East Coast fever (ECF) is a fatal disease of cattle, caused by Theileria parva, an intracellular haemoproteozoan parasite. Immunity in cattle immunized against T. parva using the infection and treatment method (ITM) is mediated by parasite-specific CD8+ cytotoxic T lymphocytes (CTL) that destroy schizont-infected cells. Ten schizont antigens (Tp1-Tp10) recognised by CD8+ CTL have been identified and vaccine potential of six of these has been evaluated. Initial results have shown a significant correlation between CD8+ CTL responses and survival after challenge, but CTL responses have only been induced in a proportion of vaccinated cattle. Since CD4+ T cells play a crucial role in the induction of naïve CD8+ T cell responses, it may be critical to incorporate antigens that contain CD4+ helper T cell epitopes in such a CTL-targeted vaccine. This work aimed at determining whether six of the CD8+ CTL-targeted vaccine candidate antigens contain CD4+ T cell epitopes and to establish whether cattle immunized by ITM mounted CD4+ T cell responses to these antigens. Peripheral blood mononuclear cells (PBMC) obtained from ITM-immunized cattle or those that had recovered from challenge infection following immunization with the candidate vaccine antigens, were used to generate antigen-specific CD4+ T cell lines by repeated stimulation with autologous T. parva infected cells (TpM) or pools of overlapping synthetic peptides. Screening and mapping of CD4+ T cell epitopes was carried out using both Lymphocyte proliferation and IFN-γ ELISpot assays. Two TpM-stimulated CD4+ T cell lines, generated from ITM-immunized animals, did not recognize any of the CTL target antigens. However, ex vivo CD4+ T cell responses were detected to Tp1, Tp4, Tp5, Tp6 and Tp8 following immunization with CTL target antigens and challenge with T. parva sporozoites. CD4+ T cell lines specific to antigen Tp1 were generated from one of these animals (BZ001) by stimulation with synthetic peptides and two antigenic peptides mapped on Tp1 (Tp11450-1459 and Tp11983-1992). Interestingly, this CD4+ T cell line recognized the recombinant Tp1 protein but did not respond to autologus schizont infected cells. The results of this study indicate that the CTL target antigen Tp1 contains CD4+ T cell epitopes which are sub-dominant or if dominant, are not sufficiently expressed on the surface of schizont-infected cells. The work also suggests the need to develop methods of screening for CD4+ T cell antigens from T. parva that could be used to identify novel, immunodominant antigens presented on T. parva schizont infected cells that may enhance the potency of the sub-unit vaccine under development.