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Identification of Metabolically Stable Selective Androgen Receptor Modulators (SARMs) in Human Urine

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Extended abstract*

A novel class of therapeutics presumably complementing anabolic steroids in the near future contains so-called selective androgen receptor modulators (SARMs) that have been under clinical investigations for several years. Although not commercially available yet, their potential for misuse in sports is high as serious shortcomings of anabolic-androgenic steroid therapies such as decreased levels of HDL cholesterol and negative influences on prostate and cardiovascular systems have been eliminated. SARMs have proven to act as full agonists in anabolic target tissues (e.g. muscle and bone), but demonstrated only partial agonist activity in androgenic tissues such as prostate and seminal vesicles. Their main advantage over steroids in testosterone replacement therapies is the fact that they do not represent substrates for 5α-reductases, one main route of metabolism of steroids related to testosterone. The resulting metabolic product of testosterone, dihydrotestosterone (DHT), is considered a more potent androgenic steroid than testosterone itself, and due to the significant amount of DHT locally produced in organs such as the prostate, DHT is believed to amplify the androgenic activity of testosterone. This effect is excluded by synthetic SARMs that are not amenable for this particular metabolic pathway. Currently, four classes of SARMs are under clinical investigation and can be categorized by their chemical structures into 1) aryl-propionamide, 2) bicyclic hydantoin, 3) quinoline, and 4) tetrahydroquinoline analogues[1].

*for further details please refer to:

Thevis *et al*. Rapid Commun Mass Spectrom (2006), **20** : 870

In the present study we describe an assay to determine three selected aryl-propionamide-derived SARMs and related compounds in spiked urine specimens using liquid chromatography – electrospray ionization – tandem mass spectrometry (LC-MS/MS) employing multiple reaction monitoring and simultaneous precursor ion scanning. Four aryl-propionamide-derived SARMs (**1**-**4**, Figure 1) were chemically synthesized[2-4] in order to establish a screening procedure, and all compounds were characterized by high resolution/high accuracy mass analysis.

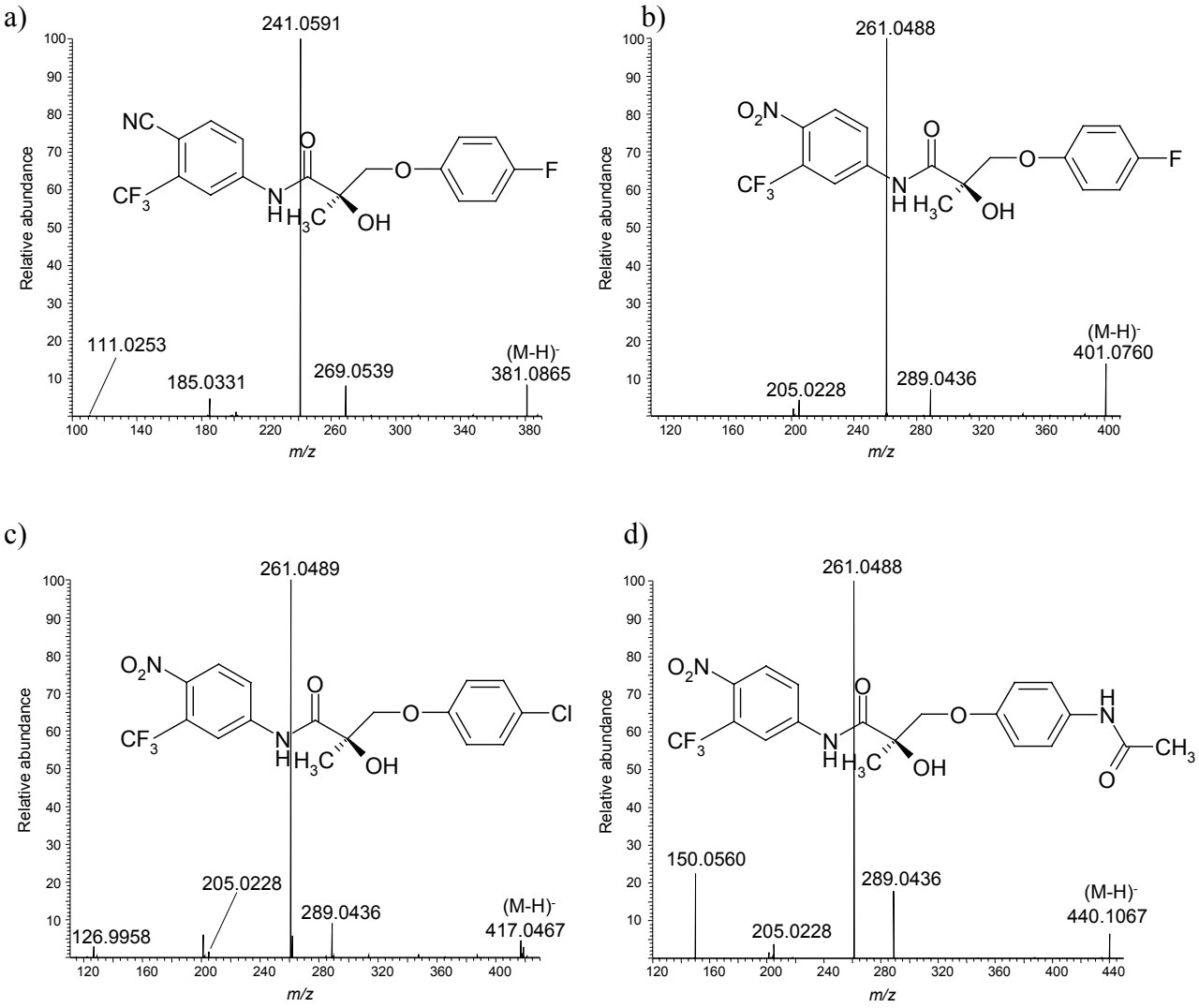

Figure 1: ESI product ion spectra of four model aryl-propionamide derived SARMs analyzed using an LTQ-Orbitrap mass spectrometer at a resolution of 60.000. Compounds **1**-**3** (a, b and c) were used for assay validation purposes while compound **4** (d) was employed to test the efficiency of precursor ion scanning in screening analyses.

Routine analyses for model SARMs were conducted after solid-phase extraction using PAD-I adsorber resin employing a triple quadrupole mass spectrometer. Characteristic product ions obtained from deprotonated precursor ions $(M-H)^-$ by collision-induced dissociation were found at *m/z* 289 and 261 as well as *m/z* 269 and 241 representing the bisubstituted aniline residues of selected model compounds. Assay validation was performed regarding lower limit of detection (1 ng/mL), recovery (85-105%), intraday precision (7.6-11.6%) and interday precision (9.9-14.4%). Precursor ion scan experiments on diagnostic product ions enabled the detection of compound **4** at 50 ng/mL.

Acknowledgments

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References

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