

BOLSA DE INVESTIGAÇÃO (M/F)

Referência: PTDC/SAU-FCF/101017/2008

Título do Projecto: Role and mechanism of action of immunoregulatory iNKT and DC cells in *Leishmania* infection: new approaches for vaccination?

Código interno: PR303004

Está aberto concurso para recrutamento de um(a) bolsheiro(a) de Investigação para colaborar no projecto acima referido, financiado pelo programa COMPETE - Programa Operacional Factores de Competitividade na sua componente FEDER e pelo orçamento da Fundação para a Ciência e a Tecnologia na sua componente OE.

A bolsa, em regime de exclusividade, terá a duração de 1 ano, eventualmente renovável, com início previsto em 15 de Julho de 2010.

O valor mensal da bolsa será de € 980,00, pago por transferência bancária (preferencialmente).

Local de trabalho: IBMC - Instituto de Biologia Molecular e Celular (Grupo “Parasite Disease”)

Programa de trabalho: ver anexo.

Perfil pretendido:

Os candidatos devem ter o grau de mestre em Bioquímica, Ciências Biológicas, Farmacêuticas ou afins, sendo dada preferência a quem possuir experiência pós-graduada em Imunologia, Parasitologia, Biologia molecular e Celular.

O prazo para recepção de candidaturas decorre de 28 de Junho a 12 de Julho de 2010.

As propostas deverão incluir carta de motivação, CV e ser enviadas por correio electrónico para candidaturas@ibmc.up.pt com indicação do código interno (PR303004).

A contratação será regida pelo estipulado na legislação em vigor relativamente ao Estatuto de Bolsheiro de Investigação Científica, nomeadamente a Lei 40/2004, de 18 Agosto, e o Regulamento de Bolsas de Investigação Científica do IBMC (www.ibmc.up.pt/fellowships.php).

“Role and mechanism of action of immunoregulatory iNKT and DC cells in *Leishmania* infection: new approaches for vaccination?”

Supervisor:

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Project summary:

Several million people worldwide are affected by *Leishmania* infection, especially in tropical areas and in Southern Europe, where leishmaniasis has emerged as an AIDS-associated opportunistic infection. However, classical methods failed to obtain an efficient vaccine. It was reported that surface glycoconjugates of *Leishmania* are potential vaccine candidates, and that these antigens could stimulate a new type of immunoregulatory cells, the invariant Natural Killer T (iNKT) cells. Antigens can activate directly or indirectly iNKT cells; direct activation could be by microbial cell-wall lipids with structural similarities to α -GalCer; or indirect activation through the recognition of their endogenous ligands presented by CD1d molecules.

The aim of this project is to determine whether *Leishmania* antigens (glycoproteins, lipophosphoglycan or galactosylceramide compounds) presented by dendritic cells (DC) can activate iNKT cells, and whether this complex could act on B cells to amplify anti-*Leishmania* immune responses. We will also analyze the role played by the subsets of DC and iNKT cells and the mechanisms implicated. Our study will contribute to the knowledge of iNKT cell cross-talk with DCs and B cells in a context of anti-*Leishmania* immune responses. In conclusion, the characterization of iNKT cell responses to glycoconjugates of *Leishmania*, which are potential vaccine candidates, could contribute to the development of new therapies against *Leishmania* infections.

Since that, the major aim of our project is to determine whether iNKT cells can respond to *Leishmania infantum* antigens, specially their glycoproteins, lipophosphoglycan or galactosylceramide compounds, we will take advantage of the expertise of Parasite Disease lab (Santarém et al, 2005) and first perform the purification of these antigens. Secondly, we will analyze if they can activate iNKT cells. Recent studies revealed that microbial stimulation of iNKT cells can be direct, by TCR recognition of cell-wall glycolipids, or indirect, through the recognition of their endogenous (self) ligands presented by CD1d molecules. After infection, this latter recognition is amplified by IL-12, IL-18 or IL-23 secreted by activated DC (Bendelac et al, 2007; Kronenberg, 2005). Recently, it was demonstrated by Leite-de-Moraes' team that, in contrast to the majority of iNKT cells, a iNKT subset produce high levels of IL-17 and low levels of IL-4 and IFN- γ (Michel et al, 2007). Knowing that IL-23 favors while IL-12 reduces IL-17 production by T cells (Weaver et al, 2007), and that the secretion of these cytokines by DC depends on the nature of the antigen and of the type of DC (myeloid or plasmacytoid), we propose to analyze the profile of cytokine produced by different types of DC when loaded with *Leishmania* antigens and their effect on iNKT cell subsets.

Another important point is that activated NKT cells enhance the level of protective anti-parasitic immune response (Spath et al, 2004) through their action on B lymphocytes but no data exists concerning the capacity of iNKT cells to help B cells in an anti-parasitic condition. Thus, thirdly we propose to stimulate iNKT cells with DC loaded with *Leishmania* antigens and analyze their ability to influence immunoglobulin production by both naïve (CD27-) and memory (CD27+) B cell subsets. The results obtained will get

insights into anti-parasitic immune responses and even contribute to obtain an efficient vaccine against leishmaniasis.

Specific objectives are:

- 1- Purification of *Leishmania* antigens will be done in order to analyze if they can activate iNKT cells that will in turn contribute to anti-parasitic immune responses and even improve vaccination against this parasite.
- 2- Study the subsets of DCs and iNKT cells will be done; we will test the capacity of plasmacytoid and myeloid DC loaded with Ag to stimulate different types of iNKT cells according to their expression of CD4, CD8, CD161, CCR6, CCR4 or CXCL10 markers.
- 3- Analyze the signaling pathways that are involved in the iNKT cell activation by these antigens.
- 4- Test the capacity of activated iNKT cells to influence Ig production by both naïve (CD27-) and memory (CD27+) B cell subsets.
- 5- Study the immune responses in mice induced by the *Leishmania* iNKT stimulating antigens, using DC(s) loaded with these antigens.
- 6- Test the effect of these *Leishmania* iNKT-stimulating antigens in the context of the infection.
- 7- Depending of the immunological role of *Leishmania* iNKT stimulating antigens a vaccine approach will be tested.