2. AIM OF THE STUDY

DCs are a heterogeneous population of cells with different myeloid or plasmacytoid origin and differential phenotype and function (Shortman and Naik, 2007; Villadangos and Heath, 2005)and are crucial cells in the induction and regulation of adaptive immunity (Lanzavecchia and Sallusto, 2001; Pape et al., 2007).

Several BM and blood precursors of DCs have been identified, including monocytes (Shortman and Naik, 2007; Villadangos and Heath, 2005). Evidences for the capacity of monocyte to differentiate into DCs are reported in the mouse models, where monocytes have been *in vivo* tracked using internalized fluorescent latex microspheres (Geissmann et al., 2003; Ginhoux et al., 2006; Qu et al., 2004; Randolph et al., 1998; Randolph et al., 1999) or in the *Listeria monocytogenes,* and *Leishmania major* infection systems (Leon et al., 2007; Serbina et al., 2003).

On the other hand, in humans it has not been defined which DCs subset originates from monocytes and experiments of transendothelial migration of DCs generated from monocytes (Randolph et al., 1998) have not conclusively established which are the processes that promote monocytes differentiation into DCs or into macrophages (Krutzik et al., 2005).

In culture, human monocytes acquire a macrophage phenotype both in the presence and in the absence of added cytokines, such as M-CSF (Gangenahalli et al., 2005), thus differentiation of monocytes into macrophages seems to represent a default differentiation program of monocytes upon extravasation (Lewis et al., 1999). Conversely, well-defined cytokine cocktails are required for monocytes to differentiate into DCs (Comes et al., 2002; Mohamadzadeh et al., 2001; Santini et al., 2000; Zou and Tam, 2002).

Several cell types have been indicated as a possible source of cytokines capable of inducing monocytes differentiation into DCs *in vitro* (Brossart et al., 1998; Hegde et al., 2007; Wirths et al., 2002; Zhang et al., 2007).

However, the stimulus and context in which these cells would promote the monocyte differentiation into DCs instead of macrophages could not be unambiguously defined.

Starting from the hypothesis that monocytes could represent progenitor cells of tissue macrophages under physiological conditions and cells committed to the local replacement of migrating or dying DCs following an inflammatory process (Shortman and Naik, 2007; Villadangos and Heath, 2005) and taking into consideration that monocytes are co-recruited with lymphocytes in chronic inflammation sites, the aim of this thesis was to analyze the consequence of T cell activation on the monocytes differentiation into DCs.

In details, a huge panel of human Th1, Th2 and Th0 antigen specific T cell clones (TCC)

was generated and the consequence of antigen presentation on monocytes differentiation into DCs was evaluated.

It was evaluated whether soluble factors released by activated T lymphocytes or their intimate interaction with monocytes were crucial to induce monocytes differentiation.

The different types of DCs populations were then analyzed for their phenotype characteristics.

Finally it was tested the functionality of the diverse DCs through the analysis of cytokines release, their capacity to phagocytose and endocytose, their antigen-presentation capability and their ability in inducing priming of naïve $CD4^+$ T lymphocytes and activation of antigen specific TCC.

A model of monocyte differentiation not based on the use of synthetic cytokines or factors and that reasonably reproduces inflammatory microenvironments allowing an easier extrapolation of *in vitro* data was obtained.