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► **To cite this version:**

Pascal Kintz, Laurie Gheddar. Evidence of use of drostanolone, an anabolic steroid, at the time the subject committed a murder: Place of hair analysis. *Toxicologie Analytique et Clinique*, 2021, 33 (3), pp.222-225. <10.1016/j.toxac.2021.05.002>. <hal-03622309>

HAL Id: hal-03622309

<https://hal.science/hal-03622309v1>

Submitted on 16 Oct 2023

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Case report

Evidence of use of drostanolone, an anabolic steroid, at the time the subject committed a murder: place of hair analysis

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Declaration of interest: none

Abstract

A 21-year old male was arrested after killing a rival during a dramatic episode of violence, due to jealousy. The perpetrator was sent to prison, where he revealed, several months later, that he was using anabolic steroids at the time of the assault. The steroids were used to enhance his muscular mass. To document possible steroids abuse, a hair specimen (9 cm in length, black in colour) was collected 7 months after the event and segmented in 3 x 3 cm segments. A specific steroids screening procedure, involving both LC-MS/MS and GC-MS/MS, was used, which demonstrated the presence of drostanolone and its ester form, drostanolone enantate in the distal segment (i.e. the one corresponding to the period of the assault). Concentrations were 3 and 14 pg/mg for drostanolone and drostanolone enantate, respectively. [The etiology of this murder was finally considered to be due to an aggressive behaviour, which can have been enhanced by side effects due to anabolic steroids abuse.](#)

Key words

Violence, anabolic steroids, drostanolone, hair

Introduction

Athletes, but also amateurs, can use anabolic steroids because it has been claimed that these drugs increase lean body mass, strength, aggressiveness, endurance and lead to a shorter recovery time between workouts (1). Today, anabolic steroids are mostly available on the Internet, at low costs, both as oral or injectable forms (generally as esters). In most cases, these compounds are used in dosages much higher than what is recommended by the manufacturers. Therapeutic uses of anabolic steroids are very limited, and, for example, in France, only testosterone enantate (Androtardyl[™]) is available after medical prescription for intramuscular injection into the buttock.

Numerous side-effects after long-term anabolic steroids abuse have been described, and include increase in blood pressure, alterations of the cardiovascular system, liver toxicity, acne, hirsutism, gynecomastia, atrophic testes and sexual dysfunction (2). When abuse is prolonged, subjects can develop psychiatric effects, which include antisocial lifestyle, disinhibition, frustration, psychosis, low self-confidence, childhood conduct disorders, delirium, depression and aggressiveness (3-5).

Epidemiological studies, both in Europe (6) and in the United States of America (7) have demonstrated that the abuse of anabolic steroids constitutes an increased risk for violence and that there is an association between their use and criminality. There is an increased risk of developing an antisocial lifestyle but the frequency of crimes of violence and weapons offences is increased once anabolic steroids are consumed for a long period (8).

Because anabolic steroids are seldom tested in forensic laboratories, and even less in hair specimens, the present observation allows establishing a link between the aggressive behaviour of a murderer and his addiction to this class of compounds.

Case history

A 21-year old male was arrested after killing a rival during a dramatic episode of violence, due to jealousy. He was the new boyfriend of a girl, who was previously in couple with the victim. The perpetrator was sent to prison, where he revealed, several

months later, that he was using anabolic steroids at the time of the assault. The exact regimen of the subject is unclear and the steroids were not identified. He refused to indicate which compounds were used and how they were purchased.

A medical doctor was requested to collect a hair specimen about 7 months after the assault. The perpetrator only accepted to give one strand of hair, which was 9 cm in length and black in colour.

Toxicological analyses

Because the identity of the involved steroid(s) was not known, a screening procedure, already published (9) was used. Briefly, 30 mg of finely cut hair, previously decontaminated by 2 dichloromethane bathes (5 mL, 2 min) and segmented (3 x 3 cm) were weighted. The hair was mixed with 1 ng of testosterone-d₃, used as internal standard and 1 mL of methanol and submitted to an ultrasonic bath for a 90 min. Then, in one side, the organic phase was evaporated and diluted with phosphate buffer pH 7.0 (part A), and in another side, the hair remain was incubated with 1 mL NaOH 1 M (part B). Liquid-liquid extractions were operated on parts A and B with 2 mL ethyl acetate. Extracts were combined and evaporated to dryness. The residue was dissolved in 1 mL dichloromethane, which was submitted to solid phase extraction, using Isolute C18 columns. [The final elution phase was evaporated and TMS derivatization \(MSTFA/NH₄I/2-mercaptoethanol, 1000:2:5\) was achieved.](#) Drostanolone and drostanolone enantate were tested by GC-MS/MS (Agilent 7010B) (Table 1).

Results and discussion

The validation of the analytical procedures for drostanolone and drostanolone enantate in hair is presented in Table 2.

In the recent years, hair specimens have been proposed to document drug exposure in any situation in which a history of past rather than recent drug use is requested. In addition, keratinous matrices are the unique matrices allowing documenting long-term exposure to drugs. Finally, testing head hair by segmentation allows establishing a retrospective calendar of an individual's drug use and the distinction between single and repetitive use (10). Segmental analyses of the hair of the murderer went back positive for the distal end and allowed interpreting the results as steroid consumption

at the period corresponding to the assault, assuming a normal hair growth rate at 1 cm per month. Concentrations of drostanolone and drostanolone enantate were in the low pg/mg range, *i.e.* 3 and 14 pg/mg, respectively. Although concentrations were low, a suitable chromatographic signal was obtained in the **distal** hair segment (Figure 1). **The proximal segment and the segment of the middle were both negative for drostanolone and drostanolone enantate, roughly corresponding to the period the subject was in jail.** Testosterone concentrations were less than 1 pg/mg in the 3 segments, **which is the limit of quantitation of the method.** Anabolic steroids have been seldom reported in hair and, in most cases, bodybuilders have been tested. Only Thieme and Anielski (11), in their compendium presented concentrations for drostanolone and drostanolone enantate, which were 4 and 30 pg/mg in one case and 2 and 2 pg/mg in a second one.

Drostanolone, or dromostanolone, is a potent synthetic androgenic anabolic steroid. Currently, there is no human indication for drostanolone, either in the United States or in European countries. In the past, it has been indicated in postmenopausal women with recurrent breast cancer, in a combined hormone therapy.

As drostanolone has the potential to be misused for performance enhancement in sport, it can attract athletes looking for power and strength improvement. As a consequence, drostanolone has been prohibited by the World Anti-Doping Agency (WADA) at-all-times in the category of “anabolic agents” under section S1 of the WADA List. On the WADA 2019 testing figures report (12), there were 4180 adverse analytical findings (AAF) and among them, 163 were for drostanolone. This indicates that drostanolone is easy to obtain and is largely abused.

According to witnesses, the murderer was aggressive at the time of the assault. The test in an alternative matrix, *i.e.* scalp hair, allowed confirming long-term drostanolone abuse. This seems of major importance to establish a clear picture of drug involvement. However, the authors can accept that some parameters about the detection of drostanolone in hair have not been addressed: dose necessary to give a positive result, repeatability, robustness, contamination, carry-over, interferences, variable incorporation into hair, external factors that may have an impact, etc. The minimum dose that had to be ingested by a subject in order to get a “positive” detection in one of the hair sections is unknown.

Conclusion

Since many years, numerous publications have indicated that there is an association between the abuse of anabolic steroids and violence. Abusers of steroids are more prone to be involved in criminal acts, as demonstrated by epidemiological studies. However, in all these reports, none are supported by toxicological analyses, which seem to be the basis in forensic science. The authors have presented toxicological investigations in a murderer who was abusing drostanolone at the time of the assault. This situation appears to be highly suitable for the trial hearing by demonstrating that a prohibited and harmful substance was involved.

Table 1

GC-MS/MS mass spectrometric data for the tested compounds

Drug	Transitions m/z	Collision energy eV	Retention time min
Drostanolone	448.0 > 141.0	20	10.94
	448.0 > 156.0	20	
Drostanolone enantate	488.4 > 141.1	25	15.42
	488.4 > 156.1	15	
Testosterone-d3	435.4 > 209.1	15	10.89

Table 2

GC-MS/MS validation parameters in hair

Parameters	Drostanolone	Drostanolone enantate
Linearity	1 to 100 pg/mg, $r^2 = 0.9998$	5 to 100 pg/mg, $r^2 = 0.9992$
Limit of detection	0.5 pg/mg	2 pg/mg
Limit of quantitation	1 pg/mg	5 pg/mg
Precision (20 pg/mg, n=8)	16.2 %	17.9 %
Matrix effect (20 pg/mg, n=15)	< 20 %	< 20 %

References

1. Cadwallader AB, Lim CS, Rollins DE, Botrè F. The androgen receptor and its use in biological assays: looking toward effect-based testing and its applications. *J Anal Toxicol* 2011;35:594-607.
2. Van Amsterdam J, Opperhuizen A, Hartgens F. Adverse health effect of anabolic-androgenic steroids. *Regul Toxicol Pharmacol* 2010;57:117-123.

3. Trenton AJ, Currier GW. Behavioural manifestations of anabolic steroid use. *CNS Drugs* 2005;19:571-595.
4. Pope HG, Kouri EM, Hudson JI. Effects of supraphysiologic doses of testosterone on mood and aggression in normal men: a randomized controlled trial. *Arch Gen Psychiatry* 2000;57:133-140.
5. Khodoruth MAS, Khan AA. Anabolic steroids-induced delirium. *Medicine* 2020; doi.org/10.1097/MD.00000000000021639.
6. Lood Y, Eklund A, Garle M, Ahlner J. Anabolic androgenic steroids in police cases in Sweden 1999-2009. *Forensic Sci Int* 2012;219:199-204.
7. Beaver KM, Vaughn MG, Delisi M, Wright JP. Anabolic-androgenic steroid use and involvement in violent behaviour in a nationally representative sample of young adult males in the United States. *Am J Public Health* 2008;98:2185-2187.
8. Skärberg K, Nyberg F, Engström I. Is there an association between the use of anabolic-androgenic steroids and criminality? *Eur Addict Res* 2010;16:213-219.
9. Kintz P, Gheddar L, Raul J-S. Simultaneous testing for anabolic steroids in human hair specimens collected from various anatomic locations has several advantages when compared with the standard head hair analysis. *Drug Test Anal*, 2021, doi: 10.1002/dta.3020.
10. Kintz P. Hair analysis in forensic toxicology: an updated review with special focus on pitfalls. *Curr Pharm Des* 2017;23:5480-5486.
11. Thieme D, Anielski P. Doping, application of hair analysis. In, Kintz P, Salamone A and Vincenti M, eds. *Hair analysis in clinical and forensic toxicology*, Academic Press, London, 2015;pp275-299.
12. https://www.wada-ama.org/sites/default/files/resources/files/2019_anti-doping_testing_figures_en.pdf (Accessed 24 January 2021)

Figure 1

GC-MS/MS chromatogram obtained after extraction of the distal hair segment of the suspect. Drostanolone and drostanolone enantate concentrations were 3 and 14 pg/mg, respectively. From top to bottom: 2 transitions for drostanolone (11.03 min, m/z 448.0 > 141.0 and 448.0 > 156.0), 2 transitions for drostanolone enantate (15.51 min, m/z 488.4 > 141 and 488.4 > 156.1) and 1 transition for testosterone-d3 (10.92 min, IS, m/z 435.4 > 209.1)

