Tropentag, September 20-22, 2023, hybrid conference

"Competing pathways for equitable food systems transformation: Trade-offs and synergies"

Development of a fluorescent RBL reporter system for diagnosis of porcine cysticercosis

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Abstract

Porcine cysticercosis (PCC) is a World Organisation for Animal Health listed notifiable disease in pig, caused by the larval stage of Taenia solium. Pigs get infected by ingesting human stool or water/vegetation contaminated with *Taenia solium* eggs. The disease is endemic in Latin America, sub-Saharan Africa, as well as South and South East Asia. PCC hampers food security and affects livelihoods of pig farmers resulting in reduced pork value and economic loss, especially in developing countries. Available serological diagnostic tests based on IgG detection are characterised by low specificity. This study aims to assess suitability of detecting parasite-specific Immunoglobulin E (IgE) using IgE based reporter cell lines. Our objectives are to create a reporter cell line that is able to bind pig IgE and to identify, clone, and recombinantly express candidate *T. solium* allergens, which will be assessed for their suitability as 'diagnostic allergens' for diagnosis of PCC. We cloned a synthetic pig high affinity IgE receptor alpha chain (Ss-FCER1A) into a plasmid vector (pcDNA5). After ligation and bacterial transformation, we transfected our target transformant (Ss-FCER1 $^{A}/_{pcDNA5}$) into Rat basophilic leukemia (RBL) cells stably transfected with neuropeptide Y monomeric red fluorescent protein (NPY-mRFP) and developed a chimeric pig/rat 'porcinised' reporter system (RBL NPY-mRFP Ss-FCER1A). The developed cell lines were then continuously sub cultured for selection of stable transfectants. Five diagnostic *T. solium* oncospheral allergens (Q9NI46, Q2XNL7, K0A0S9, E5LBB8, W8P1J2) were identified through employing several immunological and bioinformatic techniques aiming to identify IgE-immunoreactive antigens. These coding sequences for the selected allergens were then cloned into pTT expression vectors and subsequently transformed into bacteria cells for plasmid amplification. We are currently attempting recombinant expression of those cloned products in HEK293–6E cells grown in suspension. Once the expressed IgE immunoreactive antigens have been purified, we will assess their suitability as diagnostic antigens using our reporter system. This study is expected to enable development of a novel IgE- based serological diagnosis of PCC, which will be helpful for reliable identification of *T. solium* infection in endemic countries.

Keywords: Allergens, diagnosis, IgE, NPY-mRFP, porcine cysticercosis, RBL, reporter system, *Taenia solium*

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