"Confirming the synthetic origin of perdnisolone/prednisone by GC-C-IRMS"

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Prednisolone and prednisone are two glucocorticosteroids included in the section S9 of the WADA Prohibited List of substances and methods. Glucocorticosteroids are prohibited in competition when administered by oral, intravenous or rectal routes. A reporting level of 30 ng/mL has been established by the WADA in the attempt to disclose permitted from illicit administration of GC.

Urine samples used for Antidoping purposes are not collected under sterile conditions. This implies that depending on the healthy conditions of the athlete, quality of collection material used and transport conditions to the laboratory, microorganisms’ growth may occur. The metabolic activity of the microorganisms has several impacts on the validity of the sample including the interpretation of the results. Antidoping laboratories have been dealing since many years with “active” urines where atypical findings have been ex-vivo in the urine sample by the microorganisms’ activity. For a correct identification of the origin of such findings into, the application of isotope ratio mass spectrometry (IRMS) has become mandatory.

More recently, the unusual presence of prednisone/prednisolone has been observed due to steroid 1-dehydrogenase activity of the microorganisms over naturally occurring compounds present in urine (cortisol or cortisone). For prednisone/prednisolone findings between the reporting level and 60 ng/mL, in addition to verify markers of microbial activity and the presence of specific metabolites (20b-hydroxy metabolite), WADA recommends to use GC/C/IRMS analysis to confirm the exogenous origin. In order to close the debate, the aim of this study is to develop and validate a GC/C/IRMS method to confirm the origin of prednisone/prednisolone in human urine in samples collected during Antidoping sessions and showing prednisone/prednisolone concentrations between 30 and 60 ng/mL, without modifying the structure of the compounds of interest, fulfilling WADA requirements.