Protein C assays: an analysis of North American Specialized Coagulation Laboratory Association proficiency testing

Jason Baron¹, Catherine Hayward², Stephen Johnson³, Elizabeth Van Cott¹
¹Massachusetts General Hospital, Boston, MA, United States/²McMaster University, Hamilton, ON, Canada/³Tufts Medical Center, Boston, MA, United States

Objectives:
Clinical coagulation laboratories use a variety of reagents and methods to assay protein C function and antigen. The purpose of this study was to evaluate the performance of these various methods and to gauge the relative frequency with which each is used.

Methods:
We analyzed proficiency testing results from three 2010 surveys conducted by the North American Specialized Coagulation Laboratory Association (NASCOLA), one of the major providers of proficiency testing for specialized coagulation tests. We aggregated results reported by individual laboratories using the same reagent.

Results:
The most common reagents for protein C antigen were Stago (35%), Corgenix (28%) and Helena Laboratories (17%); for protein C function (chromogenic) the most common were Chromogenix/IL (33%), Stago (25%), Precision Biologic (17%) and Siemens (17%) and for protein C function (clot-based) the most common were Siemens (38%), Precision Biologic (25%) and Stago (25%). For the first survey, the mean protein C result was 26% antigen, 24% chromogenic function and 29% clot function. All 46 laboratories (73 independent observations) correctly identified this sample as abnormal and the differences in levels as measured by various ELISA assays (inter-group range 22-28%), chromogenic assays (inter-group range 22-26%) and clot-based assays (inter-group range 21-31%) were not clinically significant. The number of laboratories using immuno-diffusion or immuno-electrophoresis assays (inter-group range 34-47%) was small, but nevertheless these methods provided somewhat higher antigenic results than the ELISA assays (mean ELISA 24%, immuno methods=42%; p<0.05, Student’s t-test, two-tails, unequal variance). Similarly, for the second survey, (mean results 21%, 23% and 24% by antigen, chromogenic and clot-based methods, respectively), and the third survey (mean results 46%, 49% and 50% by antigen, chromogenic and clot-based methods, respectively), no clear clinically significant biases between groups emerged, and all results were correctly classified as abnormal except for a very small number in the third survey, which evaluated a sample with a higher protein C level. For protein C antigen ELISA, inter-lab CV was as follows: Corgenix (35%, 24%, 12%, for surveys 1,2 and 3, respectively), Helena Laboratories (16%, 12% and 16%), and Stago (14%, 17%, 16%). For chromogenic function, CV estimates were Chromogenix/IL (13%, 12%, 4% across multiple manufacturer’s analyzer platforms), Precision Biologic (3%, 17%, 8% across multiple manufacturer’s analyzer platforms), Siemens (18%, 12%, 4% across a single analyzer platform), and Stago (6%, 3%, 5% across multiple analyzer platforms from the same manufacturer). For clot-based function, CV estimates were Precision Biologic (16%, 23%, 12% across multiple manufacturer’s analyzer platforms), Siemens (11%,
14%, 6% across multiple manufacturer’s analyzer platforms), and Stago (17%, 8%, 11% across multiple analyzer platforms from the same manufacturer). These CV values are across laboratories and in most cases across analyzer platforms; estimates for intra-laboratory CV would be expected to be considerably lower.

**Conclusions:**
We conclude that, although certain immune-diffusion or immuno-electrophoresis methods tended to run higher than other methods, all methods performed acceptably well in detecting low protein C and that most variation among different methods is likely to be clinically insignificant.