

## **Abstract**

The genus *Flavivirus* is composed of about 73 viruses. Of these viruses, 34 are mosquito borne, 17 are tick borne, and 22 are zoonotic agents transmitted with now known vector. Forty species of the flavivirus family have been associated with human disease. Yellow Fever, Dengue Fever, and Japanese Encephalitis, are the most important arboviral infectious diseases across every continent and have been legitimized into Class I and Class III transmissible disease, must be reported, in current issue of Regulation of Prevention and Control for Transmission Disease at Taiwan. The specific aim in this proposal is generation of monoclonal antibodies (MoAb)s against four flaviviruses, including the emerging West Nile in addition to the three viruses mentioned above for CDC at Taipei.

In the third year of three-year proposal, BALB/c mice will be immunized by these 4 different flaviviruses and hybrids will be generated by fusion of myeloma cells and immunized splenocytes. After selection the virus-specific antibody-producing hybridoma in HAT culture medium, cloning will be done with limiting dilution method. The virus-specific (MoAb)s will be purified from ascites of NOD/scid mice injected intraperitoneally with hybridoma cells. The antigenic specificity and typing of immunoglobulin subclass of (MoAb)s will be characterized and optimal sandwich ELISA pairing between MoAbs will be crossmatched.

In the third year, 37 (MoAb)s were successfully generated against West Nile virus. However, five of these 37 (MoAb)s cross-react to all flaviviruses tested, and most of other mAbs not only recognize WNV but also cross-react to KUN virus. Only one is qualified as WNV-specific (MoAb)s, do not react any other flavivirus except E protein of WNV. Furthermore, western blot results showed that conformational epitopes are recognized by most (MoAb)s, linear epitopes only be recognized by 3 (MoAb)s, one linear epitope on NS-1 protein and other 2 on E protein. Nevertheless, the linear epitopes are also found cross-react to KUN virus.

In the third year, we also completed the work, characterization of 12 (MoAb)s against type 4 dengue (Den 4) virus, left from the 2nd. year. All of them react to NS-1 protein of Den 4 virus but one of the twelve recognized a linear epitope on NS-1 of all flaviviruses be tested. Conformational epitopes on NS-1 of Den 4 were recognized by other 11(MoAb)s, Two of the eleven (MoAb)s showed cross-react to all 4 types of dengue viruses and the other 9 are Den 4-specific.

**Keywords : FLAVIVIRUS ; EPIDEMIOLOGY ; MONOCLONAL ; ANTIBODY**