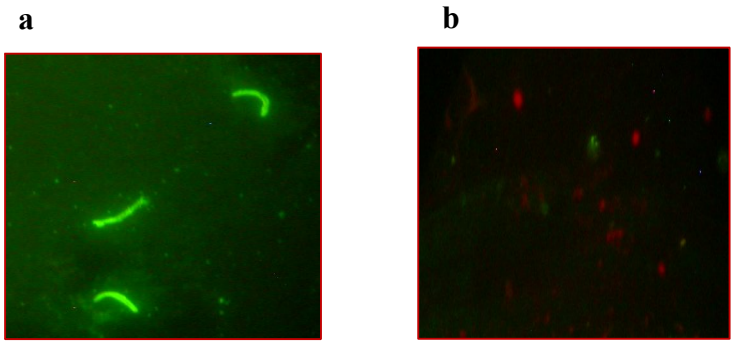

Additional file 1: Figure S2

Representative photographs of indirect immunofluorescence antibody test (IFAT). IFAT was performed to assess the ability of anti-rPfCelTOS sera of the immunized mice to recognize the native form of the CelTOS antigen on the sporozoite surface of *P. falciparum*. Multi-spot slides (a kind gift from Prof. G. Snounou), which were coated with 20 µL/well of *P. falciparum* sporozoites were blocked with 20 µL of PBS 1× containing 2.5% BSA (Roche, Basel, Switzerland) in a dark humidified chamber at room temperature for 30 min. After the slides were washed three times with PBS 1× (pH 7.4), a dilution of anti-rPfCelTOS polyclonal mouse serum in PBS 1× (1:200) was added to the duplicated wells and incubated in a wet chamber for 60 min. After three-times washing with PBS 1× (pH 7.4), each well was covered with 20 µL of the fluorescein isothiocyanate (FITC)-labeled rabbit anti-mouse IgG (1:40) and Evans blue (1:100) and then left in a wet chamber for 45 min. Each slide was examined under a fluorescence microscope (Nikon E200, Tokyo, Japan) with an oil immersion objective (100×). The serum samples obtained from the normal mouse were used as negative controls. The representative pictures show the recognition of native form of PfCelTOS in the surface of *P. falciparum* sporozoite using polyclonal antibodies (1:200) produced in mouse against rPfCelTOS. (**a**) Pooled sera derived from the immunized mice with rPfCelTOS (groups 1–5). There was no recognition

of native protein on *P. falciparum* parasite by control mouse sera (**b**), indicating rPfCelTOS-specific responses.