Project Review

"Hydropyrolysis as a preparative technique for compound-specific carbon isotope ratio measurement of endogenous steroids"

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The use of anabolic steroids in sport has been banned since the 1960s and anabolic androgenic steroids are named on the WADA 2006 prohibited list. Yet the abuse of these substances can be difficult to detect. No more so than when steroids such as testosterone that are found naturally in the body are used to top up normal levels. Recent advances have enabled the discrimination of natural from administered molecules because the type of carbon in the body's molecules reflects the carbon ingested as part of an athlete's diet.

Drugs manufactured in the lab contain very different carbon, allowing the two types of molecules to be distinguished by scientific equipment. The dissimilarity is present in the relative abundance of stable isotopes of carbon (carbon-12 and carbon-13) which can be detected by modern instrumentation. In their natural form, however, the body's molecules react too aggressively with the laboratory equipment designed to separate them for accurate measurements to be made. Some methods overcome this problem by adding carbon to the target molecule, but this irreversibly overprints the carbon source 'signal'.

Hydropyrolysis is a newly-applied catalytic technique that delicately strips molecules of their chemically aggressive parts but retains the carbon skeleton intact, allowing easy detection of the carbon source. Free from conventional analytical problems, the new detection system will allow scientists to pinpoint banned substances in an athlete's system more accurately.

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Results and Conclusions

Pilot project advances include the development of a universal deconjugation technique that removes both glucuronide and sulfate moieties from steroids. This procedure is relatively rapid (2 hours) taking ten times less than some enzyme based methods. When it is considered that the process also defunctionalizes the steroids removing the need for derivatization prior to GC, the rapidity appears even more remarkable.

The development of a low temperature platinum catalyst that shows close to 100% conversion efficiency and a high degree of product specificity can produce one or a small number of isomers from functionalized starting materials. The high conversion has an impact on technique sensitivity and the selectivity enables ease of analysis. Minimal carbon isotope fractionation during all procedures ensures that carbon isotope ratios for the hydropyrolysis products are faithful representatives of the steroid carbon isotope ratios.

Publications

Articles

Meredith, W. Snape, C.E., Cooper, M, Gomes, R.L. and Sephton, M.A. (Submitted) Hydropyrolysis over a platinum catalyst as a preparative technique for compound-specific carbon isotope ratio measurement of endogenous steroids. Rapid Commun. Mass Spectrom.

Gomes, R. L., Meredith, W., Sephton, M. A., and Snape, C. E., (Submitted) Hydropyrolysis as an alternative to deconjugation and derivatisation for steroid conjugate determination by GC/C/IRMS. Rapid Commun. Mass Spectrom.

Gomes, R. L., Meredith, W., Sephton, M. A., and Snape, C. E., (Submitted) Analysis of conjugated steroid androgens: preparative approaches and associated issues. Anal. Bioanal. Chem.

Presentations

Gomes, R. L. (2008) Determination of steroid conjugates by hydropyrolysis-GC/C/IRMS, in London Biological Mass Spectrometry, London, UK. Invited Speaker.

Meredith, W., Snape, C. E., Sephton, M. A., and Love, G. D. (2008) Hydropyrolysis as a preparative method for the compound specific carbon isotope analysis of steroids, in 23rd International Meeting on Organic Geochemistry 9th -14th September 2007, Torquay, Devon, United Kingdom.