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Influence of AOD-9604 on the WADA hGH isoform immunoassay

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Abstract

AOD-9604 is a modified peptide fragment of the C-terminal region of human growth hormone (hGH). It consists of 16 amino acids, is a peptide hormone of 1815 Da, and corresponds to the amino acids 177 to 191 of hGH bearing an additional N-terminal tyrosine. AOD-9604 is reported to stimulate lipolysis and inhibit lipogenesis and was developed for the treatment of obesity. Recently AOD-9604 was ascribed to the S.0 category by the World Anti-Doping Agency (WADA) and is therefore prohibited in sports. To which extent the use of this peptide leads to detectable amounts in human urine or plasma is yet unknown. An unlabeled vial obtained from the environment of elite athletes was investigated with LC-Q-TOF/MS-techniques and AOD-9604 was identified as the main ingredient, demonstrating that the substance has found its way into the world of sport. Besides its potential as doping agent, its possible influence on the WADA hGH isoform immunoassay was to be assessed since the drug candidate is a fragment of hGH and thus could potentially interact with antibodies employed in the isoform test. Different concentrations of AOD-9604 were tested on Kit 1 and Kit 2 of the hGH isoform immunoassay. Serum samples containing recombinant hGH, negative serum sample and human zero serum (supplied by the manufacturer of the assay) were tested for the influence of AOD-9604 on the different antibodies of rec- and pit-assay of Kit 1 and Kit 2. First results showed no binding of the fragment to the antibodies of the assay (no increase of the concentration of the negative sample and the human zero serum) and no ‘false positive’ results were generated. Further, the peptide did not affect the test results of adverse analytical findings of hGH in serum samples and is therefore not of concern regarding the detection assay for hGH in doping controls.