

Tox typer[®] Introduction



潘晨松 博士
高级应用工程师



毒物检测的历史状况

天才、耗时、危险



“Elementary my dear Watson”



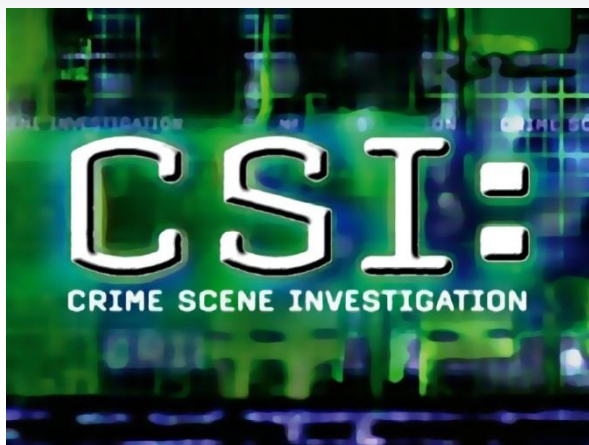
“The lab results are in....
Book him Danno,
Murder 1”



“Don't you
understand?
...these things take
time!”

毒物检测的“现状”

高科技、“无所不能”



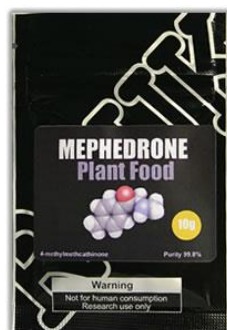
“There we go...it’s heroin, 2.1ng/ml with 32.6% purity taken at 8:52 amNow that didn’t take long did it?”

Simple as that??



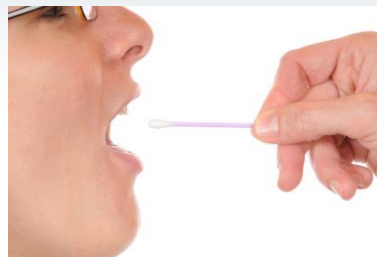
毒物检测的种类

种类众多



毒物检测的样本

复杂多样

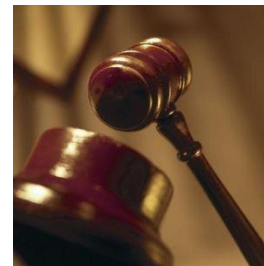


毒物检测数据需要极高的可靠性



- ***“Due to the serious legal consequences of forensic cases, particular emphasis must be placed on the quality and reliability of analytical results”.*** -TIAFT STA Committee June 13-2013
 - False Positive– Wrongful conviction / wrongful acquittal
 - False Negative – Wrongful conviction / wrongful acquittal

Data **MUST** stand up to legal scrutiny and cross examination in a court of law



毒物检测的方法

从免疫学方法走向质谱方法



- **Very easy to use**
- **High sample throughput ability**
- Lack of specificity (high false positives)
- Poor sensitivity (high cut-off limits: high false negatives)
- Unable to multiplex different drugs classes (e.g. Opioids and benzodiazepines).
- Not applicable to legal highs
- Positive results must be confirmed by GC-MS or LC/MS

- Historically, immunoassay has primarily been used as the first tier toxicological screening tool. Positive results are **confirmed by GC-MS or LCMS**.

Today, leading forensic labs are reversing this trend by using LC/MS/MS as their frontline screening tool. Some advantages of **LC/MS/MS over immunoassay include improvements in sensitivity, specificity, cost effectiveness and wider compound coverage with superior quantitation.**



Lean Toxicology: A Simplified Single Workflow!



Toxtyper[®]

MS MS² MS³



● www.bruker.com

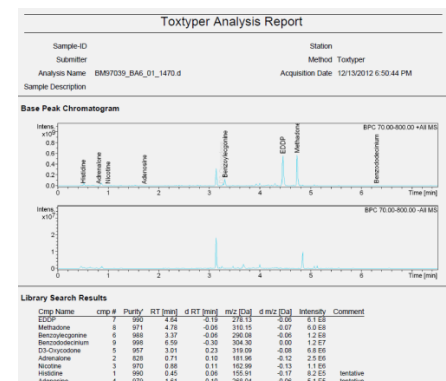
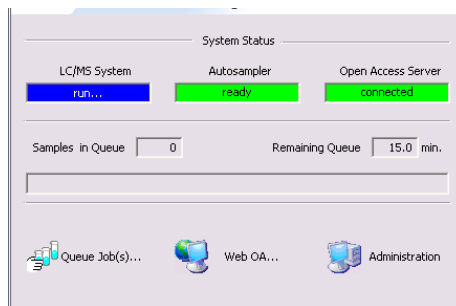
For research use only. Not for use in diagnostic procedures.

Toxtyper[®] 的工作流程

从样品到报告一键式标准控制和管理



样品制备
~ 5-10 min



鉴定报告



样品标准控制管理和检测
~ 5-10 min

From one sample to result: ~ **20 min**

?
Unknown Intox.
or drug taken

Toxtyper[®] 的样品制备 (< 10min)



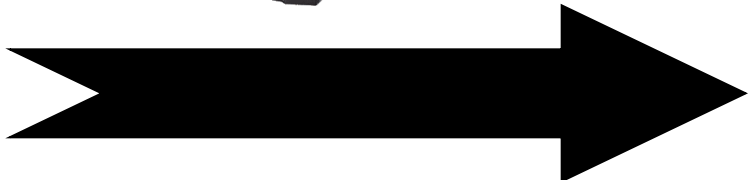
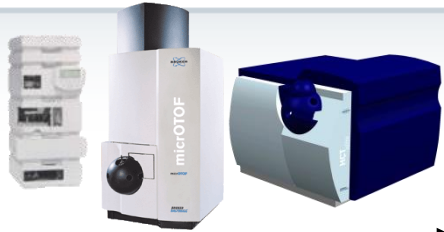
标准化可重复的高通量样品制备

- Alkaline Liquid/Liquid (LLE) extraction protocol:
 - Broad range of compounds, good removal of matrix components
 - Used for serum and urine providing high concentration effect up to 40fold
- Protein precipitation (PPT):
 - Fast and simple sample cleanup procedure, favorable if only proteins need to be removed or glucuronides should be extracted.
 - Less effective in removing some interfering compounds as compared to LLE or SPE
- Solid Phase Extraction (SPE):
 - SPE generally provides very clean extracts
 - Automation possible , slight concentration effect, up to 4fold

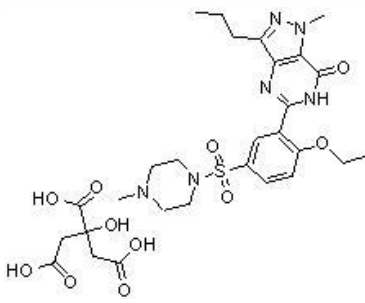
Toxtyper[®] 标准、智能的检测和报告



规范化、高通量、智能报告模式



Answer!



Toxtyper[®] 高性能的检测平台

Bruker amaZon Speed 大容量离子阱质谱



- A Bench Top LC/MS System

- Full Scan spectral information

- SmartFrag[™]



- MSⁿ at his best

- Highest Sensitivity by dual ion funnel

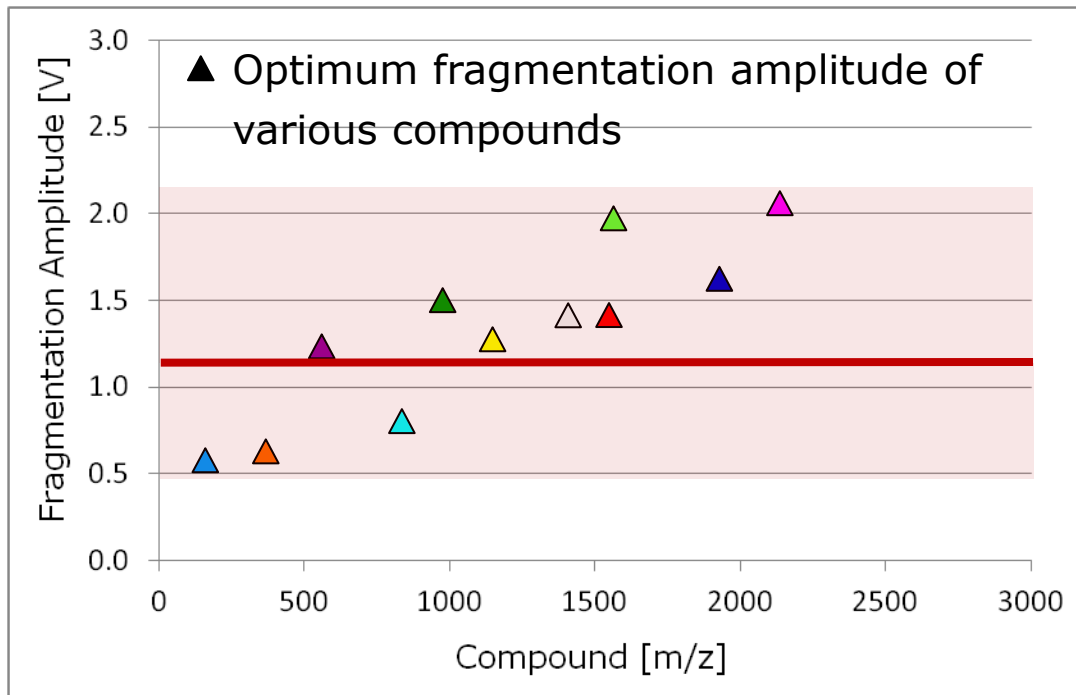
- UHPLC MS acquisition

- Zero Delay Alternating (ZDA[™]) switching

SmartFrag™: One fragmentation Fits All! Bruker patented technology



- **SmartFrag™** ramps the fragmentation amplitude for each compound:
 - successful fragmentation of **every** compound in a given sample
 - **No collision energy method development necessary**
 - Reproducible fragmentation results from **lab-to-lab** and **instrument-to-instrument**



Typical Amplitude 1.0 V

Toxtyper[®] 最全面的毒物数据库

837种欧盟规定检测毒物，并持续更新中...

Toxtyper library	Description
Content	Drugs and drugs of abuse, Neuroleptics, antidepressants, hypnotics, benzodiazepines and metabolites
Number of compounds	> 830
Number of spectra	> 2600
Characterization of spectra	ion trap, MS, MS ² and MS ³ , positive and negative ion mode
UHPLC	Retention time information included

- The comprehensive Toxtyper library covers the majority of compound classes, which are necessary for identification in clinical toxicology screening
- **Open Toxicology** library concept allows an easy input of user defined library entries by a few mouse-clicks



Toxtyper® 最全面的毒物数据库

毒物类型

抗抑郁药

Amitriptyline
Amitriptylinoxide
Clomipramine
Desipramine
Dibenzepin
Dosulepine
Doxepin
Duloxetine
Imipramine
Melitracen
Nortriptyline
Oxypertine
Protriptyline
Reboxetine

抗真菌、抗菌药

Griseofulvin
Isoconazole
Naftifine
Ornidazole
Phthalylsulfathiazole
Sulfadiazine
Sulfadoxine
Sulfaethidole
Sulfalene
Sulfamerazine
Sulfamethizole
Sulfapyridine
Terconazole
Tolnaftate

鸦片类制剂

Codeine
Heroin
Hydrocodone
Morphin-3-beta-D-glucuronide
Morphine
Morphine-6-beta-D-glucuronide

安他非命类

Amphetamine
MBDB
MCPD
MDA
MDDMA
MDEA
MDMA
Methamphetamine
PMMA
Pseudoephedrine
Sibutramin

抗组胺类药物

Bamipine
Cyclizine
Isothipendyl
Mequitazine
Methaphenilene
Oxememazine
Pyribenzamine
Pyrilamine
Tritoqualine

合成大麻类

AM-1220
AM-2233
JWH-007
JWH-015
JWH-018
JWH-019
JWH-020
JWH-072
JWH-073
JWH-081
JWH-122
JWH-122-5-fluoropentyl-derivate
JWH-200
JWH-210
JWH-250
JWH-307
JWH-387
JWH-398
JWH-412
Methanandamide
RCS-4 ortho-isomer

代谢产物

Desmethyl-chlordiazepoxide
Desmethyl-Citalopram
Desmethylclobazam
Desmethylclomipramine
Desmethylclozapine
Desmethyl-Mirtazapine
Desmethylvenlafaxine
Nordoxepine
THC-COOH
THC-OH

镇痛类

Acetylsalicylic acid
Amidopyrine
Fentanyl
Nifenazone
Salicylamide
Salicylic acid

苯并二卓氮类

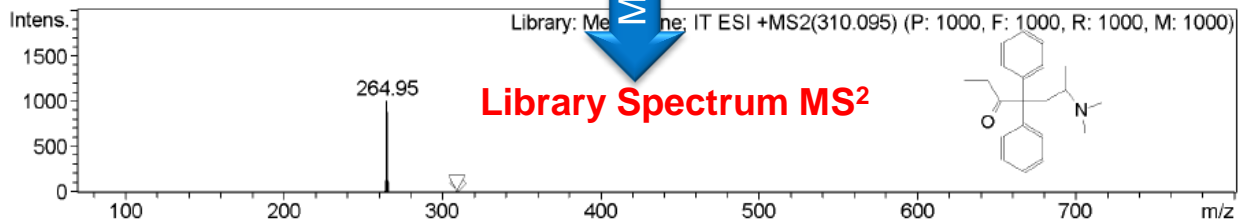
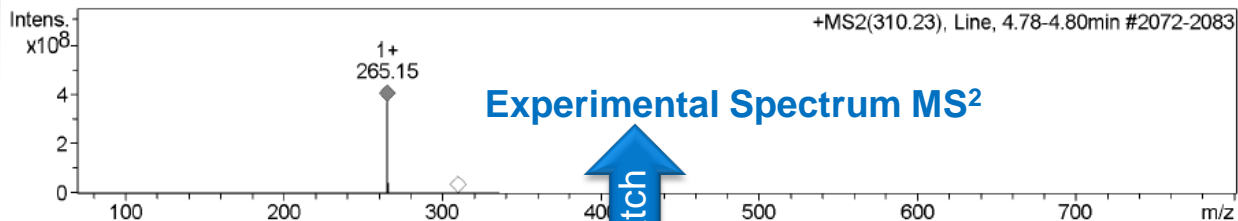
7-Aminoflunitrazepam
Alprazolam
Bromazepam
Brotizolam
Chlordiazepoxide
Clobazam
Clobenzepam
Clonazepam
Clotiazepam
Delorazepam
Desalkylflurazepam
Diazepam

农药类

Alachlor
Flurochloridone
Isoproturon
Metamitron
Methabenzthiazuron
Methoprotryne
Metsulfuron-methyl
Monolinuron
Napropamide
Sebuthylazine
Terbuthylazine
Terbutryn
Triasulfuron

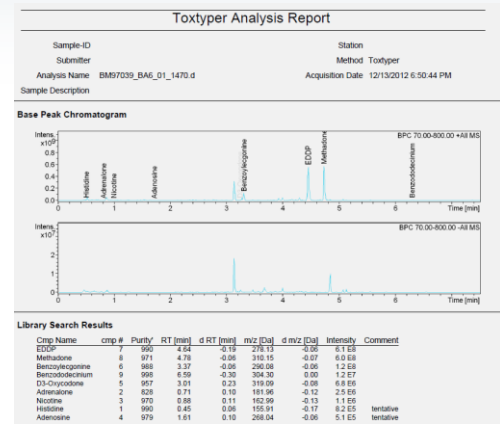
Toxtyper[®] 毒物数据库图谱匹配

自动评估并生成报告

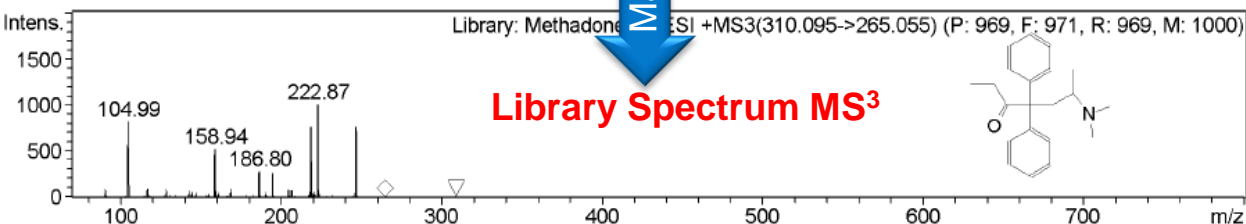
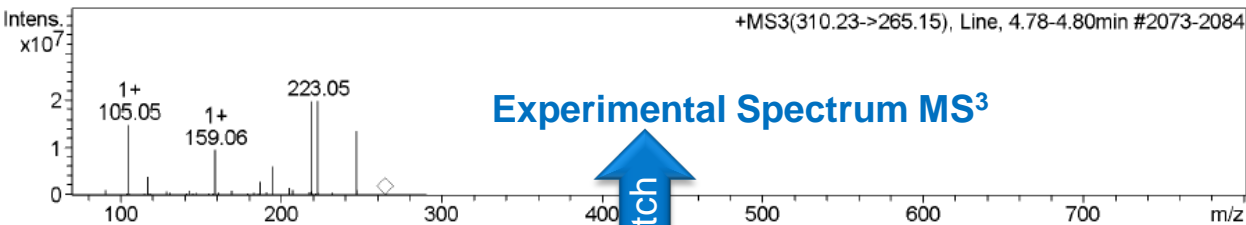


Match

Cross match of MS²/MS³ spectra



Report



Match

- Inter-laboratory test with **7 Toxtyper** systems organized by the Institute of Forensic Medicine , Freiburg

Study design:

17 compounds spiked in 3 human serum samples for an Inter-laboratory test. Spiked concentrations in ng/mL:

no therapeutic level known, sub-therapeutic, therapeutic, toxic

Sample 1

Methadone (**250**)
EDDP (**50**)
Diazepam (**100**)
Nordazepam (**500**)
Oxazepam (**200**)
Temazepam (**100**)

Sample 2

Trimipramine (**100**)
Amitryptiline(**100**)
Zolpidem (**500**)
Midazolam (**150**)
 α -OH-midazolam (**50**)
Fentanyl (**3**)
Lidocaine (**200**)

Sample 3

Duloxetine (**600**)
Nordoxepine(**300**)
Mirtazapine (**50**)
Metoprolol (**200**)

Toxtyper[®] 检测结果

不同实验室的重复性



Application Note # LCMS-73

Interlaboratory Tests Demonstrate the Robustness and Transferability of the Toxtyper[™] Workflow

Abstract

There is high demand in clinical and forensic toxicology for comprehensive, specific and transferable techniques that overcome the well-known limitations of current GC-MS, LC-UV/DAD and immunoassay solutions. Liquid chromatography-tandem mass spectrometry (LC-MS) combined with library searching is an emerging screening solution for toxicology.

Here we describe a robust and automated solution for the detection and identification of drugs and drugs of abuse in biological specimens. The workflow was tested with regard to method- and result-transferability from lab to lab.

Authors

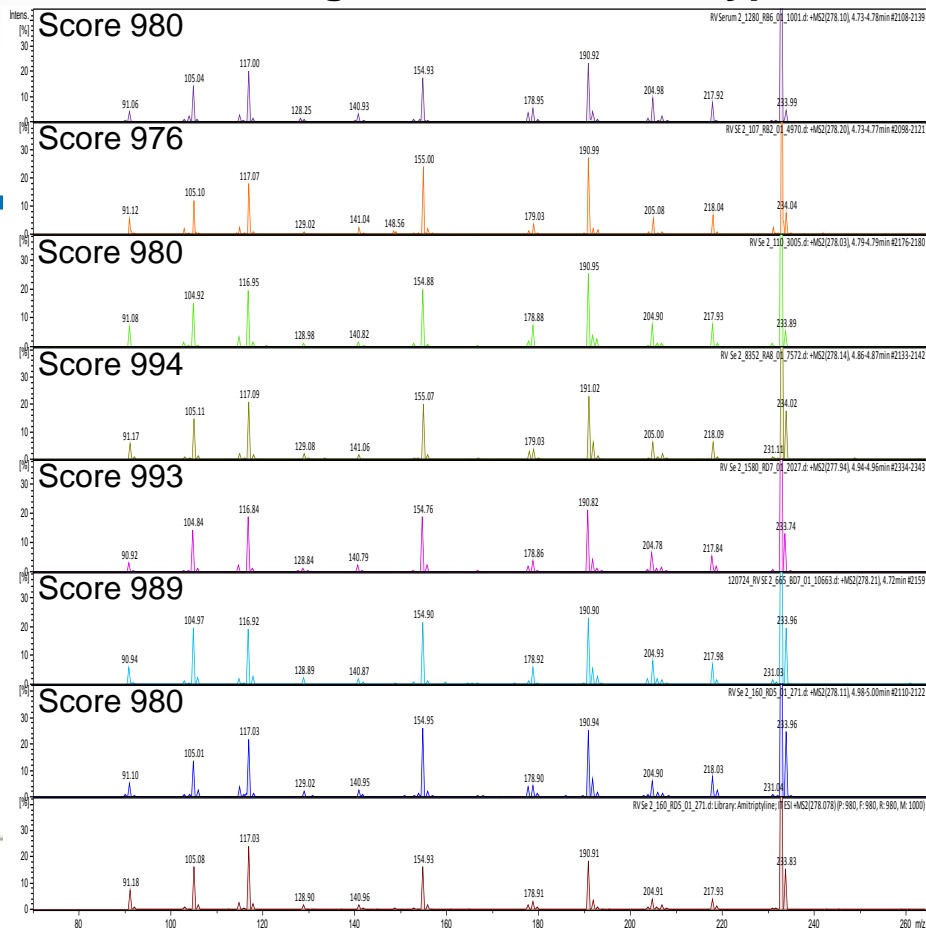
Jürgen Kempf, Laura Huppertz, Susanne Vogt
Institut für Rechtsmedizin, Universitätsklinikum Freiburg, Germany

Markus Meyer, Sebastian Götz, Birgit Schneider
Bruker Daltonik GmbH, Bremen, Germany

Keywords

Keywords	Instrumentation and Software
Library Search	amaZon speed
Toxicology Screening	Compass OpenAccess 1.4
Drug Screening	DA 4.1
Drugs of Abuse	Ion Trap

MS² fragmentation of Amitriptylin



Instruments

Bdal 1

Bdal 2

IRM

HUG 1

HUG 2

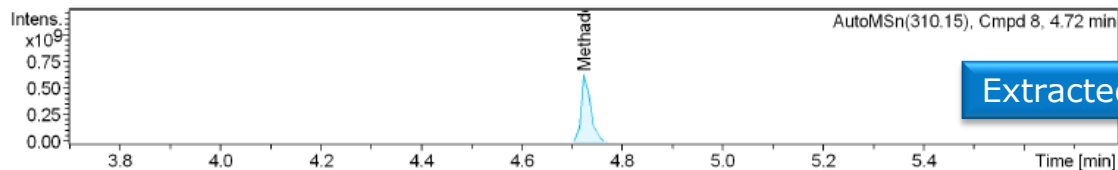
IKC

UK

Library spectrum

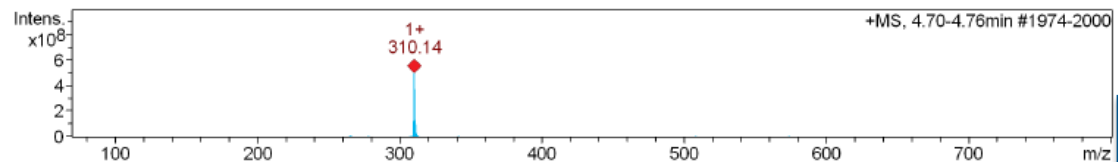


Extracted Ion Chromatogram

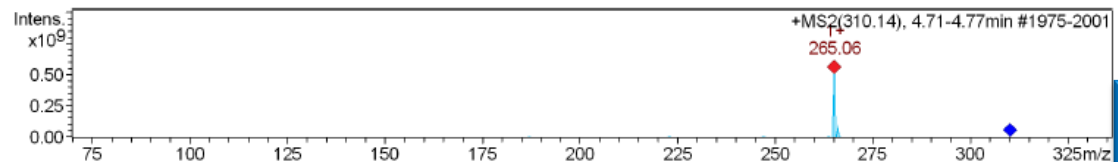
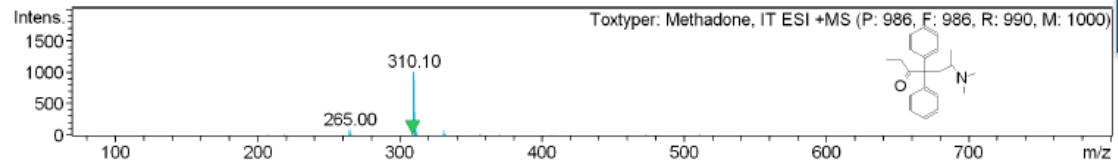


Extracted ion chromatogram

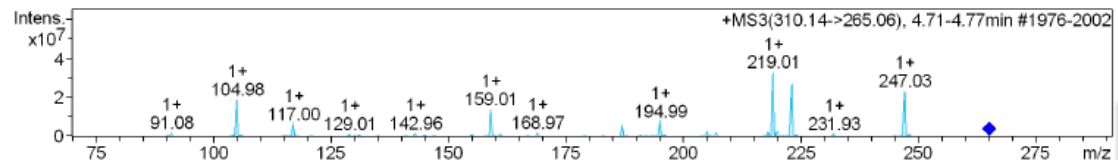
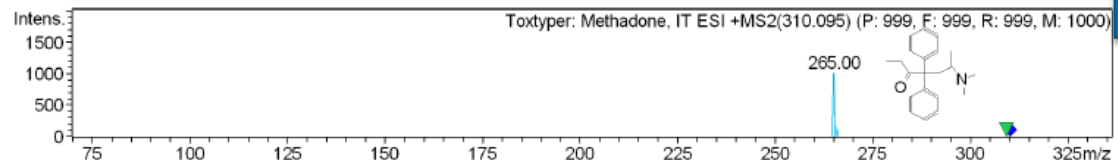
Compound Spectra



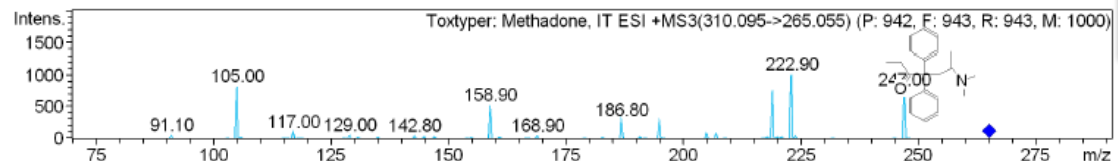
MS – experimental vs. library



MS² – experimental vs. library



MS³ – experimental vs. library





Tox typer[®] 检测结果

不同实验室返回结果

Spiked Compounds	Participants						
	IKC	IRM	HUG 1	HUG 2	UK	BDal 1	BDal 2
Sample 1							
Methadone	✓	✓	✓	✓	✓	✓	✓
EDDP	✓	✓	✓	✓	✓	✓	✓
Diazepam	✓	✓	✓	✓	✓	✓	✓
Nordazepam	✓	✓	✓	✓	✓	✓	✓
Oxazepam	✓	✓	✓	✓	✓	✓	✓
Temazepam	✓	✓	✓	✓	✓	✓	✓
Sample 2							
Amitriptyline	✓	✓	✓	✓	✓	✓	✓
α-OH-midazolam	✓	✓	✓	✓	✓	✓	✓
Fentanyl	✓	✓	✓	✓	✓	✓	✓
Lidocaine	✓	✓	✓	✓	✓	✓	✓
Midazolam	✓	✓	✓	✓	✓	✓	✓
Trimipramine	✓	✓	✓	✓	✓	✓	✓
Zolpidem	✓	✓	✓	✓	✓	✓	✓
D5-diazepam (IS)	✓	✓	✓	✓	✓	✓	✓
Ingredient of Serum							
Caffeine	✓	✓	✓	✓	✓	✓	✓
Theobromine	✓	✓	✓	✓	✓	✓	✓
Sample 3							
Duloxetine	✓	✓	✓	✓	✓	✓	✓
Metoprolol	✓	✓	✓	✓	✓	✓	✓
Mirtazepine	✓	✓	✓	✓	✓	✓	✓
Nordoxepine	✓	✓	✓	✓	✓	✓	✓
D5-diazepam (IS)	✓	✓	✓	✓	✓	✓	✓
Ingredient of Serum							
Caffeine	✓	✓	✓	✓	✓	✓	✓
Theobromine	✓	✓	✓	✓	✓	✓	✓

Toxtyper® 欧洲用户



- Prof. Denis Hochstrasser, Dr. Marc Fathi, HUG - Hôpitaux Universitaires de Genève :
 - “We are using **two Toxtyper systems** for *the routine toxicological screening of patients admitted to the emergency room and profit from the ease-of-use and fast result generation of this solution.*”
- Jürgen Kempf, Institute of Forensic Medicine in Freiburg:
 - *The combination of a simple push-button solution with the performance of the trap technology makes the **Toxtyper solution** a valuable tool for routine and research forensics labs.*
- PD Dr. Peter Findeisen, Institute for Clinical Chemistry, Mannheim (UMM):
 - *We are using the system for routine screening of patients that are admitted to the emergency room of the UMM with suspected intoxications. The specificity of the method and shortened turnaround times are rather impressive, and perfectly fit the demands for general unknown screening.”*
- **The German BKA (Federal Criminal Agency)** got their **first Toxtyper** system beginning of 2013

Toxtyper[®] 总结



- 使用方便，适合日常检测，无需专业质谱知识；
- 快速报告，响应迅速；
- 一键式分析、报告，规范化操作流程；
- 全面、信息丰富的毒物图谱数据库；

质谱在毒物检测的应用



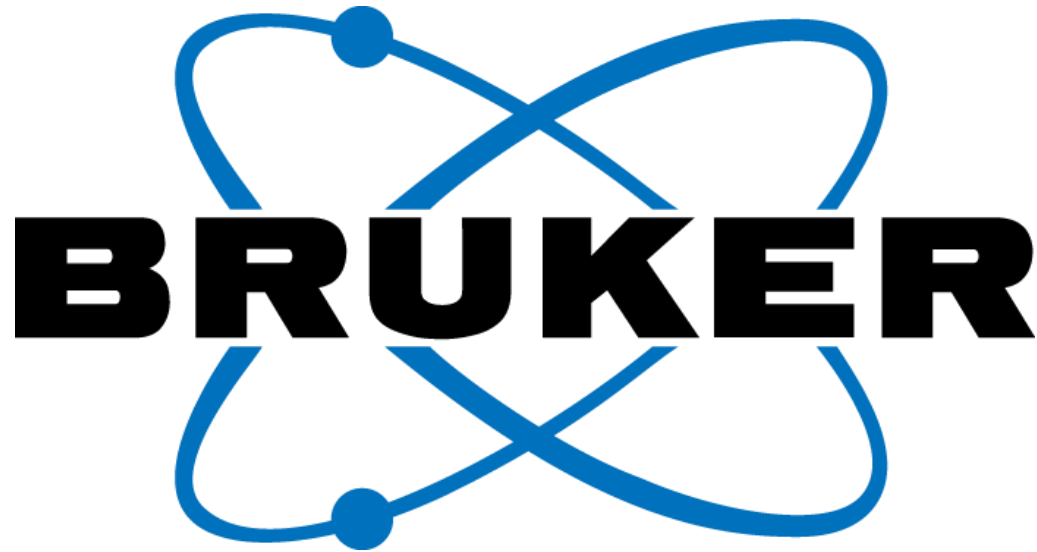
	MTS (n > 300)	Quant	LOD / Sensitivity	GUS	Price	Compound Coverage	Sample prep	Ease of use
Q-TOF	++++	++	+++	++++	+	+++	++++	+++
ToxTyper	++++	++	++	++	+++	+++	++++	++++
Triple Quad	+++	++++	++++	NO	++	+++	++++	++
GC-MS(MS)	++	++++	+++	++	++++	+	++	+++



Bruker 北京实验室欢迎你

液质联用 (q-TOF和离子阱)





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