



Analysis of gene expression by quantitative polymerase chain reaction of CD209, CLEC7A, interleukin (IL)-15RA, Toll-like receptor (TLR)-1, TLR-2, TLR-9, IL-12B, tumour necrosis factor (TNF), transforming growth factor (TGF)-β1, IL-15 and IL-10 in mature dendritic cells (mDCs) stimulated for 48 h with a specific maturation cocktail [IL-1β (25 ng/mL), IL-6 (1,000 U/mL), TNF (50 ng/mL) and prostaglandin E2 (10⁻⁶M)] or in *Mycobacterium leprae* stimulated DCs (ML DCs) pulsed with sonicated antigen of *M. leprae* (10 µg/mL) compared to nonstimulated immature DCs (iDCs). DCs derived from monocytes were obtained from healthy controls (HCs) (n = 14), tuberculoid patients (n = 9) and lepromatous patients (n = 11). *: p < 0.05; **: p < 0.01; ***: p < 0.001; #: p < 0.05; ##: p < 0.01, Friedman nonparametric ANOVA, Kruskal-Wallis nonparametric ANOVA and Dunn's multiple comparison test.

tmRNA analysis

For gene expression analysis by mRNA, dendritic cells (DCs) from nine tuberculoid, 11 lepromatous leprosy patients and 14 healthy controls (HCs) were harvested in Trizol Reagent (Invitrogen, USA) after two days of stimulation and kept at -80°C until processing. Total mRNA (1 µg) was transcribed by using 200 U of SuperScript III Reverse Transcriptase enzyme (Invitrogen) and 500 ng of oligo(dT) 12-18 primer (Invitrogen) according to the manufacturer's instructions. The expression of target genes CD209, CLEC7A, interleukin (IL)-10, IL-12B, IL-15, IL-15RA, transforming growth factor (TGF)-β1, Toll-like receptor (TLR)-1, TLR-2, TLR-9 and tumour necrosis factor (TNF) was evaluated by real-time polymerase chain reaction using Taqman assays (Applied Biosystems Inc, USA) in a StepOne Plus equipment (Life Technologies, USA), as indicated by the manufacturer. *GAPDH* and *B2M* genes were used as endogenous controls. Gene expression was estimated by relative quantification using a relative standard curve composed by five serial dilutions from a pool of DCs mRNA.

Supplementary Figure shown that TLR-1, TLR-2, CLEC7A, CD209 and TGF-β1 genes showed lower expression in mature DCs (mDCs) and sonicated *Mycobacterium leprae* (ML DCs) compared to immature DCs. On the other hand, IL-15, IL-12B and IL-15R genes increased expression in mDCs and a tendency to higher expression was also observed in ML DCs, agreeing with the results of flow cytometry that indicated some grade of activation of DCs by *M. leprae*. For TNF gene (Fig. 4), low expression in tuberculoid patients compared to lepromatous patients and HCs can be seen in all conditions of DCs. The same pattern was observed in TLR-9 gene. IL-10 gene expression showed a tendency to decrease in ML DCs.