

PROJECT REVIEW

"Overall approach for blood transfusion detection (autologous/homologous)"

J. Segura, R. Ventura, R. Abellan (Fundacio IMIM, Barcelona, Spain), **O. Fornas**, (Universitat Pompeu Fabra, Barcelona, Spain), **M. Lopez** (Hospital del Mar, Barcelona, Spain)

Homologous blood transfusion is at present detectable by flow cytometry methodology applied to blood samples. Nevertheless, blood sampling for antidoping control is carried out only in some selected situations and in no way can be considered a universal screening approach. The existence of a methodology applicable to urine to suspect of potential blood transfusion will be of enormous benefit for the fight against doping.

In the other hand, autologous blood transfusion remains elusive to detection. However, if evidence of blood transfusion of any type could be obtained but signs of homologous transfusion be absent, the conclusion of autologous blood transfusion would be a direct outcome.

The present project aims at developing screening and confirmatory methods able to detect a blood or red blood cells transfusion process to a given individual. The screening approach is based on the search in urine of plasticizers known to be leaked from blood bags towards the liquid and solid portions of bags contents and to be incorporated to donor's body after transfusion. The presence of unusually high amounts of metabolites of plasticizers in urine would alert on the use of a potential prohibited method.

Subsequent blood analysis could confirm the transfusion process by different approaches. As an extension of the screening procedure, unchanged plasticizer in plasma and red cells membranes would confirm the screening finding. Also residues of solvents used for cryogenic storage of red blood cell will be investigated for that kind of blood preservation. Additionally, blood analysis will allow to ascertain the presence of aged red blood cells deteriorated during the blood storage process. Accordingly, several markers known to be related to red cells aging will be studied by flow cytometry of red blood cells and those more sensitive be proposed as physiological confirmatory measurements. In conjunction with the present flow cytometry method to detect homologous blood transfusion, discrimination of type of transfusion appears feasible.

In summary, screening and confirmatory methodologies of general application to detect blood transfusion appear feasible to be obtained through the present project.

"Overall approach for blood transfusion detection (autologous/homologous)

J. Segura, R. Ventura, R. Abellan (Fundacio IMIM, Barcelona, Spain), **O. Fornas**, (Universitat Pompeu Fabra, Barcelona, Spain), **M. Lopez** (Hospital del Mar, Barcelona, Spain)

Results and Conclusions

No detection method for autologous blood transfusion exist, but the results recently obtained in the project suggest us that DEHP metabolites could be specific markers for the detection of transfusions (homologous or autologous).

An extraction and detection method has been developed for quantitative determination of DEHP metabolites in urine. Liquid-liquid extraction has been used and the compounds were unequivocally and efficiently detected by UPLC MS/MS in very widely concentration range. Much higher concentrations of DEHP metabolites have been detected in samples of transfused individuals compared to the general population. The majority of the samples analyzed from sportsmen behave as non exposed subjects.

In summary, the screening methodology proposed and the results obtained indicate that the procedure, to be carried out primarily in urine, could be applicable to all urine samples submitted to doping control. Thus, the approach is a promising tool for detecting of the use of blood transfusions (autologous, homologous) in sportsmen, being a fast and efficient method

Publications

1. Monfort N., Ventura R., Latorre A., Belalcazar V., López M., Segura J., Phthalate exposure: new clue to suspect illicit blood transfusion, *Recent advances in Doping Analysis*, 2010, 17:147
2. Monfort N., Ventura R., Latorre A., Belelcazar V., López M., Segura J., Urinary di-(2-ethylhexyl)phthalate metabolites in athletes as screening measure for illicit blood doping: a comparison study with patients receiving blood transfusion, *Transfusion*, 2010, 50:145-149