SUMMARY

Inhibitory activity of CaeA on proliferation of naïve and effector CD4 T cells and CD8 T cells were checked at different concentration using *in vitro* system. Dose dependency in the inhibition was seen on increasing the concentration of CaeA. Further, investigation of CaeA on different subtypes of CD4 T cells was essential to prove its efficacy. CD4 T cells are subdivided into different subtypes viz. Th1, Th2, Th17 and Tregs, on the basis of cytokines or transcription factors they express. CaeA inhibited the differentiation of Th1 cells and Th17 cells *in vitro*. It also suppressed the secretion of IFN-γ and IL-17. The results were further confirmed by the inhibition of transcription factors T-bet and RORγt for Th1 cells and Th17 cells, respectively. Further, CaeA was able to regress the functionality of effector Th1 cells and Th17 cells. Presence of CaeA in the culture resulted in the decrease of percentage of both $CD4^+IFN\text{-}\gamma^+$ T cells and $CD4^+IL\text{-}17^+$ T cells. Surprisingly, we observed unique and interesting results in the case of Treg cells. CaeA promoted the differentiation of Tregs, since augmentation in the percentage of $Foxp3^+$ T cells was eminent. Even in the absence of TGF-β, CaeA could enhance the percentage of Tregs. Further, it worked synergistically with TGF-β and considerably increased the percentage of Tregs compared to the TGF-β alone. Intriguingly, restricted proliferation of CD8 T cells was also noted when cultured with CaeA. Both naïve and effectors cells were inhibited in the presence of CaeA. A sharp decline in the secretion of IFN-γ was also noticed. In addition, CaeA inhibited the proliferation of MHC-incompatible CD8 T cells, along with decrease in the yield of granzyme. In the current study, the reduced clinical score and pathologies of EAE mice treated with CaeA were likely due to the combinatorial effect of CaeA by suppressing the autoreactive Th1 cells and Th17 cells. Interestingly, we observed improved Treg response, as evidenced by the

expansion in the percentage of these cells during the EAE course. We also noted dramatic reduction in the generation of autoreactive Th1 cells and Th17 cells and secretion of IFN-γ and IL-17 in CaeA treated mice. Our study shows that CaeA repressed the endurance of mature antigen-specific effector T cells and memory T cells in the CaeA treated EAE mice. Impudently, CaeA may act directly on naïve T cells. Thus, restricting the naïve T cell differentiation to effector T cells. This was evident from the fact that CaeA acted on T cells as early as 1h of initial culture. Positively, we noted that antigen specific T effector cells obtained from EAE mice were repressed by CaeA *in vivo,* as well in their ability to proliferate and release IFN-γ and IL-17, upon re-stimulation with MOG peptide. Present study shows diametric role of CaeA. In essence, the study suggests that CaeA can be an important future drug that may help to treat autoimmune diseases.