

Highlights of Analytical Sciences in Switzerland

Division of Analytical Sciences

Longitudinal Monitoring of Endogenous Blood Steroids as a Tool to Detect Testosterone Abuse in Sport

Federico Ponzetto^a, Julien Boccard^b, Norbert Baume^a, Serge Rudaz^b, Martial Saugy^a, and Raul Nicoli^{*a}

*Correspondence: Dr. R. Nicoli, E-mail: raul.nicoli@chuv.ch; *Swiss Laboratory for Doping Analyses, Chemin des Croisettes 22, CH-1066 Epalinges; ^bSchool of Pharmaceutical Sciences, University of Geneva, Boulevard d'Yvoy, CH-1211 Genève

Keywords: Doping · Longitudinal follow-up · Steroid profiling · Steroidal passport · Testosterone

Testosterone, as the major sex hormone secreted by endocrine glands, constitutes the key male endogenous anabolic androgenic steroid. Nowadays, this compound still represents one of the most widely used doping agents in strength and endurance sports, mainly due to its anabolic action on skeletal muscles. Quantification of testosterone and its major precursors and metabolites in an athlete's urine samples by GC-MS(/MS) is routinely achieved in anti-doping laboratories. To detect testosterone abuse in sport. the most sensitive biomarker is obtained by the glucuronidated testosterone over epitestosterone (T/E) concentration ratio. The latter is monitored over time using 'intra-individual reference values' for a more accurate evaluation of abnormal fluctuations that may indicate steroid misuse. This approach constitutes the so-called 'steroidal module' of the Athlete Biological Passport.

To overcome well-known weaknesses of this 'urinary steroid profile', such as genetic polymorphism (lack of enzyme responsible for testosterone glucuronidation), the monitoring of biosynthetic and metabolic pathways of testosterone in blood matrix could constitute a complementary approach. In this research, pills and patches of testosterone have therefore been administered to healthy volunteers and, after serum sample collection, selected endogenous blood steroid levels were monitored by LC-MS(/MS) with the aim of highlighting relevant biomarkers of exogenous testosterone abuse in the blood matrix. By applying multivariate data analysis, testosterone and especially dihydrotestosterone concentrations in serum were highlighted as significantly influenced by testosterone administration. Monitoring these two hormone levels in a longitudinal manner demonstrated that



intra-individual reference values were clearexceeded 1v following testosterone administration compared to negative control (same person in the absence of administration).







Steroidogenesis: processes by which cholesterol is converted to biologically active steroid hormones.

Results of this work show how the future implementation of a longitudinal follow-up of endogenous steroids in the blood matrix could increase the capabilities of antidoping laboratories for detecting exogenous testosterone administration.

Received: December 14, 2015

Reference

F. Ponzetto, F. Mehl, J. Boccard, N. Baume, S. Rudaz, M. Saugy, R. Nicoli, Anal. Bioanal. Chem. 2016, 408, 705



Example of a longitudinal profile in serum in the presence (red) or absence (green) of testosterone patch administration.