

Verification of Erythropoiesis Stimulating Agents analytical method (sensitivity and specificity) in a second laboratory

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Project overview

Recent work in the anti-doping field has clearly shown that the future of blood collections is dependent on direct capillary blood collection, moving away from traditional venous draws. In that light, this project will focus only on the detectability of endogenous (blood EPO, or bEPO) and recombinant EPO (rEPO) in samples collected directly with capillary collection devices and will avoid venous blood spotted onto a matrix. Blood samples, both from control volunteers and patients who have been administered rEPO, will be collected using Tasso and OneDraw devices, and finger pricks.

Extraction of blood (and EPO) from the spot will be optimized using in-house protocols. Once extracted, three methods of EPO purification will be attempted. First, a conventional magnetic bead immunopurification method utilizing anti-EPO antibodies to capture the EPO in the dried blood sample will be tested (adapted from Desharnais, 2017). Next, the commercially available MAIIA EPO purification gel kit will also be utilized for efficacy. And finally, the StemCell ELISA plate will be assessed for immunopurification (adapted from Reverter-Brnachat, 2018). Once purified, samples will be analyzed via SAR-PAGE and Western Blotting, using the biotinylated monoclonal anti-EPO antibody described in previous Cologne Workshops (Reichel 2018, Dehnes 2020) to provide a higher quality protein signal. Using the methods described above, the sensitivity and specificity of bEPO and rEPO in DBS will be characterized.