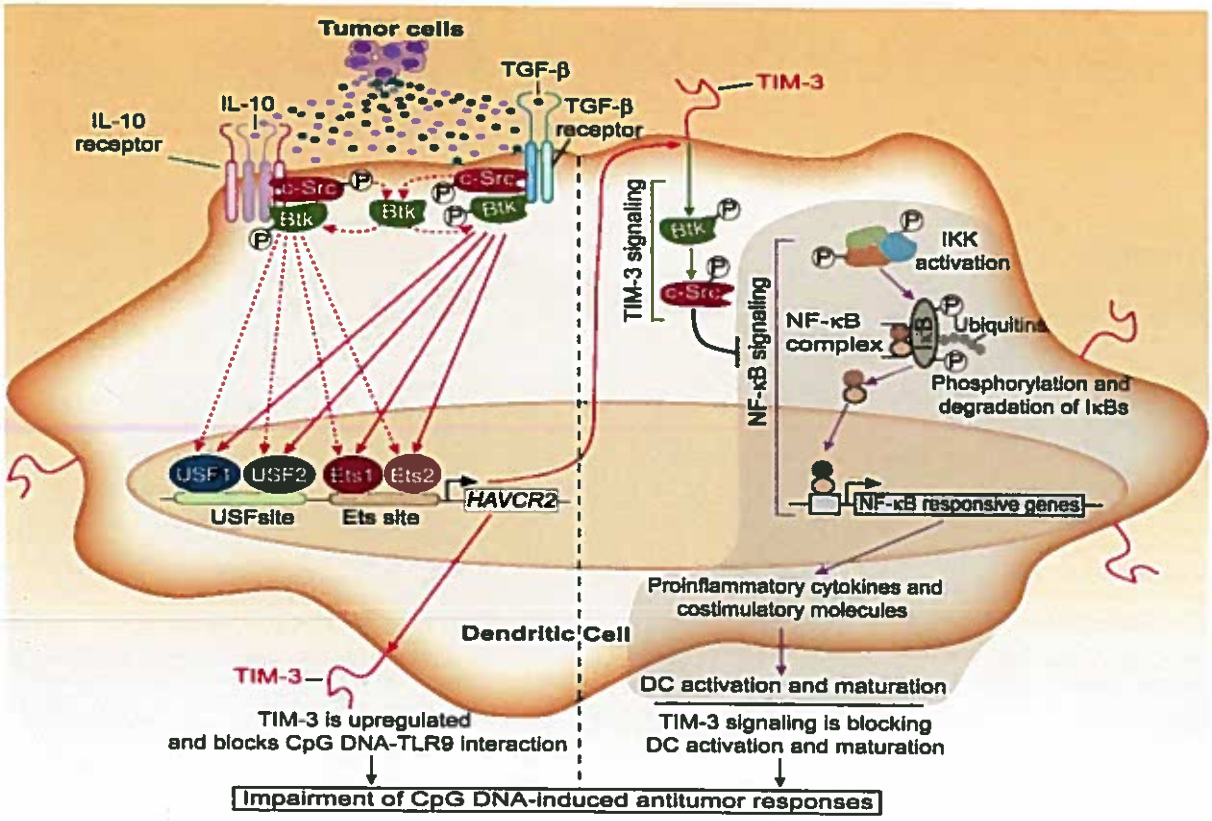

Fig. 5 Model depicting role of TIM-3 in CpG DNA-induced antitumor immune responses.