

Reprint from

RECENT ADVANCES
IN DOPING ANALYSIS
(7)

W. Schänzer
H. Geyer
A. Gotzmann
U. Mareck-Engelke
(Editors)

Sport und Buch Strauß, Köln, 1999

A. POKRYWKA, D. KWIATKOWSKA, D. STANCZYK, K. CHROSTOWSKI:
Midodrine (Gutron) Detection in Urine Samples. Should Midodrine be Added to the IOC
Doping List?

In: W. Schänzer, H. Geyer, A. Gotzmann, U. Mareck-Engelke (eds.) Recent advances in
doping analysis (7). Sport und Buch Strauß, Köln, (1999) 383-387

MIDODRINE (GUTRON) DETECTION IN URINE SAMPLES.

Should Midodrine be added to the IOC doping list?

Department of Antidoping Research, Institute of Sport, Warsaw, Poland.

Background

Midodrine-Gutron (HAFSLUND NYCOMED PHARMA AG) is the amphetamine-like drug, which exclusively stimulates peripheral alpha receptors of the sympathetic nervous system and improves tonicity of the vessels leading to rise of peripheral resistance of arterioles and prevents orthostatic hypotension disorders. Gutron rather weakly stimulates of the central nervous system because it passes through the barrier of the brain very slowly [2, 4].

Therefore indications for application of this drug are: orthostatic hypotension, blood pressure liability due to weather sensitivity, high temperature and difficulties to start in morning hours [2, 3, 4]. Same of the properties of the midodrine may be desirable for sportsmen too.

Pharmacokinetics

Midodrine by itself is biological inactive and acts only by active metabolite [2-amino-1-(2',5'-dimethoxyphenyl)-propan-1-ol] named desglymidodrine (de-glymidodrine) which is created after hydrolysis and split off the aminoacid -glycine [Fig. 1]. Midodrine and its metabolite are excreted almost entirely in the urine as the parent compound 5%, active metabolite as 40% and rest of inactive metabolites approximately of 55% [2, 3, 4].

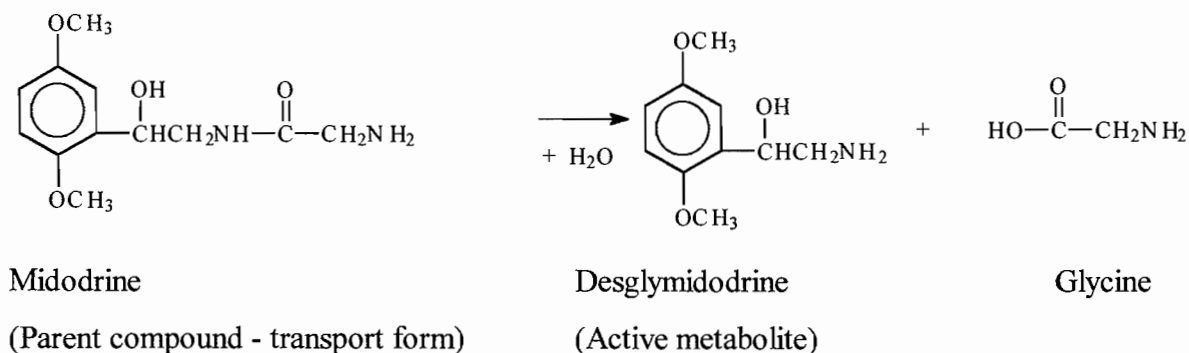


Fig. 1. Midodrine - enzymatic hydrolysis

Material and method of detection.

A single dose of Gutron (equivalent to 2.5 mg midodrine HCl) was administered orally (in tablet) to the male volunteer (28 years old). Urine samples were collected after 4, 5, 8, 11, 13, 25.5, 27, 34, 37.5, 44.5 hours. Samples were prepared accordingly to the procedure for detection of heavy volatile nitrogen containing compounds (acid hydrolysis, based extraction, derivatisation MSTFA and MBTFA) [1]. Cathine of the concentration 1 µg/ml as the internal standard was used. Samples were analysed on a Hewlett Packard GC/MS system (HP 5890 Series II Plus / HP 5972) - acquisition SCAN and SIM.

Sample preparation

1 ml 6M HCl, 100 mg cysteine and 5 µl solution of cathine (IS - final concentration 1µg/ml) were added to 5 ml of urine and the mixture was hydrolysed for 30 min at 105°C. Extraction was performed by shaking (20 min) with 5 ml diethylether. The organic layer was separated and discarded. The liquid layer was adjusted to pH 9.6 with borate buffer and K₂CO₃. After that, 5 ml of mixture diethylether/t-butanol (10/1 v/v) and 1 g Na₂SO₄ were added and extraction by shaking (20 min) was performed. The organic layer was separated, one drop TMCS was added and evaporated to dryness.

Derivatization

To dry residue was added 50 µl mixture of acetonitrile/TFA (3:2)/trace methylorange titrate. After that, MSTFA was added (methylorange from red to yellow) and kept for 10 min at 80°C. After the above mixture was cooled to room temperature, 10 µl MBTFA was added and incubated for 5 min at 80°C. 2 µl were injected to GC/MS.

GC/MS parameters

GC/MS: HP 5890 Series II Plus / 5972 MSD

Column: HP-5MS 12.5 m, 0.2 mm i.d., 0.33µm film thickness

Carrier gas: 0.9 ml/min helium, split 15:1

Temperature program: 100°C for 0.5 min, then to 220°C at 15°C/min, then to 290°C (4 min) at 20°C/min

Acquisition mode:

scan (qualitative analyse)

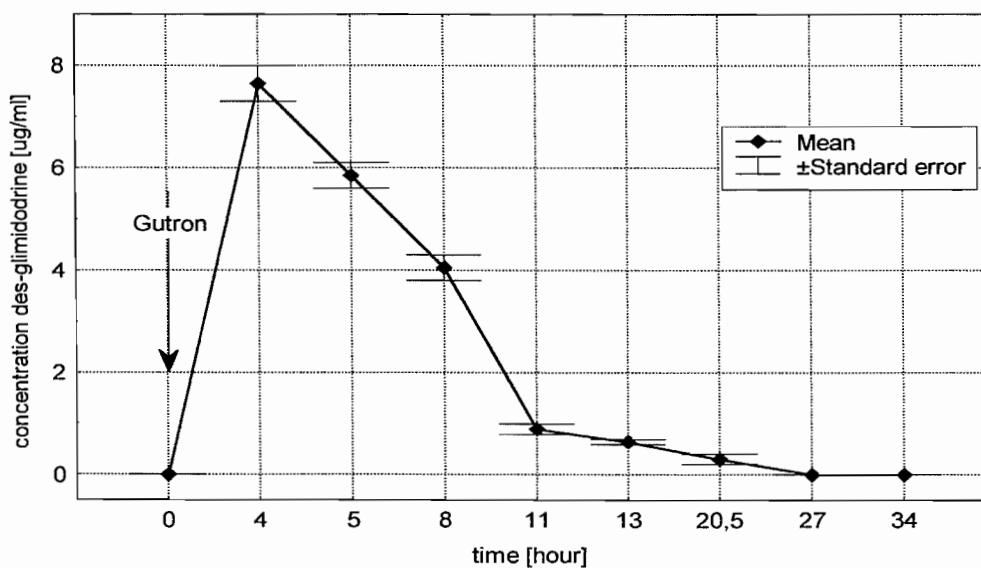


Fig. 2. Desglymidodrine excretion after Gutron (2.5 mg) application [man 28 years old]

Conclusion

Although midodrine is not formally included to the IOC list of the banned substances, detection of desglymidodrine in urine samples, in our opinion, should be reported as a doping agent.

References

1. Donike M., Geyer H., Gotzmann A., Kraft M., Mandel F., Nolteernsting E., Opfermann G., Sigmund G., Schänzer W., Zimmermann J.: Dope Analysis. In: P. Bellotti, G. Benzi, A. Ljungquist (eds): Official Proceedings: International Athletics Foundation World Symposium on Doping in Sport, Florenz, 1987. International Athletic Foundation, Florenz 1988.
2. Hafslund Nycomed Pharma AG: Gutron - monograph of product, Linz 1995.
3. Parfitt K. Ed. Midodrine Hydrochloride. In: Martindale: The Complite Drug Reference. 32 ed. Pharmaceutical Press London 1999, p: 910.
4. McTavish D., Goa K.L.: Midodrine: a review of its pharmacological properites and therapeutic use in orthostatic hypotension and secondary hypotensive disorders. *Drugs* 1989; 38: 757-77.

SIM (quantitative analyse) - monitored ions: 179, 191, 304, 239, 209, 365
(time 3.50 - 7.00 min.)

Results

In the urine samples after Gutron application we found two peaks characteristic for desglymidodrine: N-trifluoroacetyl-O-trimethylsilyl-desglymidodrine and N-trifluoroacetyl-N,O-bistrimethylsilyl-desglymidodrine [Fig. 2].

The mass spectra of the two derivatives are shown in Fig. 3 and Fig. 4. The N-trifluoroacetyl-O-trimethylsilyl derivative is main and N-trifluoroacetyl-N,O-bistrimethylsilyl is minor for desglymidodrine.

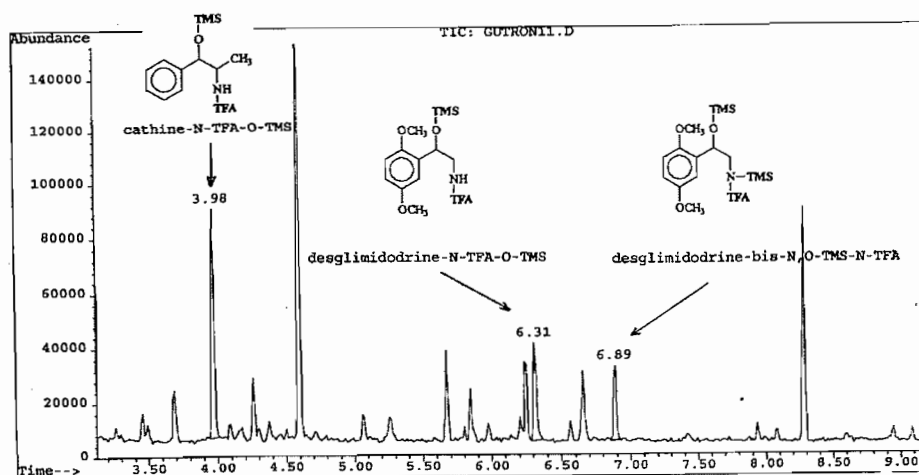


Fig. 2. GC/MS analysis of the urine sample of the man (28 years old) after application Gutron (2.5 mg)

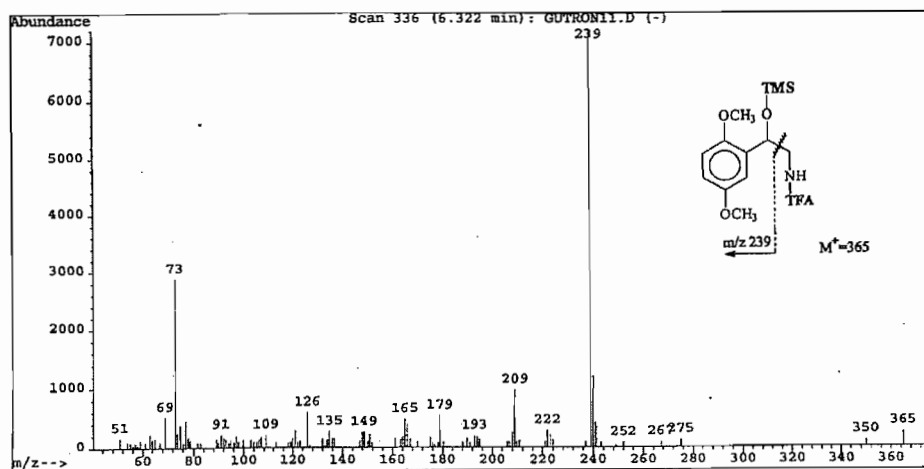


Fig. 3. Mass spectra of desglymidodrine-N-TFA-O-TMS

