



Centre
Universitaire
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Médecine
Légale



**Nouvelles approches
analytiques pour
améliorer la détection du
dopage à la testostérone**



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Laboratoire Suisse d'analyse du dopage

Why testosterone doping

Juve's Pogba provisionally suspended after testing positive for testosterone

Reuters

September 12, 2023 12:37 AM GMT+2 · Updated 14 hours ago

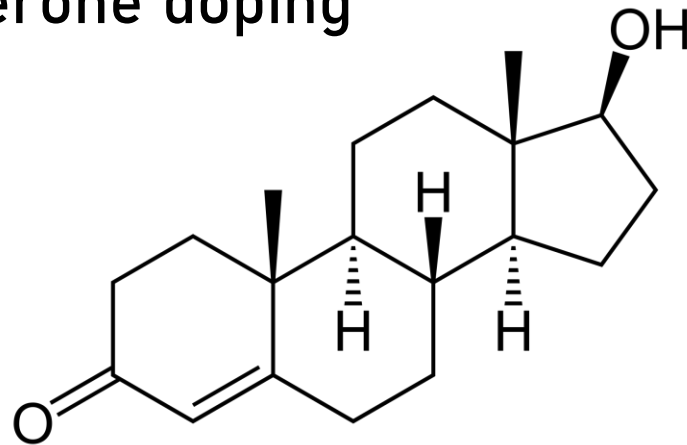


Soccer Football - Europa League - Semi Final - First Leg - Juventus v Sevilla - Allianz Stadium, Turin, Italy - May 11, 2023 Juventus' Paul Pogba reacts REUTERS/Massimo Pinca/File Photo [Aspire Licensing Rights](#)

Sept 11 (Reuters) - Juventus's France midfielder Paul Pogba has been provisionally suspended after testing positive for a banned substance, Italy's national anti-doping (NADO Italia) tribunal said on Monday.

The test, performed after Juve's 3-0 Serie A season-opening victory at Udinese on Aug. 20, detected testosterone, a hormone that increases athletes' endurance.

"In acceptance of the instance proposed by the National Anti-doping Prosecutor, it has provided for the provisional suspension of the athlete Paul Labile Pogba," NADO Italia said in a statement.



testosterone



muscle growth and strength



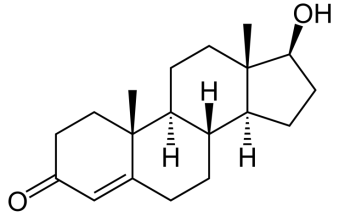
increased production of red blood cells



Detecting testosterone doping is not trivial



“My cocktail was EPO, but not a lot, transfusions and **testosterone**.”
Lance Armstrong, 2013
 (interview with Oprah Winfrey)



! ...but he was NOT tested positive for testosterone doping... !

➔ there is room for improvement (regarding anti-doping control / analysis)

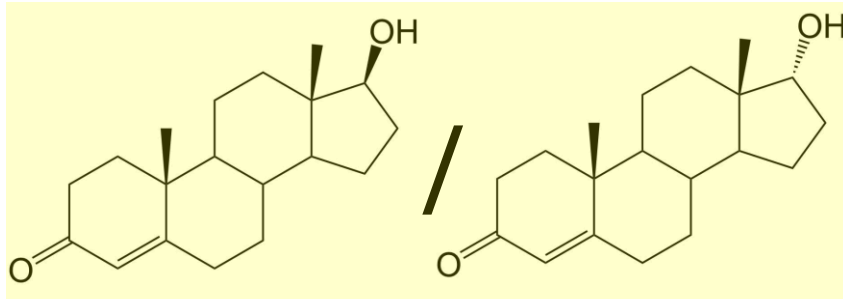


Development of the Athlete Biological Passport (ABP)

testosterone = endogenous

urinary concentrations vary throughout the day

steroid ratios in urine:

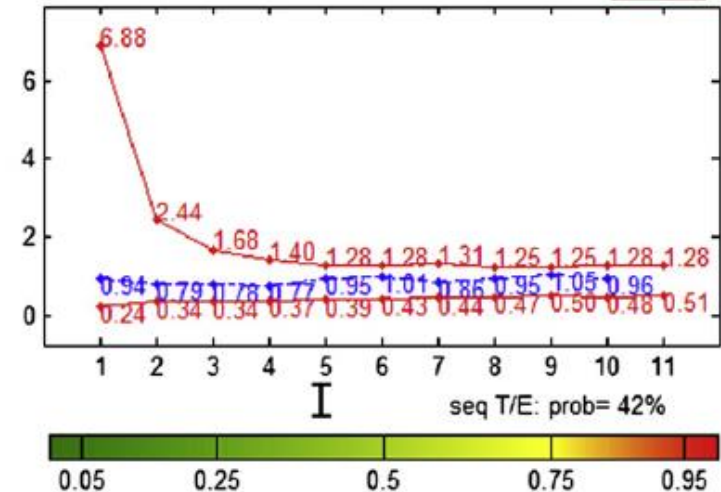


testosterone (T) / epitestosterone (E)



T/E

up

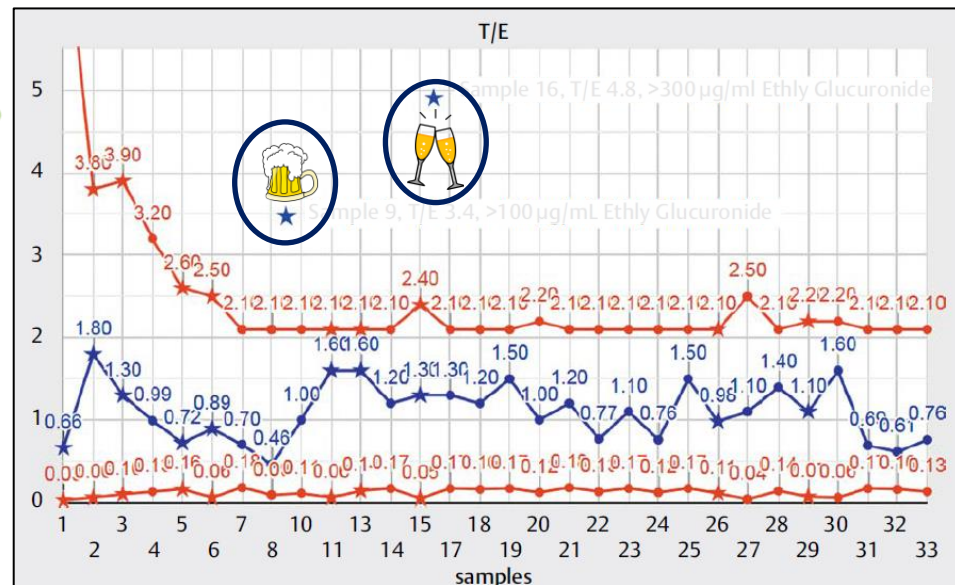
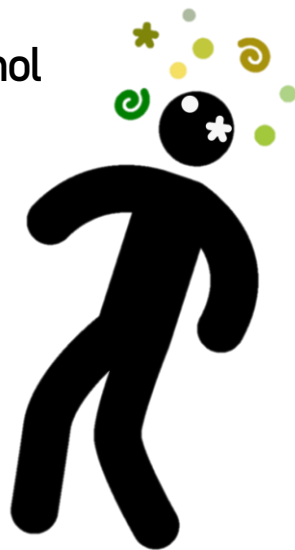


from global thresholds to individual longitudinal monitoring = urinary steroid profile



Weaknesses of the urinary steroid profile

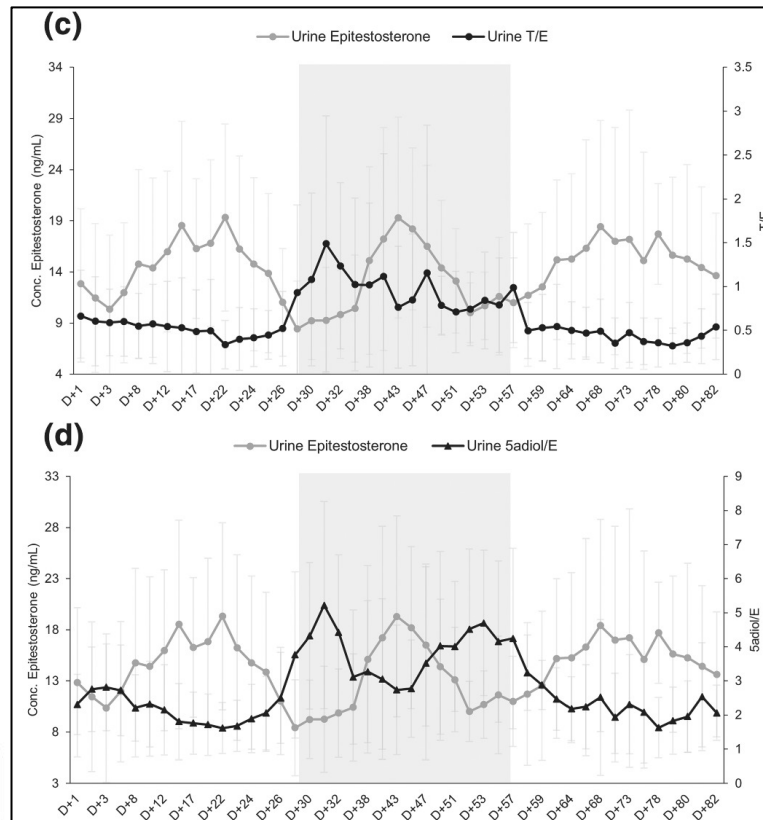
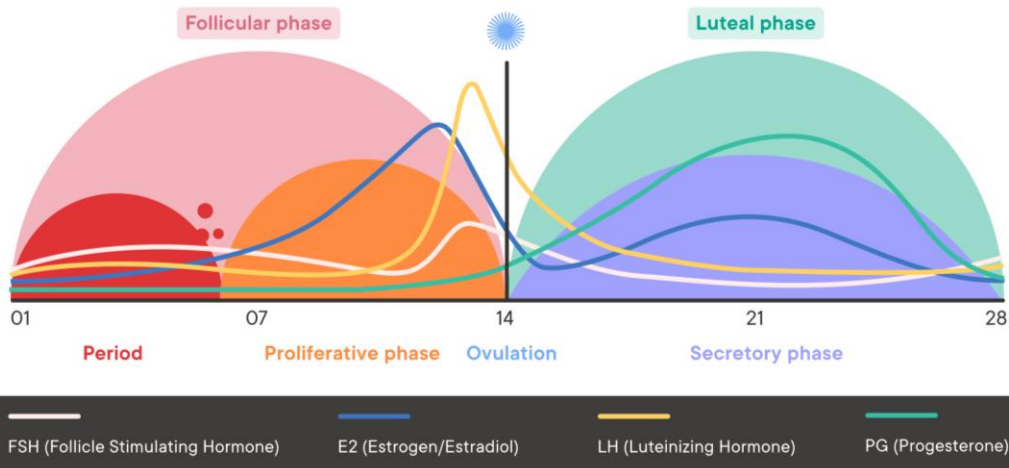
confounding factors: e.g alcohol



Weaknesses of the urinary steroid profile

confounding factors: e.g alcohol

hormonal fluctuations during the menstrual cycle

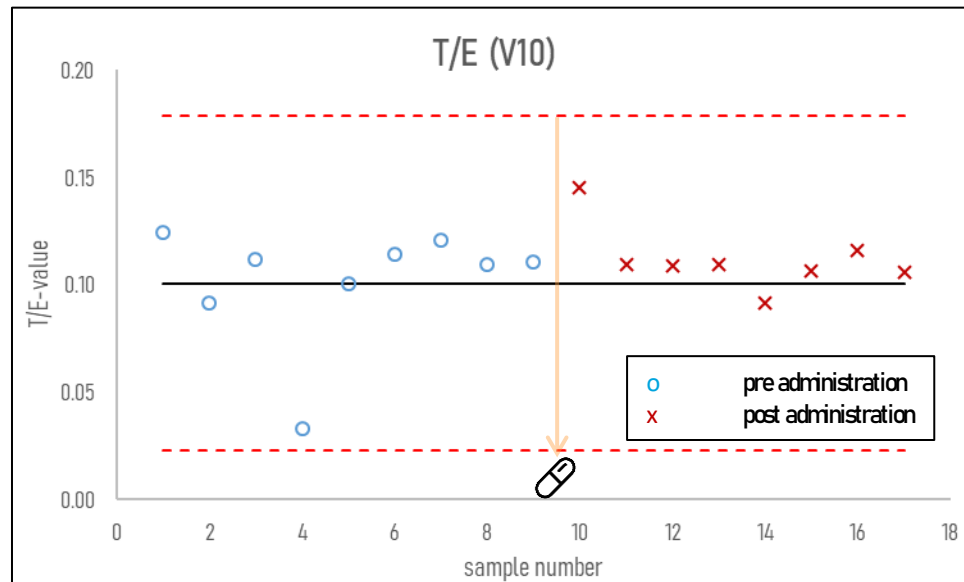


Weaknesses of the urinary steroid profile

confounding factors: e.g alcohol

hormonal fluctuations during the menstrual cycle

low excretion - low concentrations

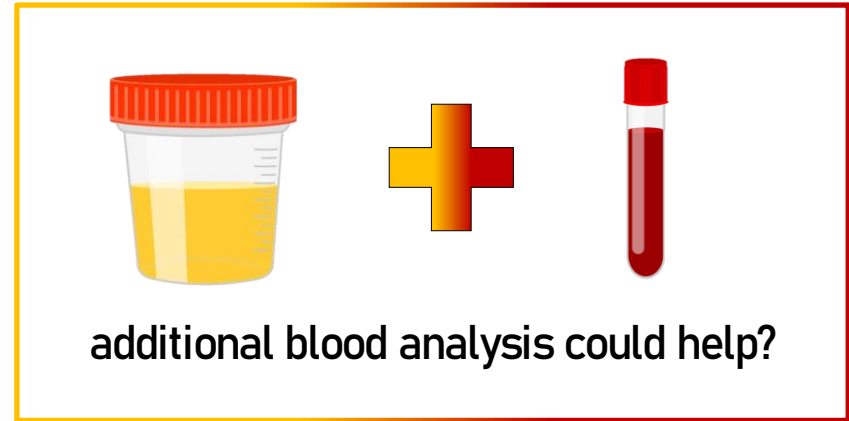


Weaknesses of the urinary steroid profile

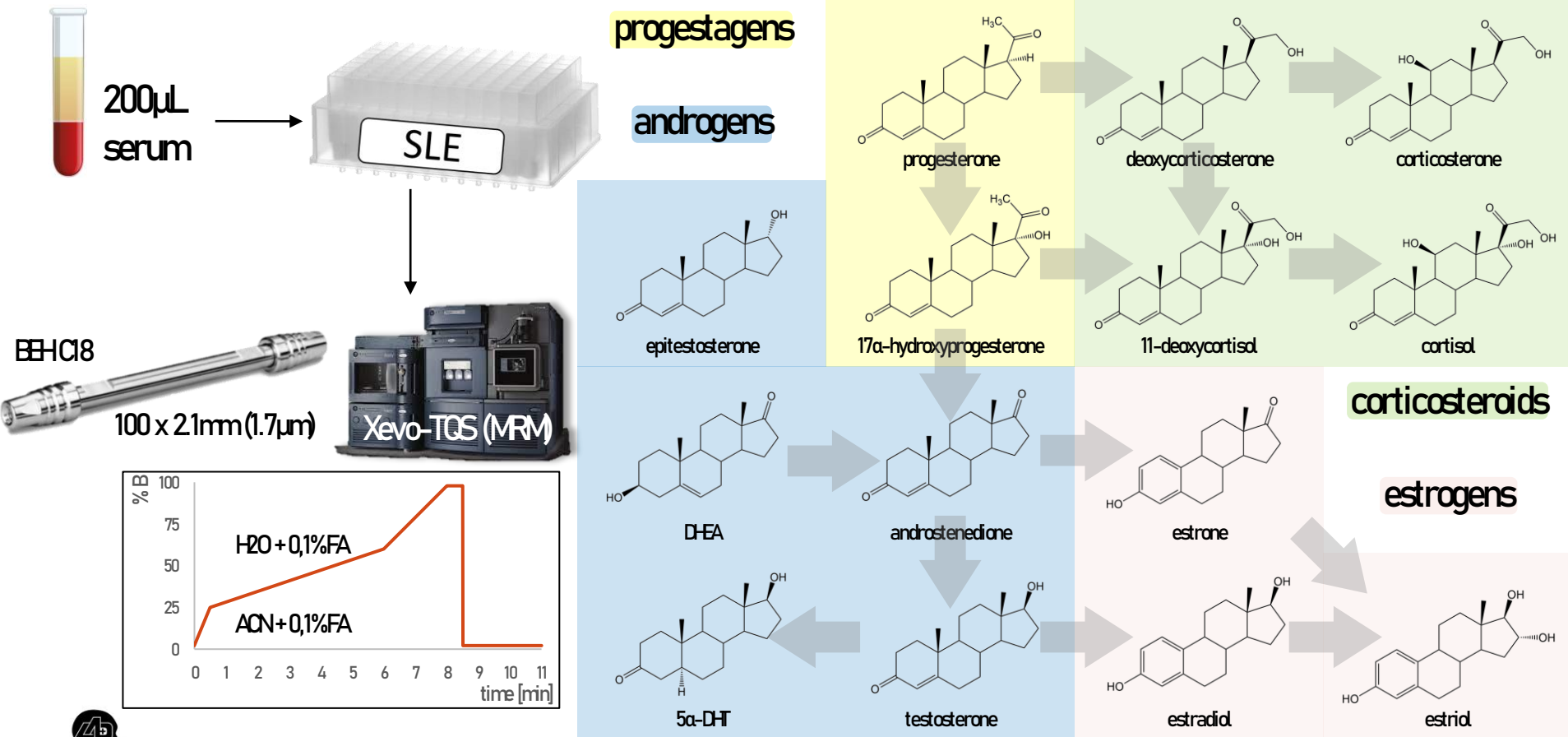
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hormonal fluctuations during the menstrual cycle

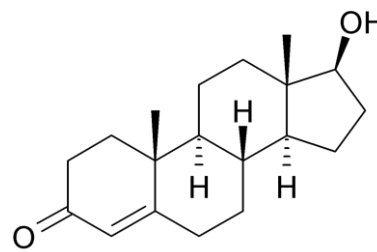
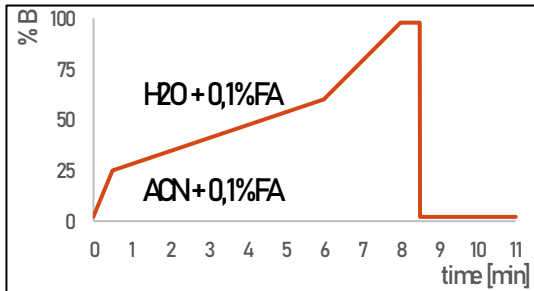
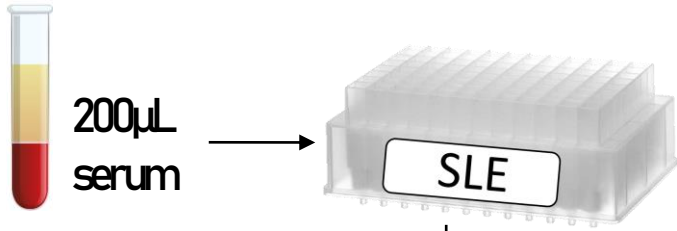
low excretion - low concentrations



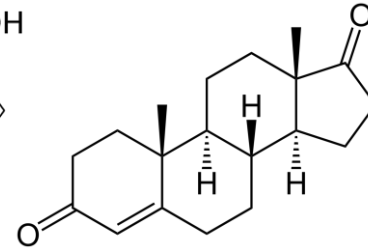
Steroid profiling in serum



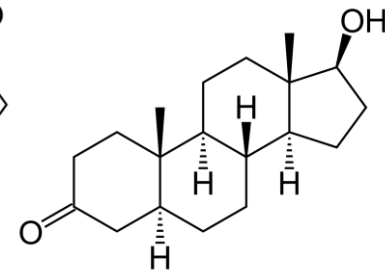
Steroid profiling in serum



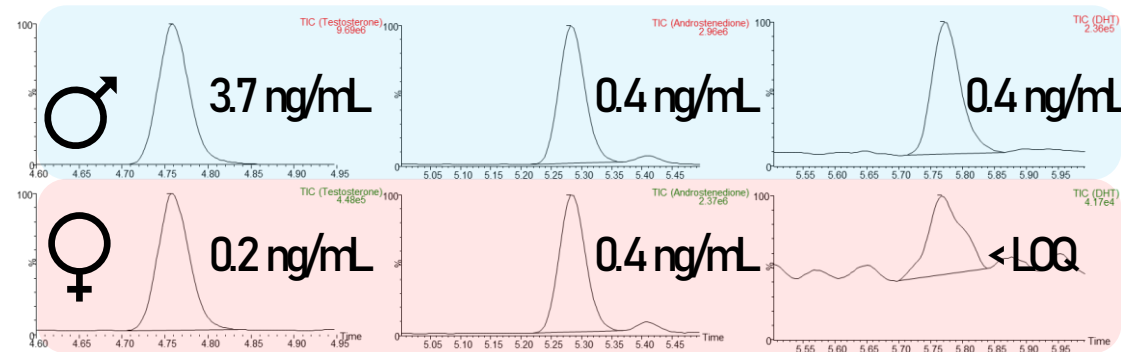
testosterone (T)



androstenedione (A4)



5α-dihydrotestosterone (DHT)



LOQs 20 pg/mL

50 pg/mL

50 pg/mL



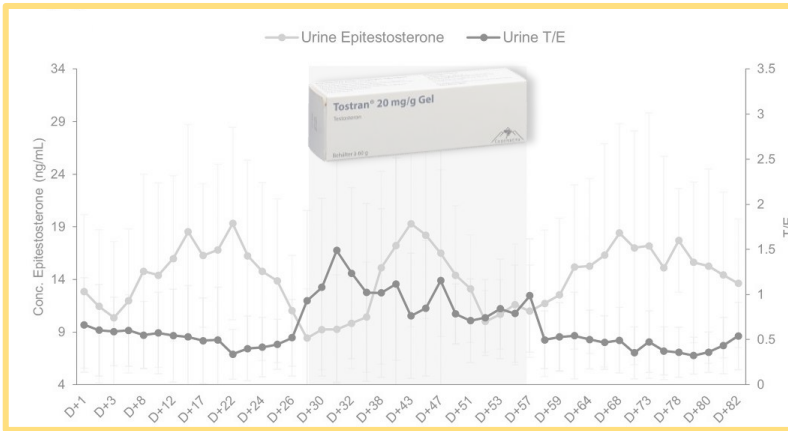
Detecting transdermal T in women

14 volunteers - urine and blood samples during 3 menstrual cycles

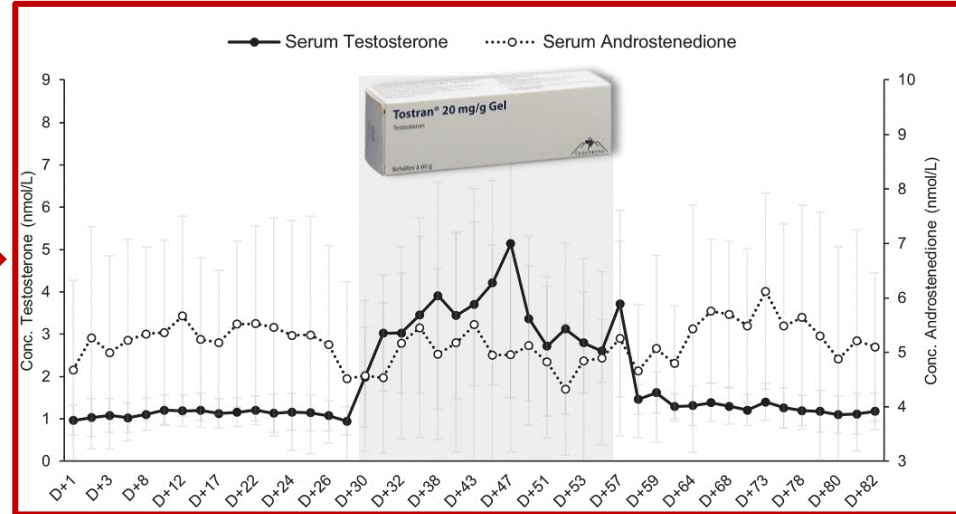
1 control-phase

2 10mg T daily (transdermal)

3 washout



urinary epitestosterone fluctuations
T/E less sensitive for women

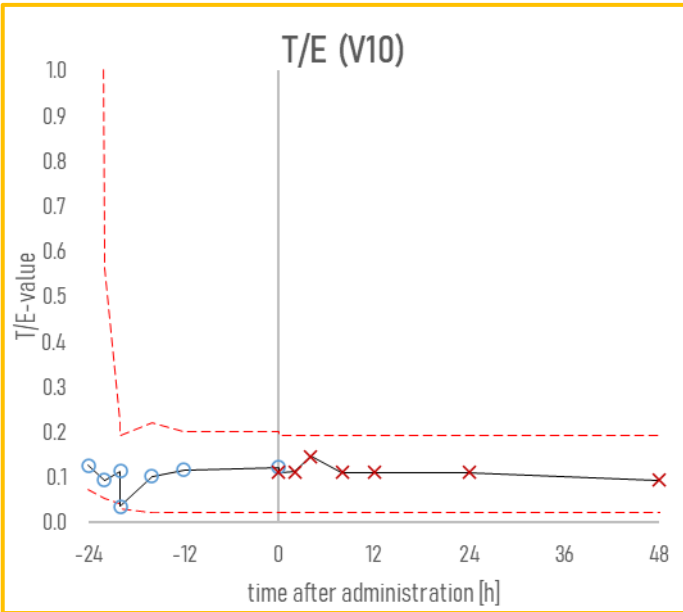


serum T more indicative
new ratio: serum T/A4



Detecting oral T in men - analysis of steroid esters

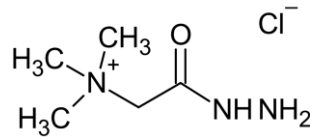
19 volunteers - 80mg **testosterone undecanoate** orally



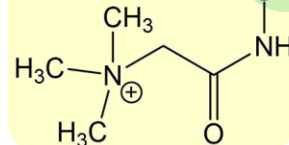
urinary steroid profile is less sensitive for low concentrations

direct detection in blood possible...

challenge: low concentrations - low signals



derivatisation with Girard's reagent T



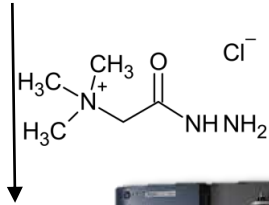
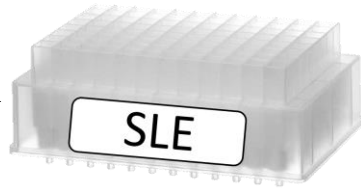
testosterone ester



Analysing steroid esters in serum



200µL
serum



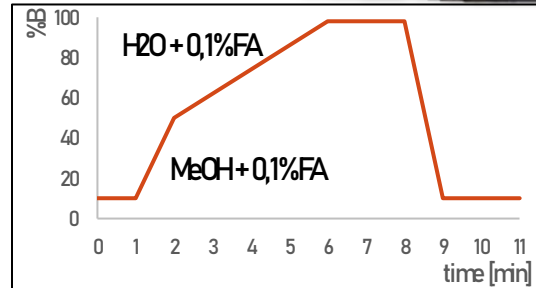
BEH C8



100 x 2.1mm (1.7µm)

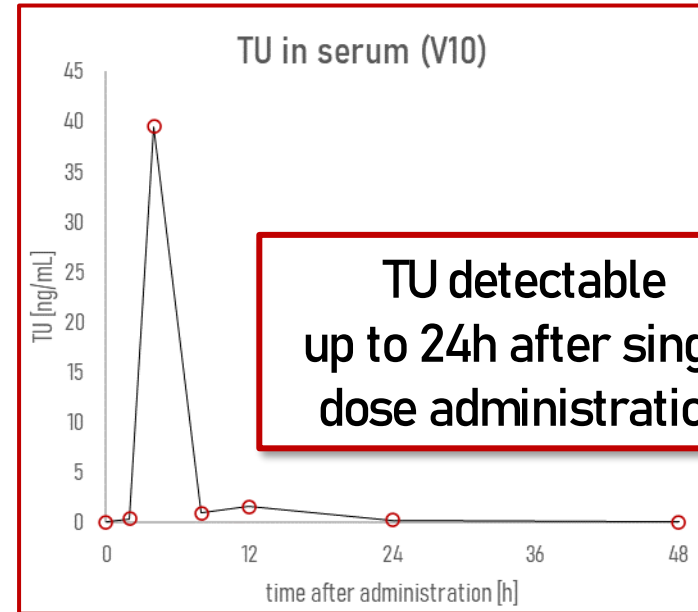


Xevo-TQS (MRM)



17 different esters
(testosterone, nandrolone, trenbolone)

LODs: 50-500 pg/mL



TU detectable
up to 24h after single
dose administration



Simplyfing blood collection with dried blood spots (DBS)



Name: Tobias Langer
Date: aujourd'hui



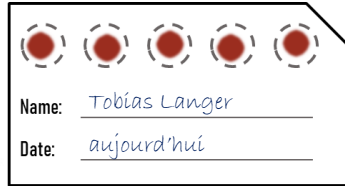
easy collection
easy storage
easy shipping
stability of substances



low volume
limited spots



Simplyfing blood collection with dried blood spots (DBS)



Name: Tobias Langer

Date: aujourd'hui

10 - 20 μ L
per spot

2x extraction with
MeOH : IPA : TBME
(25 : 25 : 50)

derivatisation
Girard's reagent T

analysis of
endogenous steroids

EC18



100 x 21mm (1.7 μ m)



analysis of
steroid esters

Cortecs C18+

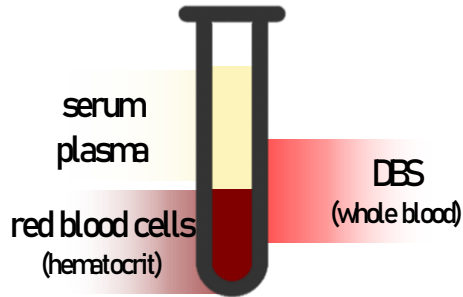


100 x 21mm (1.6 μ m)

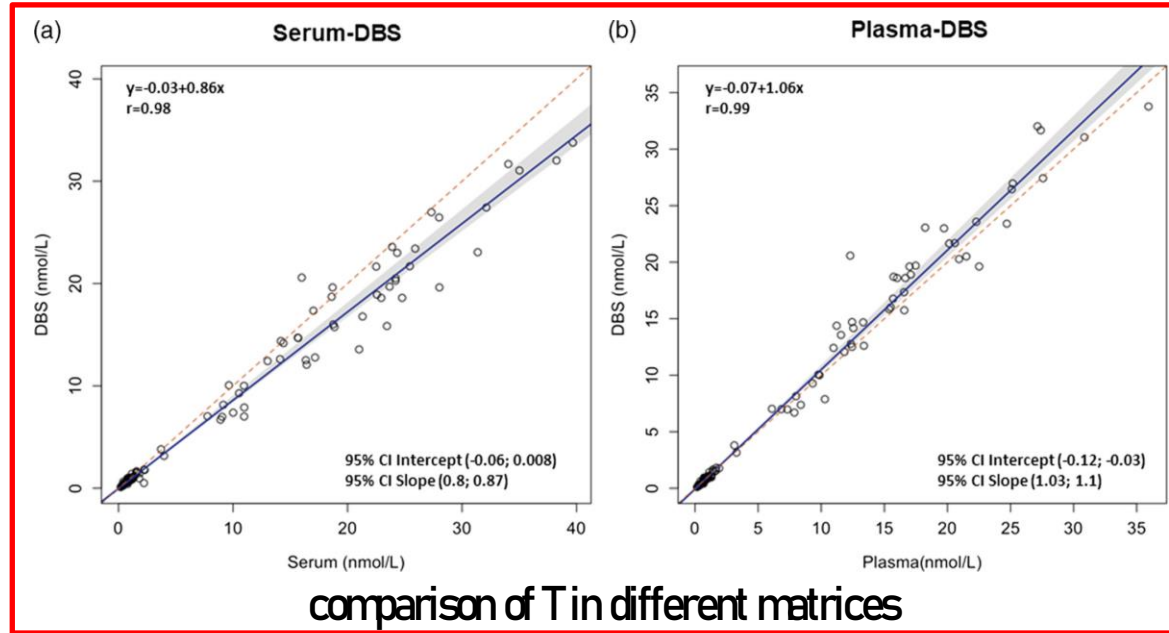


Different matrices - comparable results?

Are DBS viable for steroid profiling in comparison with serum?



analysis of:
serum, plasma, DBS
100 athletes
(51 females, 49 males)



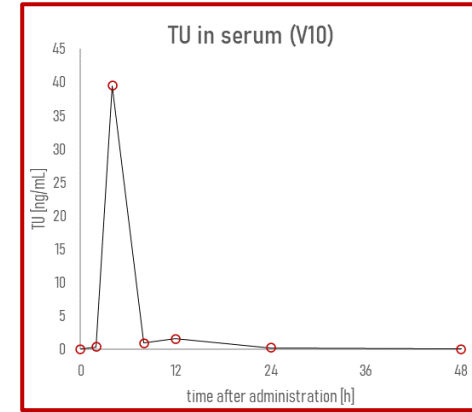
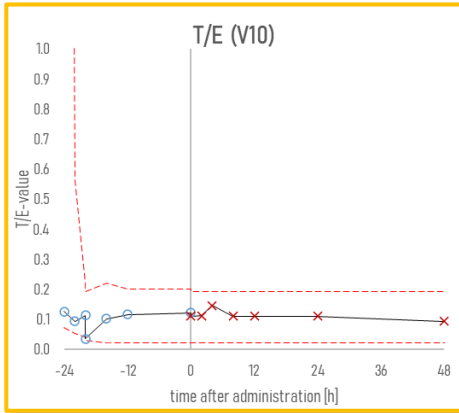
good correlation of T and A4 with generic 45% hematocrit correction
but more tests will follow...



Conclusions

Blood analysis can support urine analysis

Ester analysis as efficient alternative



Name: Tobias Langer

Date: aujourd'hui

DBS: good for research, not fully established in routine



**world
anti-doping
agency**

new module in the ABP: steroids in blood

RELEASES | 21 AUG 2023

**REMINDER: WADA to
launch new Athlete
Biological Passport
features tomorrow,
22 August 2023**



Acknowledgements

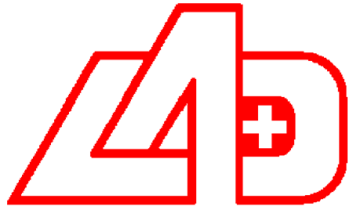
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**world
anti-doping
agency**

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Thank you
for your attention!



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