別紙２

Results report

１． Title of Research and Development：Protective and subversive mechanisms of macrophage genes in Mycobacterium tuberculosis infection

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４． Results of Research and Development:

This research aims at identification and analyses of macrophage genes that work protective or subversive in Mycobacterium tuberculosis (Mtb) infection. For this purpose, we compared gene expression dynamics of macrophage in Mtb infection with that of cytokine-induced macrophage activations. The main experimental task of both parties were selection of candidate genes using already obtained high quality transcriptome data (Japan), Creation of the constructs for the perturbation experiments (Japan), gene expression analysis of the perturbation samples (Japan), Mtb infection experiments using macrophages and experimental mice (South Africa) and preparation of samples for the gene expression analysis (South Africa). Because we have already had a great progress in our analysis, we focused on writing papers as much as possible in the last fiscal year, with minimum additional wet experiments.

In the stimulation by pathogen infection including Mtb, the activated macrophage cells, as well as classically activated one, express various host protective factor genes involved in inflammation, such as *Tnf*, etc. We successfully identified that Batf2 is a novel transcription factor (TF) involved in gene expression of those factors. In addition, we found that Batf2 associates with Irf1, another TF induced by IFNg, and induces inflammatory genes, which was published in *J. Immunology*. Further, including the above results, we have published a review article in *Oncotarget*. Furthermore, in vivo functional analysis of Batf2 is going on using the Batf2 KO mice. We have already obtained interesting results for the Mtb infection response in the KO mice and aim at publication in earlier timing.

We also analyzed transcriptional regulation dynamics of classical and alternative activations in macrophage cells. Surprisingly, the analysis revealed that almost same set of transcription factor binding motifs are involved in both activations, although those two activations shows quite different features. The results suggest that same TFs are involved in the different activations. Further, using the comprehensive promoter-level expression profiles, we successfully identified novel TF genes, peripheral genes and lncRNAs that are considered to play important role in those activations, which was published in *Nucleic Acids Research*. In addition, we explored effect of the alternative activation to Mtb infection by using the IL4 receptor KO mice that does not occur alternative activation. We found that alternative activation in macrophage cells has almost no effect in Mtb infection, which was published in *PLosONE*.

In addition to the above achievement, we are analyzing large scale time course expression data in Mtb-infected macrophages, which was obtained by the deepCAGE method. Because we have almost completed the analysis, we aim at the paper publication in earlier timing. Further, we found that Batf, a family TF of Batf2, is also transiently induced in pathogen (Mtb)-infected macrophages. We are now analyzing the function of Batf in Mtb infection. Finally, although we had a plan to have a workshop with the collaborator in the last half of the fiscal year, it was not held. It is because we decided in our discussion that we took paper publication work as our first priority.