

“Influence of athletes’ hyperhydration on sample collection procedure in terms of urine pharmacokinetics representative prohibited”

C. Georgakopoulos, M. Lyris, Y. Angelis, H. Dimopoulou, E. Sgouraki, P. Macheras (Doping Control Laboratory of Athens, Greece)

PROJECT REVIEW

Some athletes consume high volumes of drinks before the anti-doping sample collection in order either to recover from dehydration body conditions, which is normal after competition in high temperature and humid environmental conditions, or to dilute the urine and try to manipulate and mask detection of doping agents, where dilution as practice cannot be prohibited. Currently, for the International Standard of Testing, the suitable specific gravity (sg) of the doping control samples is 1.005 or higher measured with a refractometer, or 1.010 or higher with lab sticks.

Inappropriate sg for urine during sample collection procedure is faced by the collection of multiple samples until the Code requirement of the sg is met. Even if, the current specification of the appropriateness of the sg is scientifically valid, extensive knowledge on pharmacokinetics of hyperhydrated urine samples and subsequent influence on the detection of doping agents is lacking.

The goal of the current project is the performance of a series of pharmacokinetic studies of three different doping agents under various controlled hydration conditions to compare and examine the hydration influence in the urine excretion profile. Relating the hydration status to the pharmacokinetics of the three doping agents will allow drawing conclusions for the additional measures, if necessary, that can be taken during sample collection against the delivery of diluted urine that probably can mask or disturb the detection of the use of prohibited substances.

Results and Conclusions:

Hyperhydration effect on the endogenous androgenic anabolic steroids (EAAS) concentration levels was clearly demonstrated in the present study based on the individual and study volunteers population data. No significant difference was observed between the two hyperhydration agents with the water and Gatorade to act similarly on the urinary ‘steroid profile’ markers. The conventional WADA applied SG-adjustment method can eliminate the dilution induced effect and correct the EAAS concentrations by adjusting approximately to the baseline values. All the steroid ratios included to the urinary ‘steroid profile’ were remained unaffected by the hyperhydration due to the homeostasis of the steroid biosynthesis with the variability to be within 30% for the majority of data. No interference on the detectability of the selected

transitions was observed due to the dilution of the samples after hyperhydration. The evaluation of the collected data using the steroid module of the ABP would be performed in order to examine in which extent the individually calculated thresholds of the ABP software are exceeded under hyperhydration conditions.

As final statement: the use of hyperhydration as a masking procedure by altering the urinary 'steroid profile' is not effective. Although, excessive fluid intake can alter the urinary 'steroid profile' markers, the steroid ratios remain unaffected and the SG-adjustment method officially used by WADA can fully eliminate any effect caused by dilution.

Hyperhydration induced decrease in LH urine concentration levels can be eliminated by adjusting the measured concentrations. Since reduced LH concentration levels may serve as an indicator for steroid abuse, adjustment of LH concentrations would be useful for the urinary 'steroid profile' evaluation. The WADA applied SG adjustment method can compensate the dilution induced effect and correct the LH concentrations by adjusting them close to the baseline values. Based on the results of the present study, SG-adjustment of LH concentrations in all the samples, as it is applied on the endogenous 'steroid profile' markers, could be a useful practice even if the low LH threshold is not applicable in the WADA TD2017.

In the present study, hyperhydration induced by two different agents (water and Gatorade) showed no significant changes on the hematological module of the ABP at any time during the study. The magnitude of difference between pre- and post-hydration values was too small and not statistically different 30 min after the ingestion independently of the hyperhydration agent. The evaluation of the collected data using the hematological module of the ABP has been requested from WADA in order to examine the extent that the individually calculated thresholds of the ABP software are exceeded under hyperhydration conditions.

Based on the serum PK profiles as well as on SAR-PAGE analysis and analyte mobility of both urine and serum samples no effect of hyperhydration was observed under the conditions examined on the present study in the anti-doping urine and blood analysis of rHuEPO.

Urine concentrations of budesonide and its metabolites were affected due to dilution after hyperhydration leading to increased percentage of samples below the MRPL of 30 ng/mL. However, WADA SG-adjustment based on the equation of Levine-Fahy was able to compensate the dilution effect by adjusting the concentration values closed to the baseline and therefore, decreasing the % of samples below the MRPL.