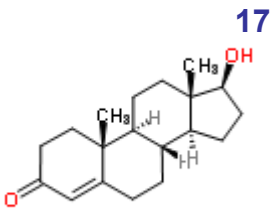
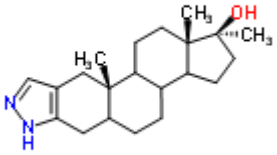
1. ANABOLIC STEROIDS

A. Steroid structures all contain the same tetracyclic nucleus, referred to as the cyclopentanoperhydrophenanthrene ring system, with substituents attached to the various rings, depending on the specific anabolic steroid. Anabolic steroids can be classified as non-esters or 17-esters.

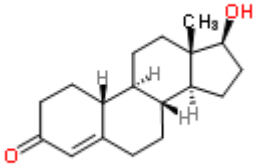
B. Structure, Empirical Formula, Molecular Weight



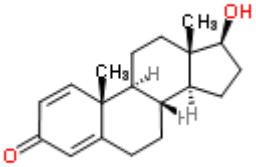
Testosterone
$C_{19}H_{28}O_2$
MW 288.4



Stanozolol
$C_{21}H_{32}N_2O$
MW 328.4



Nandrolone
$C_{18}H_{26}O_2$
MW 274.3



Boldenone
$C_{19}H_{26}O_2$
MW 286.4

C. Synonyms: Juice, roids

D. Trade Names: Winstrol®, Anadrol®-50, Oxandrin®, etc.

E. Drug Action: Anabolic-androgenic steroid

F. Common pharmaceutical/street forms: Common preparations include tablets, injectable oils and suspensions from various countries of origin.

G. Solubility: Methanol, ethanol, diethyl ether, chloroform, insoluble in water

H. Extraction:

(1) Dry extraction for tablets

(a) Determine amount to extract 5-10 milligrams of steroid from the dose.

(b) Grind up tablet(s) and transfer to a test tube.

(c) Add approximately 1 milliliter of methanol or other appropriate solvent (e.g. diethyl ether for stanozolol).

(d) Vortex. If emulsion occurs, centrifuge.

(e) Decant the methanol, filtering if necessary.

(2) Liquid/liquid extraction for oil solutions

(a) Determine amount to extract 5-10 milligrams of steroid from the dose (e.g. 1-2 drops of oil from a 300mg/mL solution).

(b) Add approximately 1 milliliter of C15 methanol.

(3) Liquid/liquid extraction for aqueous suspensions.

(a) Shake container vigorously to emulsify the suspension.

(b) Determine amount to extract 5-10 milligrams of steroid from the dose.

(c) Transfer the suspension to be extracted to a test tube.

(d) Add approximately 1 milliliter of chloroform.

(e) Vortex. If emulsion occurs, centrifuge.

(f) Remove the chloroform (bottom) layer, filtering if necessary.

(g) Evaporate the chloroform and reconstitute with C15 methanol.

(4) Stanozolol derivatization

(a) Extract as appropriate from above.

(b) Evaporate solvent.

(c) Dissolve in 100 µL caffeinated dimethylformamide (DMF).

(d) Transfer to GC/MS vial with glass insert.

(e) Add 100 µL BSTFA with 1% TMCS to GC/MS vial.

(f) Cap vial.

(5) Steroid deesterfication

(a) If using a powder steroid prepare a 20mg/mL solution of steroid in methanol. If using an oil based steroid take 2-3 drops of oil and add it to 1mL of methanol.

(b) In a test tube or GC vial add 100µL of powder steroid solution or 200µL of oil steroid solution.

(c) Then add 1.9mL or 1.8mL, respectively, of methanolic 0.2 NaOH (total volume with steroid solution 2mL)

(d) Mix and incubate (heat) @ 70˚C on heating block or 45-50˚C on analog heat block for approximately 30 minutes (this is accomplished by setting the low heat setting just below 4)

(e) Let cool

(f) Add 0.5mL deionized H2O and mix

(g) Add 1mL CHCl3 and mix

(h) Two layers will form (bottom layer is CHCl3) discard the H2O layer

(i) Dry the CHCl3 with anhydrous Na2SO4

(j) Remove CHCl3 and dry down

(k) Reconstitute with C15/MeOH and run on GC/MS using Drugs2

I. Chemical indicator tests:

(1) Liebermann's: See Table 1 in Chiong reference.

(2) H_2SO_4: See Table 1 in Chiong reference.

J. TLC:

(1) Mobile Phase: System 1: Methylene chloride:diethyl ether:methanol:water (77:15:8:1.2)

(2) Locator: Sulfuric acid/ethanol + heat, p-toluenesulfonic acid, DPST solution

K. GC/MS: Analyze using “Drugs2” program.

Note: If Stanozolol does not resolve sufficiently then it may require derivatization prior to analysis by GC/MS.

L. Comments: Check Mexican PDR if labels are in Spanish.

M. Report as: See Title 13 for list of specific controlled anabolic steroids.

Example:

Oxymetholone, a dangerous drug.

Testosterone Cypionate, an ester of testosterone, a dangerous drug.

N. References:

(1) Analytical Profiles of Anabolic Steroids, CND Analytical, Inc., Auburn, AL, 1989.

(2) Tolliver, J. M., (ed.), Anabolic Steroids, Microgram, Vol. XXIV, No.11, Nov. 1991, pp. 268-281.

(3) Tolliver, J. M., (ed.), Anabolic Steroid Products, Microgram, Vol. XXV, No. 5, May 1992, pp. 134-140.

(4) Grob, R. L., (ed.), Modern Practice of Gas Chromatography, 3rd Ed., John Wiley & Sons, Inc., 1995, pp. 708-711.

(5) Chiong, D. M. et al, The Analysis and Identification of Steroids, Journal of Forensic Sciences, Vol. 37, No. 2, March 1992, pp. 488-502.

(6) Drug Analysis Protocol, Los Angeles County Sheriffs Department, Los Angeles, CA.