PROJECT REVIEW

“Blood Doping Screening through Capillary Electrophoresis”

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To facilitate the continued anti-doping efforts it is becoming important to make the best use of the limited analysis resources available for doping detection. The primary tools for doping analysis consist of equipment that requires highly skilled operators and the analyses are often expensive and/or time consuming. As such the number of samples that can be tested for any one event is limited; affording those doping a possibility of not being tested. To overcome this deficiency we propose a method to both increase the amount of testing and focus the high quality testing on those samples most likely to be actual doping cases.

This will be accomplished through the use of highly rapid capillary electrophoretic separations; capable of identifying key indicators of blood transfusion and other methods of enhancing oxygen delivery in the blood. The benefits of capillary electrophoretic separations are numerous; the high speed of the separations, the capability of having portable instruments and the low cost of operation. Capillary electrophoresis based separations can act as frontline screening systems to identify those blood samples that present signs of doping; flagging them for further analysis, while eliminating the bulk of the clean samples from further required testing. The advantage in the size of the capillary electrophoretic systems is the ability to perform multiple analysis with as little as a single drop of blood from the athlete; a much less invasive and rapid sampling method.

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Results and Conclusions

The objective of this project is to investigate how effectively capillary electrophoresis (CE) can be applied to the analysis of athlete blood samples for the identification of blood dopants. This work has specifically targeted three blood dopants: autologous blood transfusions, hemoglobin based oxygen carriers (HBOCs), and perfluorocarbon emulsions (PFC). The benefits of working with CE include the ability to perform very rapid separations, and the versatility to handle both molecular samples (i.e. HBOCs) and cellular samples (i.e. red blood cells). Furthermore, the benefits of CE separations can be translated from bench-top instruments to lab-on-a-chip devices, for what is often termed “point of care” analyses. This could be highly advantageous in anti-doping analyses, where the samples could be rapidly tested with instrumentation brought to the site of the athletic competition, likely reducing the delay in detecting those using performance enhancing agents.

For our work, we have focused on the use of traditional, bench-top, CE instruments, as they provide the greatest flexibility in method development. In investigating the three above mentioned blood dopants we have been able to successfully apply CE to two of the three dopant methods. The one method that has been found to be incompatible with CE analysis were the PFC; we found that their inability to remain suspended in a blood sample lead to difficulties in obtaining reproducible injections into the CE. Furthermore, their significantly greater density than red blood cells (RBCs) allows for their rapid, initial identification simply through centrifugation, rendering CE analysis superfluous. The remaining dopants, HBOCs and autologous blood transfusions, have shown much better success in detection through CE. In this past year our work on the detection of HBOCs, mixed into fresh blood samples in vitro, was published in Electrophoresis (DOI:10.1002/elps.201100506). In that work we were able to detect the presence of HBOCs at concentrations down to 5.5 g/L of whole blood, an amount below a 5% increase in total hemoglobin concentration. The CE method that we have developed for the detection of autologous blood transfusions continues to show great promise. This analysis is based on the different electrophoretic mobilities experienced by RBCs of different sizes. As circulating RBCs in athletes tend to be newer, larger cells, whereas RBCs taken from storage tend to be substantially smaller, we are able to exploit the differences in size and mobility of the RBCs to identify the presence of RBCs from storage.