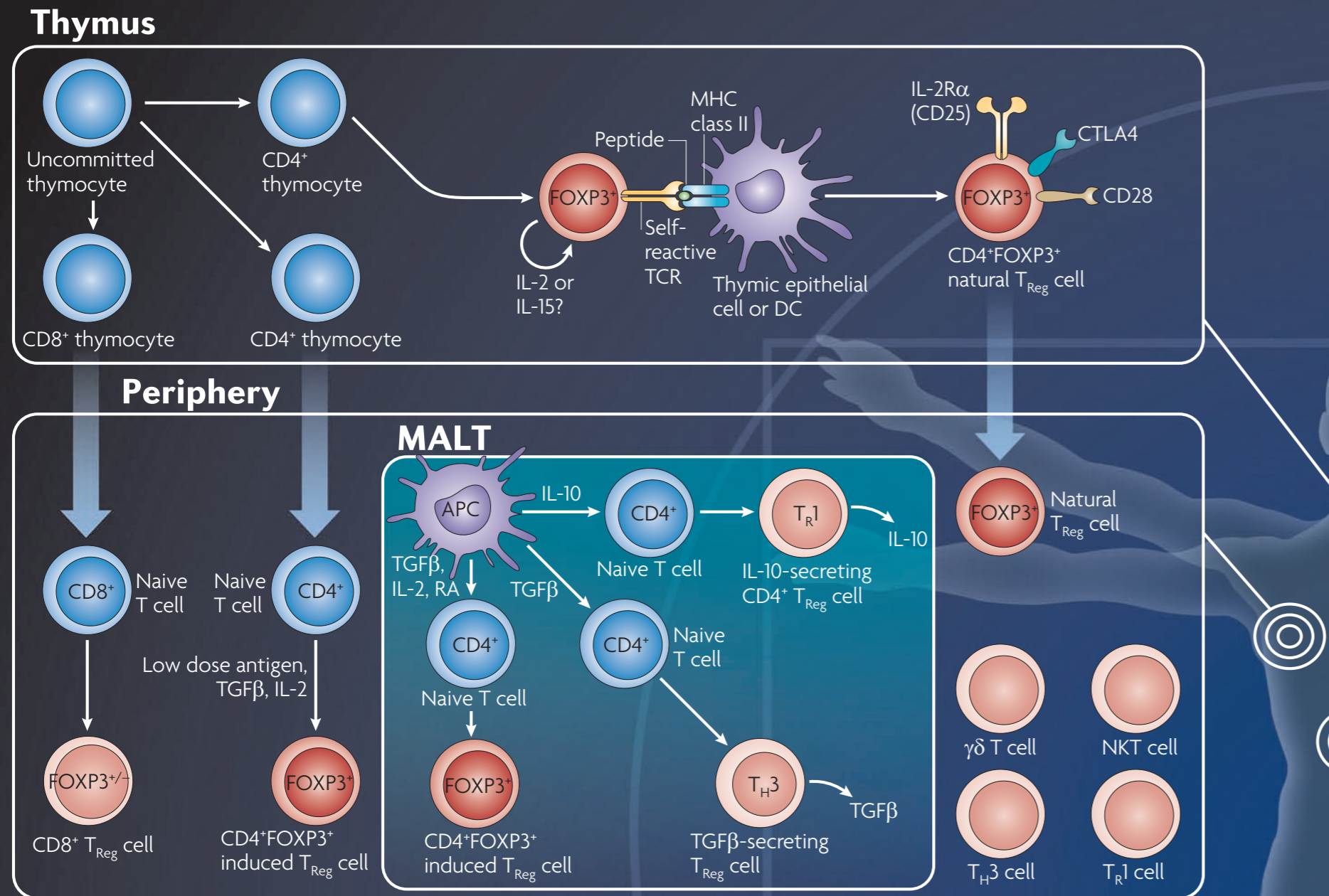


To avoid immune-mediated pathology and unrestricted clonal expansion of responder T cells, the immune system has subsets of T cells, known as regulatory T (T_{Reg}) cells, that are dedicated to mediating immune suppression. The most important subset of T_{Reg} cells expresses the transcription factor forkhead box P3 (FOXP3). Both mice and humans with genetic deficiencies of FOXP3 develop severe abnormalities of immune homeostasis. T_{Reg} cells modulate the immune response in numerous settings, including autoimmune disease, allergy, microbial infection, tumour immunity,

organ transplantation, foetal-maternal tolerance and even obesity. Defects in T_{Reg} cell function may be an important factor in the development of autoimmunity or in the failure to control immunopathology, whereas overactive T_{Reg} cell function may contribute to the suppression of tumour immunity. Enhancement of T_{Reg} cell function either pharmacologically or by cell-based therapy may prove to be an adjunct to the treatment of autoimmunity, whereas deletion or inactivation of T_{Reg} cell function may facilitate the generation of tumour immunity or enhance responses to weak vaccines.

Development and phenotype of regulatory T cells

Many T cell types have immune regulatory function, but the two most important T_{Reg} cell subsets express the transcription factor FOXP3 and develop in the thymus or can be induced in peripheral sites including the mucosa-associated lymphoid tissue (MALT). Although expression of FOXP3 is considered a useful marker for these cell subpopulations in mice, FOXP3 expression may also be induced in human T cells that lack T_{Reg} cell function. However, functional activated human FOXP3⁺ T_{Reg} cells express a unique pattern of cell surface markers that can facilitate their isolation. A third important type of T_{Reg} cell secretes the immunosuppressive cytokine interleukin-10 (IL-10) and may develop from conventional CD4⁺ T cells by activation in the presence of IL-10 or may develop from T helper 1 (T_H1) or T_H2 cell subsets. Other T cell subpopulations including natural killer T (NKT) cells, $\gamma\delta$ T cells and CD8⁺ T cells can also exert potent suppressor functions in certain settings. Although both human and mouse CD8⁺ T cells can be induced to express FOXP3, a suppressive function for these cells *in vivo* has yet to be clarified.



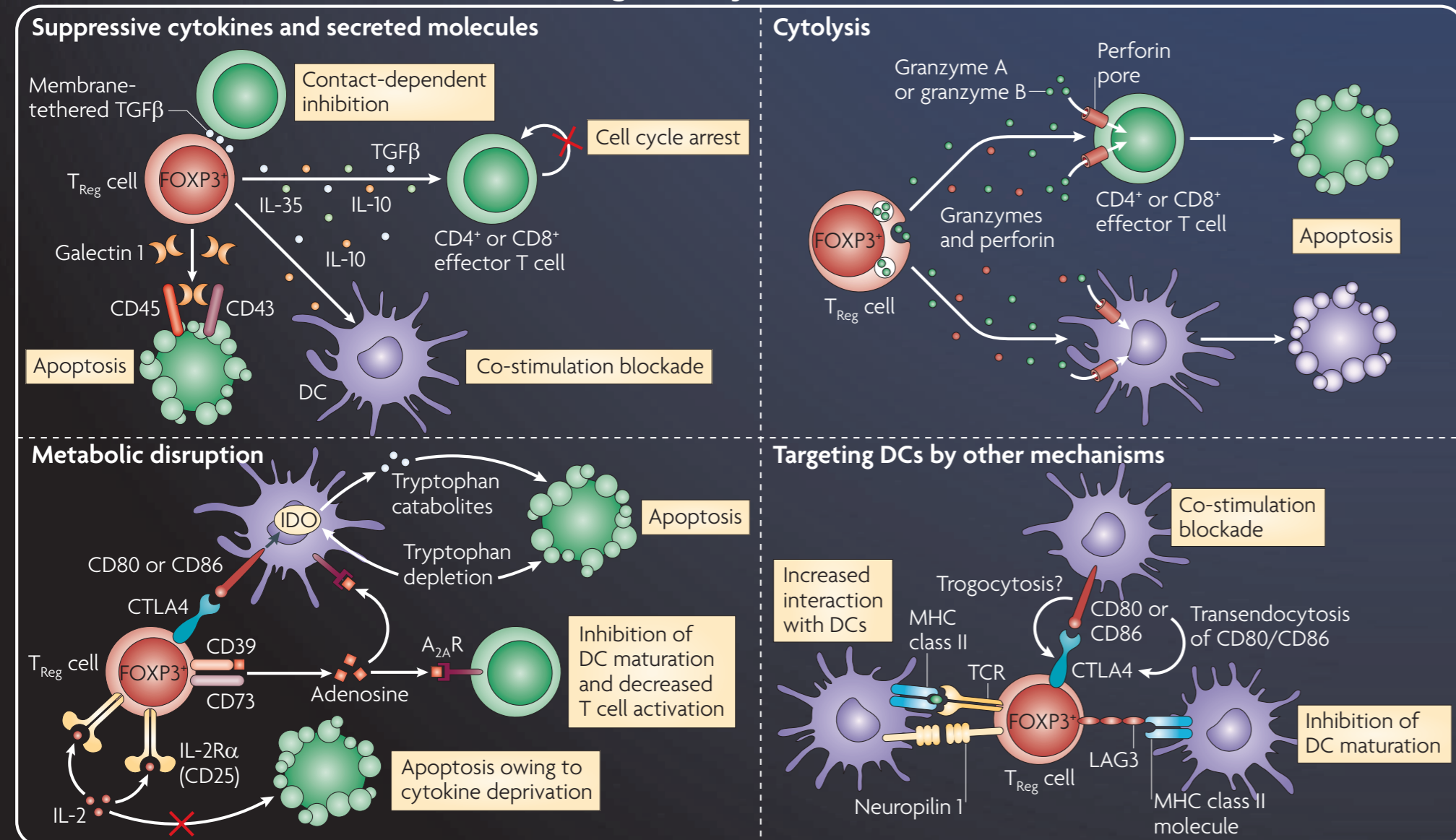
Plasticity in the periphery

Under certain conditions, FOXP3⁺ T_{Reg} cells can downregulate their expression of FOXP3, lose suppressor functions and manifest some of the functions of conventional effector T_H1 , T_H2 , T_H17 and T_H1 cell subsets. The key causes of this loss of FOXP3 expression include inflammatory environments with high levels of cytokines that are normally involved in the induction of effector T cells, such as IL-6 and interferon- γ (IFN γ). In addition, T_{Reg} cell-specific deletion of certain transcription factors that are shared between T_{Reg} cells and effector cell subsets (for example, the T_H2 cell-specific factor IRF4) results in impaired suppression of the T_H2 cell responses by the T_{Reg} cells.

Phenotypic markers of FOXP3⁺ regulatory T cells

Markers shared by FOXP3 ⁺ T_{Reg} cells and conventional activated CD4 ⁺ T cells (mice and humans)	Markers preferentially expressed by activated mouse FOXP3 ⁺ T_{Reg} cells	Markers specifically expressed by activated human FOXP3 ⁺ T_{Reg} cells
CD25 GITR CD45RB ^{low} (mice only) CD45RO (humans only) Folate receptor 4 (mice only)	FOXP3 Latent TGF β CD103	FOXP3 ^{hi} Latent TGF β GARP CD121a (IL-1R1) CD121b (IL-1R2)
	Subpopulations of human FOXP3⁺ T_{Reg} cells	
	CD45RA ⁺ FOXP3 ^{low} (naive) CD45RA ⁺ FOXP3 ^{hi} (activated) CD45RA ⁺ FOXP3 ^{low} (cytokine-secreting)	

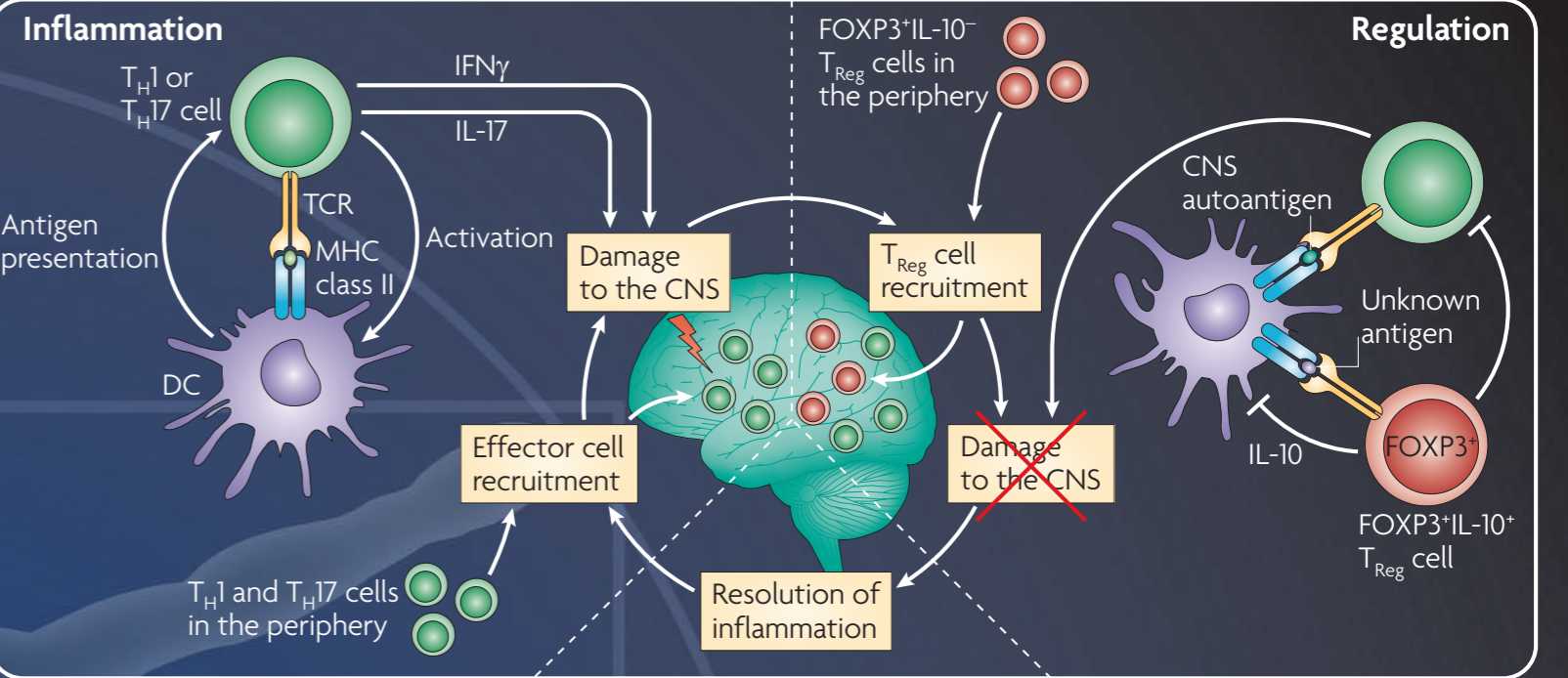
Mechanisms of action of FOXP3⁺ regulatory T cells



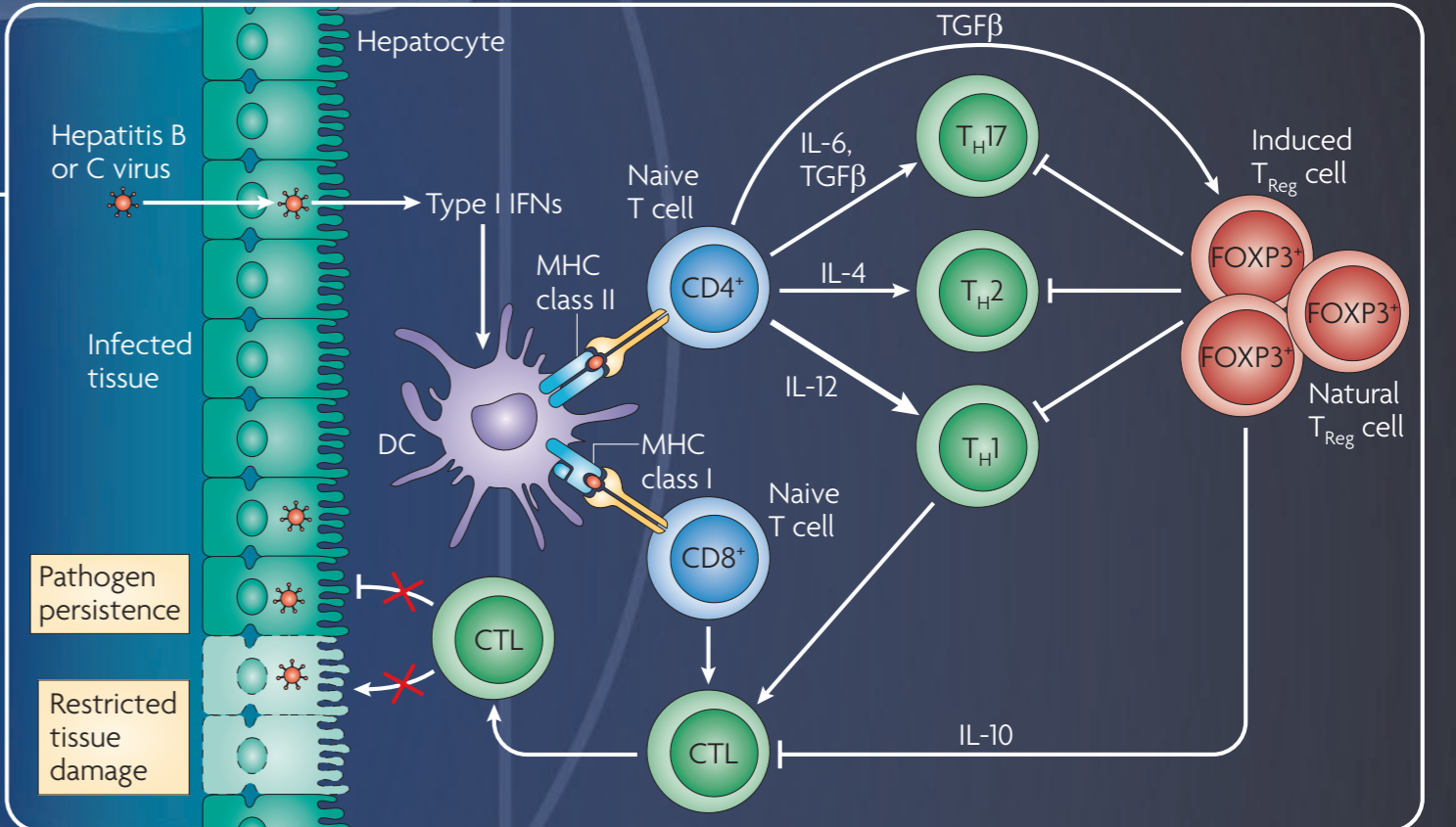
Function of FOXP3⁺ regulatory T cells

FOXP3⁺ T_{Reg} cells have been shown to influence the outcome of immune responses in several tissues. For example, in the intestine T_{Reg} cells have a key role in maintaining tissue homeostasis by inhibiting the overactivation of dendritic cells (DCs) and effector T cells. In the case of autoimmunity, such as that depicted in the central nervous system (CNS), T_{Reg} cells can have a beneficial effect by short-circuiting the inflammatory loop of T cells and antigen-presenting cells (APCs). This same general mechanism can have negative consequences in the setting of a tumour, in which T_{Reg} cells can inhibit the antitumour immune response, thereby preventing tumour clearance. During infection T_{Reg} cells carry out a delicate balancing act: preventing immunity would lead to the inability to clear the pathogen, whereas unrestrained immunity would lead to unwanted immune-mediated tissue destruction. In each of the examples shown, we focus on the role of FOXP3⁺ T_{Reg} cells, although interactions between multiple immune cell types and indeed different types of regulatory T cell are also likely to be important in the regulation of immune responses.

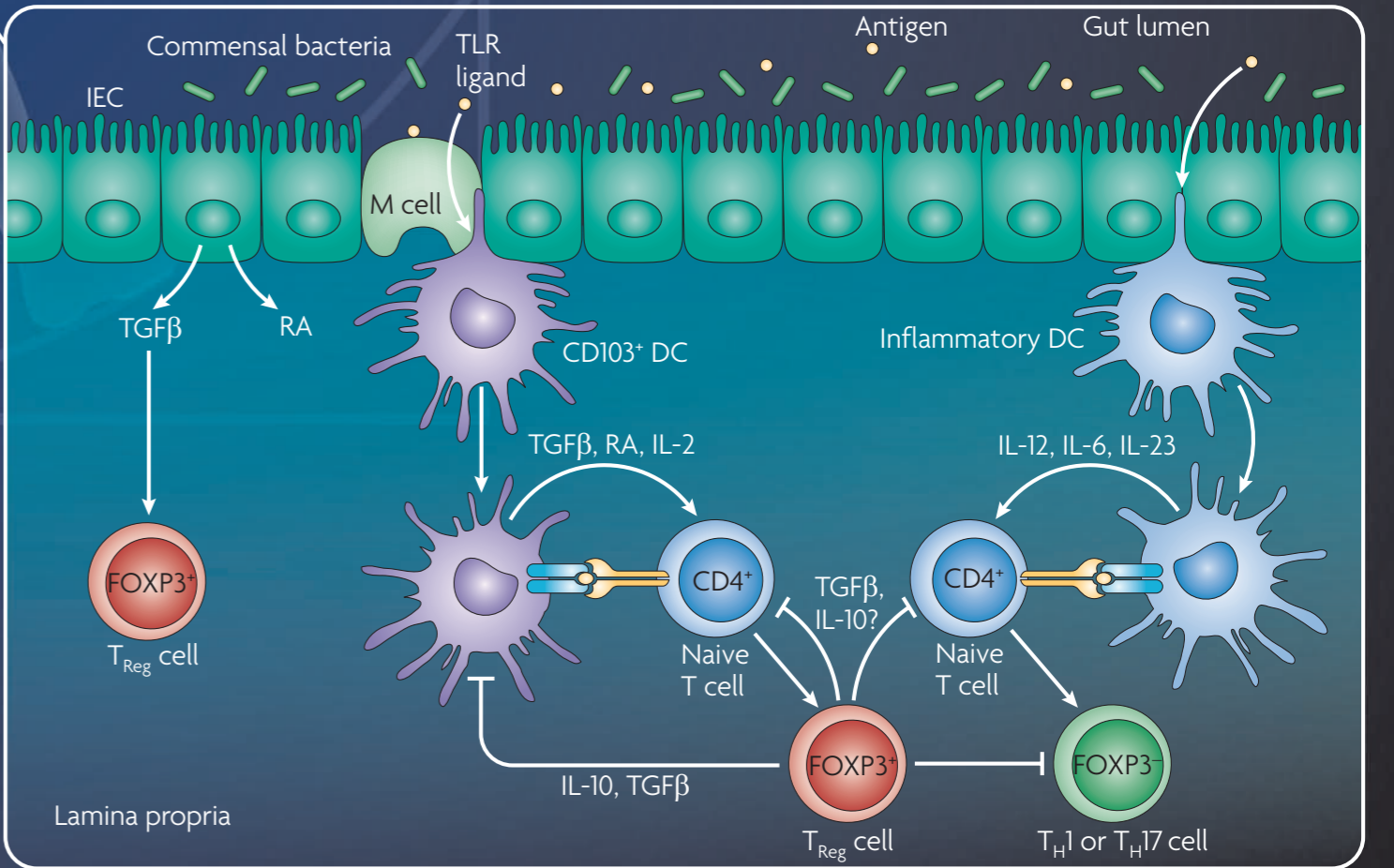
Preventing autoimmunity



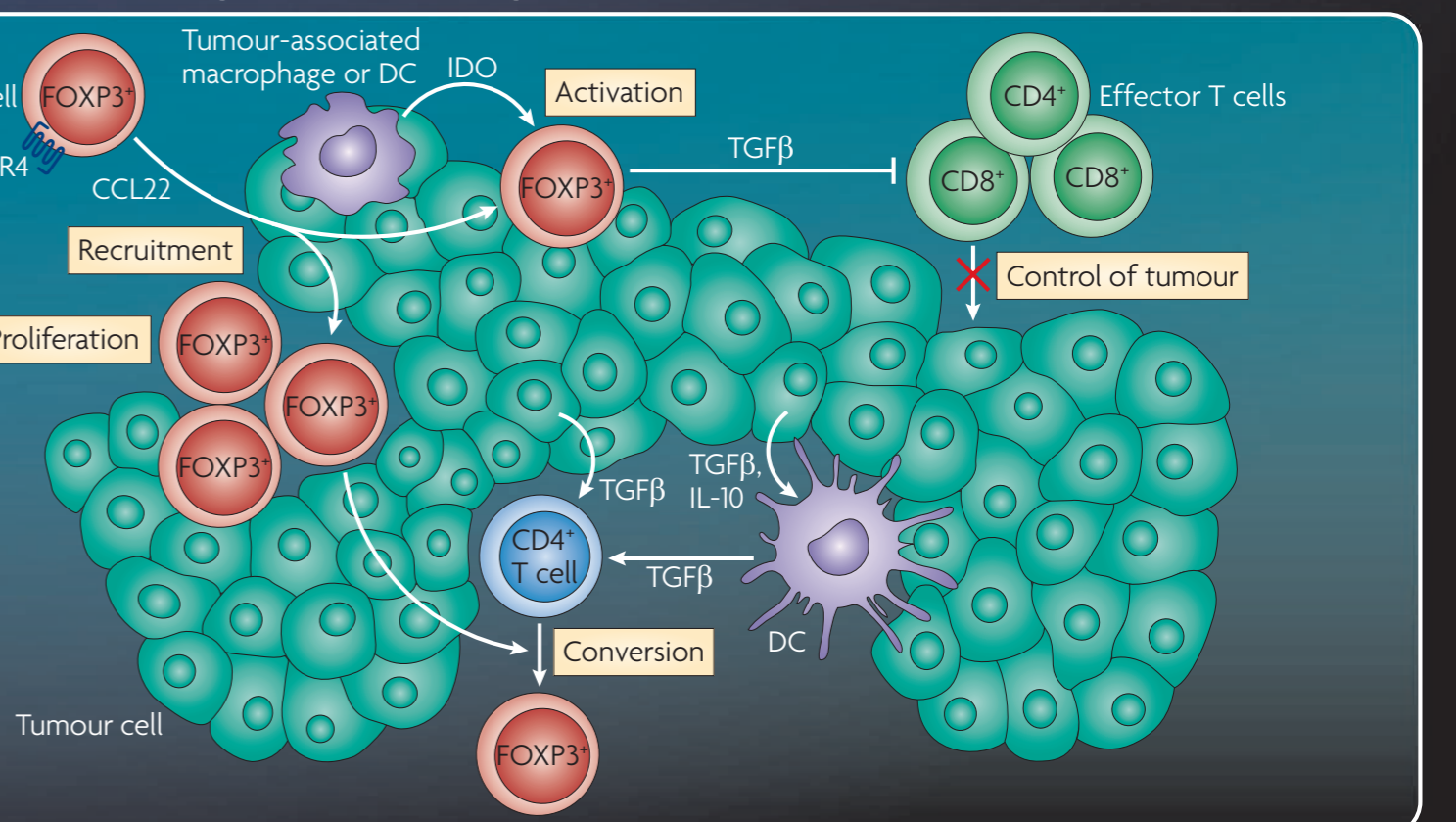
Establishing chronic infection



Maintaining intestinal homeostasis



Promoting tumour progression



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STEMCELL Technologies
Regulatory T cells (Tregs) comprise only a small fraction of total CD4⁺ T cells in human peripheral blood and mouse spleen, and therefore must be highly enriched to evaluate their suppressive function and therapeutic potential. Since Tregs lack a unique cell surface marker and often share phenotypic similarities with activated T cells, isolation of highly purified Tregs is typically difficult and time consuming, often requiring multiple steps. To meet the needs of Treg researchers, STEMCELL Technologies has developed a full range of optimized Treg isolation kits that addresses these specific challenges.

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Starting sample	Phenotype of cells	STEMCELL Cell Isolation Kit	STEMCELL Catalog number
Human			
Whole blood	CD4 ⁺ CD25 ⁺ T cells	Complete Kit for Human CD4 ⁺ CD25 ⁺ T Cells	15862
	CD4 ⁺ CD127 ^{low} CD25 ⁺ T cells	Complete Kit for Human CD4 ⁺ CD127 ^{low} CD25 ⁺ Regulatory T Cells	15861
	CD4 ⁺ CD127 ^{low} CD49d ⁺ CD25 ⁺ T cells	Complete Kit for Human CD4 ⁺ CD127 ^{low} CD49d ⁺ CD25 ⁺ Regulatory T Cells	15864
PBMC	CD4 ⁺ CD25 ⁺ T cells	EasySep [®] /RoboSep [®] Human CD4 ⁺ CD25 ⁺ T Cell Isolation Kit	18062
	CD4 ⁺ CD127 ^{low} T cells	EasySep [®] /RoboSep [®] Human CD4 ⁺ CD127 ^{low} T Cell Enrichment Kit	19231
	CD4 ⁺ CD127 ^{low} CD49d ⁺ T cells	EasySep [®] /RoboSep [®] Human CD4 ⁺ CD127 ^{low} CD49d ⁺ Regulatory T Cell Enrichment Kit	19232
Mouse			
Spleen or other tissues	CD4 ⁺ CD25 ⁺ T cells	EasySep [®] Mouse CD4 ⁺ CD25 ⁺ Regulatory T Cell Isolation Kit	19782