Project Review

"Detection of Indirect Androgen Doping with a GnRH Analog (Leuprolide)"

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This study evaluates the threat for sports doping of a class of hormones called gonadotrophin releasing hormone (GnRH) analogs. These are synthetic, small peptide superactive analogs of the natural hypothalamic hormone GnRH, a decapeptide which is a major regulator of the reproductive system. GnRH stimulates pituitary LH and testicular testosterone (T) secretion but exogenous use of both LH and T are banned in sports. Consequently, GnRH and its analogs are also prohibited in sports for their potential to act as doping and/or masking agents.

Used as intended for prolonged periods, GnRH analogs suppress gonadal function in hormone dependent diseases like breast or prostate cancers. However, when used for short periods they stimulate gonadal function, creating a so-called "flare" reaction, before suppression sets in. This "flare" reaction could be used by athletes for indirect androgen doping by stimulating the body's own LH and testosterone secretion to unnaturally high levels. So far, although GnRH analogs are banned and their use by athletes is suspected, there has been no detailed evaluation of how GnRH analogs affect sports doping tests, when to suspect their use and how to detect them in urine specimens.

This study will examine in detail the hormonal effects of a superactive GnRH analog (leuprolide) on serum and urine LH and T when used for short periods, and when repeated with a drug-free interval and well as with an androgen-suppressed gonadal axis. Our preliminary evidence proves major hormonal effects are produced so a detection test for this GnRH analog is required. We will develop suitable LC/MS/MS methodology, apply it to the clinical study and to a set of urine samples from athletes with high-normal urine testosterone but normal T/E ratio where the suspicion of GnRH analog use might be highest.

Results and Conclusions

Non-steroidal drugs that increase endogenous testosterone may be used to exploit ergogenic effects of androgens in power sports. While superactive GnRH analog use is suspected, neither screening nor detection tests are developed.

Objective: To determine if (a) stimulation for 5 days by leuprolide of serum and urine steroids and urine LH is reproducible at a 2 week interval, (b) nandrolone decanoate (ND) co-administration masks responses to leuprolide administration, (c) performance of urine measurement of leuprolide and M1, its major metabolite, as a detection test.

Healthy men randomized into a 4 week parallel group, open label clinical study. Leuprolide (1mg) was injected sc daily for 4 days in 1st & 3rd week with hormone-free 2nd & 4th

weeks. In the 3rd week, men were randomized to either N decanoate injections or no extra treatment. Serum and urine steroids and urine leuprolide and M1 and LH.

Results: Leuprolide stimulated striking, reproducible increases in serum and urine LH and steroids (serum T, DHT, 3α diol; urine T, E & A). ND suppressed basal serum T, E2, 3α diol, and urinary E but did not mask or change the magnitude of responses to leuprolide. Urine leuprolide and M1 measurement had 100% sensitivity and specificity in detecting leuprolide administration up to one day after cessation of injections with the detection window between 1 to 3 days after last dose. Screening using urine steroid and LH measurements, optimally by urinary log10(LH x T), correctly classified 82% of urine samples.

Conclusions: Leuprolide stimulation of endogenous testosterone is reproducible after a 10 day interval, is not masked by ND and is reliably detected by urine leuprolide or M1 measurement for up to 1 to 3 days after administration.

Publications

Detection and effects on serum and urine steroid and LH of repeated GnRH analog (leuprolide) stimulation. Handelsman DJ, Idan A, Grainger J, Goebel C, Turner L, Conway AJ. J Steroid Biochem Mol Biol. 2014 May; 141:113-20. doi: 10.1016/j.jsbmb.2014.01.011. Epub 2014 Feb 2.