



ADAMTS13

ADAMTS13 is a von Willebrand factor (VWF)-cleaving protease. It controls the VWF multimer interaction with sub-endothelial collagen and platelets by cleaving VWF at the A2 domain. Its deficiency can cause by abnormal VWF-platelet microthrombi formation in vasculatures of various tissues or organs, which results in a disease called Thrombotic Thrombocytopenic Purpura (TTP), a life-threatening condition with a mortality of 80-90% if not promptly treated. Since a markedly decreased ADAMTS13 activity (<10%) is considered characteristic for TTP, clinical laboratory ADAMTS13 is critical for diagnosis and management of patients with TTP.

In general, based on the differences in VWF substrates, ADAMTS13 assays can be grouped into three types. The first type uses full-length VWF. The cleavage of high- molecular-weight vWF multimers by ADAMTS13 can be assessed by western blot or a variety of von Willebrand factor activity assays, such as a collagen binding assay. The second type uses a recombinant VWF A2 domain fragments as the substrate. Cleaved A2 domain fragments are detected by ELISA assays. The third type of ADAMTS13 activity assays uses a recombinant VWF peptide, which contains the ADAMTS13 cleavage site between Y1065-M1606 in the A2 domain. The cleavage of this peptide can be measured by either a fluorescence resonance energy transfer (FRET) technique. The FRET assay uses a pair of fluorescent donor (A2pr[Nma]) and quencher (A2pr[Dnp]) molecules that are covalently attached to adjacent amino acids close to either side of the Y1065-M1606 (Q1599 and N1610) cleavage site.

When the peptide is intact, the fluorescent signal emitted from Nma is completely quenched by Dnp. The cleavage of VWF73 by ADAMTS13 causes a separation of the FRET pair and consequently a full release of fluorescent signal from Nma. The ADAMTS13 activities can be quantified by either a kinetic or an endpoint method.

The 2016-A mailing is the first CAP ADAMTS13 Survey. About 20 laboratories participated in this Survey. Approximately 16 laboratories used FRET method and 3 laboratories used /ELISA method. Though sample size is small (n=21), there are no apparent differences in performance of these two methods. Both CGS7-01 and -03 are normal-pooled plasmas and all participating laboratories correctly interpreted their results as normal. CGS7-02 is a specimen with mildly decreased ADAMTS13 activity and 18 out of 21 laboratories interpreted their results as abnormal. Of the three CGS7 specimens, 10 laboratories indicated that ADAMTS13 inhibitor screen was unnecessary, and all 4 laboratories that performed inhibitor screen assay got negative results. None of the participating laboratories performed Bethesda titer assay.

Conclusion, the first ADAMTS13 proficiency Survey went well, and distribution of the ADAMTS13 activity results are tight. However, Bethesda titer assay could not be assessed in this survey due to the specimen nature, which hopefully will be addressed in future ADAMTS13 Surveys.