

“Novel strategy for the detection of new long term AAS metabolites by low energy EI GC-QTOF”

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Project Overview

The identification of anabolic androgenic steroids (AAS) is a vital issue in doping control. Due to the performance enhancing properties of AAS, the World Anti-Doping Association (WADA) banned their use but according to the annual report of WADA, steroids are still very popular amongst athletes and are responsible for 40% of all adverse analytical findings. The search for metabolites with longer detection times remains an important task and the introduction of new long-term metabolites for exogenous AAS such as for example stanozolol, methanedione and dehydrochloromethyltestosterone, led to a 4 - 80-fold increase of adverse analytical findings due to the prolonged detection time.

Recently, our laboratory developed a new strategy for finding new long-term metabolites. The approach uses low energy EI GC-QTOF and it is applicable to a wide range of different anabolic steroids. The technique has been tested in a proof-of-concept setting and the combination with a robust polar column allows the analysis of both derivatized and non-derivatized steroids, further expanding the application window and increasing the chances of finding new long-term metabolites.

Results and Conclusions:

A search for new metabolites was performed by applying the low energy EI GC-QTOF product ion scan strategy. To increase the chances of finding new long-term metabolites this strategy was applied using different (complementary) approaches: Analyses of both the derivatized and non-derivatized steroid form in the glucuronidated, sulfated and free steroid fraction.

The main conclusion is that the strategy suffers from a lack of sensitivity that cannot be sufficiently compensated by the increased selectivity. Unfortunately, this meant that the procedure proved to be inferior to the MRM CI GC-MS/MS approach for detection of new metabolites (WADA project 16A01MP). Consequently, no new metabolites could be found within this study and the approach failed to find many of the metabolites that were detected with the MRM CI GC-MS/MS detection protocol.

However, during this study we did acquire more knowledge on the analysis of sulfated steroids on GC-MS and this provided us with some new insights on

their GC-MS behavior, prompting us to pursue a more in-depth study of directly injecting non-hydrolyzed sulfated steroids on GC-MS. We are convinced that this direct injection strategy has great potential to, in the future, lead to the discovery of new long-term metabolites and/or will allow the inclusion of long-term sulfated metabolites in a general GC-MS initial testing procedure.