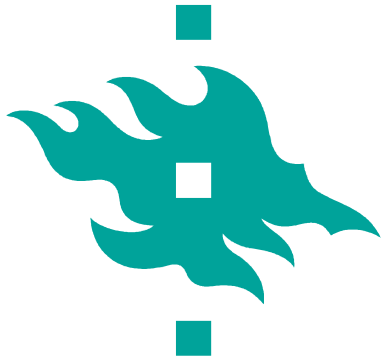




Substituting immunoassay urine drug screening by UHPLC-HR-TOFMS

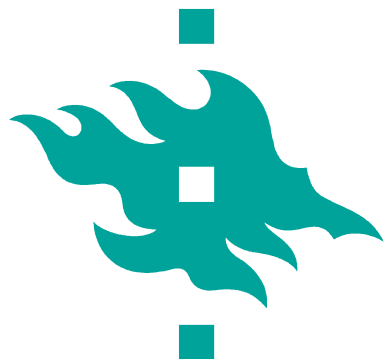
Mira Sundström, Anna Pelander, Ilkka Ojanperä

Hjelt Institute, Department of Forensic Medicine,
University of Helsinki, Finland



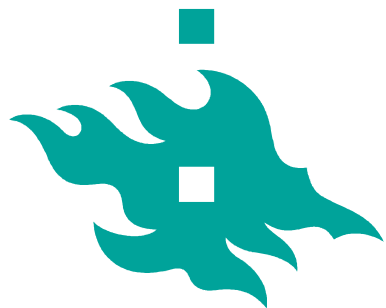
Areas of drug screening

- Post-mortem toxicology within cause of death investigation
- Clinical forensic medicine
 - Driving under the influence of drugs (DUID)
 - Offenders and victims
 - Drug-facilitated crime
- Treatment of drug addicts
- Child welfare
- Clinical and emergency toxicology
- Drug testing (workplace, prisons, armed forces etc.)
- Doping



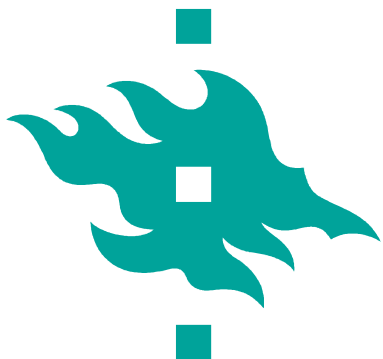
Designer drug findings in post-mortem cases

	2010	2011	2012	2013	2014
Fluorinated amphetamines	3	1	4		
Other designer amphetamines		5	8		2
Synthetic cannabinoids			1		
mephedrone (4-MMC)	2				
methylone (MDMC)	1				
3,4-dimethylmethcathinone (3,4-DMMC)		2			
pyrovalerone				1	
MDPV	14	10	8	2	
alpha-PVP			12	16	3
2-DPMP	1	3	1	1	1
bromo-dragonfly		2		2	
methiopropamine			1		
MePPP			1	1	
methoxetamine			1		
AMT				1	
TOTAL	21	23	36	24	6



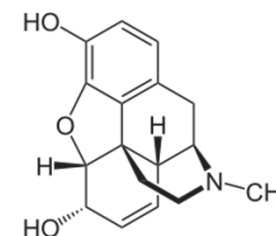
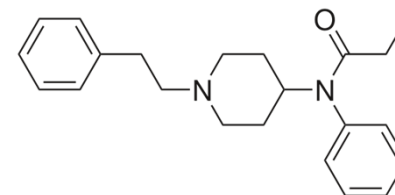
Designer drug findings in clinical samples

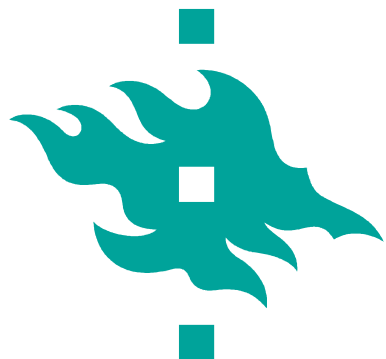
	2010	2011	2012	2013	2014
Fluorinated amphetamines		2	3	1	1
methylone (MDMC)	1		2		
MDPV	40	10	11	8	4
alpha-PVP			4	36	56
2-DPMP	1	2	16		
bromo-dragonfly				1	1
PMA		1	1		
MePPP			1		
Methiopropamine			2	7	7
25I-NBoMe			1		
2-CE			1		
methoxetamine				1	
alpha-PBP				1	
6-APB					1
3,4-CTMP					1
TOTAL	42	15	42	55	71



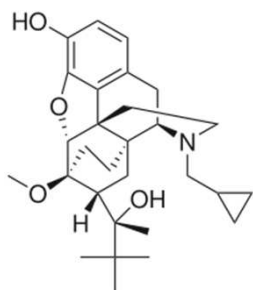
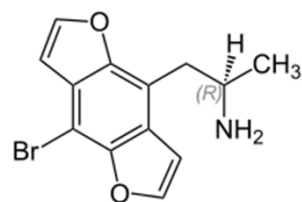
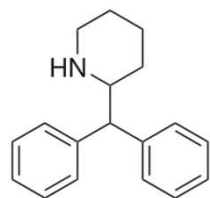
Replacing immunological drug screening by HRMS?

- The conventional way to perform drugs of abuse screening is by various immunoassay
- Typical coverage and cut-offs by immunoassay
 - Amphetamines 300 ng/mL
 - Opiates 300 ng/mL
 - Cocaine 300 ng/mL
 - Benzodiazepines 200 ng/mL
 - Cannabinoids 20 ng/mL
 - Buprenorphine 1 ng/mL

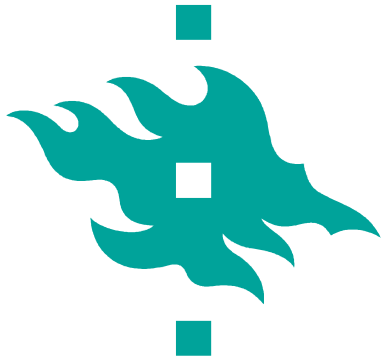




Coverage of the mass spectrometry based method: 526 compounds

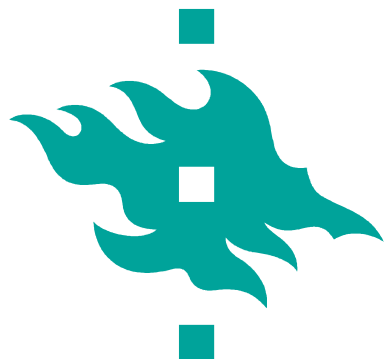


- New psychoactive substances (NPS)
 - Synthetic cannabinoids and cathinones
 - Piperidine derivatives (2-DPMP)
 - Tryptamine derivatives (5-MeO-DIPT)
 - Other phenethylamine derivatives (DOB, bromodragonfly)
- Conventional drugs of abuse (cannabis, amphetamine)
- Prescription medicines, such as morphine, buprenorphine, benzodiazepines, Z-drugs



Methods used in this study





Main results for 279 urine samples

Immunoassay

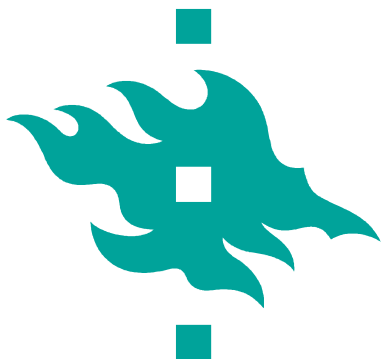
	TP	FP	FN
amphetamine	7	5	4
buprenorphine	24	10	3
THCA	21	0	6
benzodiazepines	62	0	18
opiates	22	7	9
TOTAL	136	22	40

false negatives due to high cut-off concentrations and false positives due to interfering matrix

UHPLC-HR-TOFMS

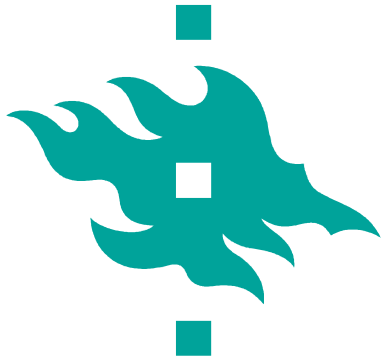
	TP	FP	FN
amphetamine	11	0	0
buprenorphine	27	0	0
THCA	27	0	0
benzodiazepines	80	0	1
opiates	31	5	0
TOTAL	176	5	1

Excellent correlation with confirmation analyses (GC-MS or LC-MS/MS)

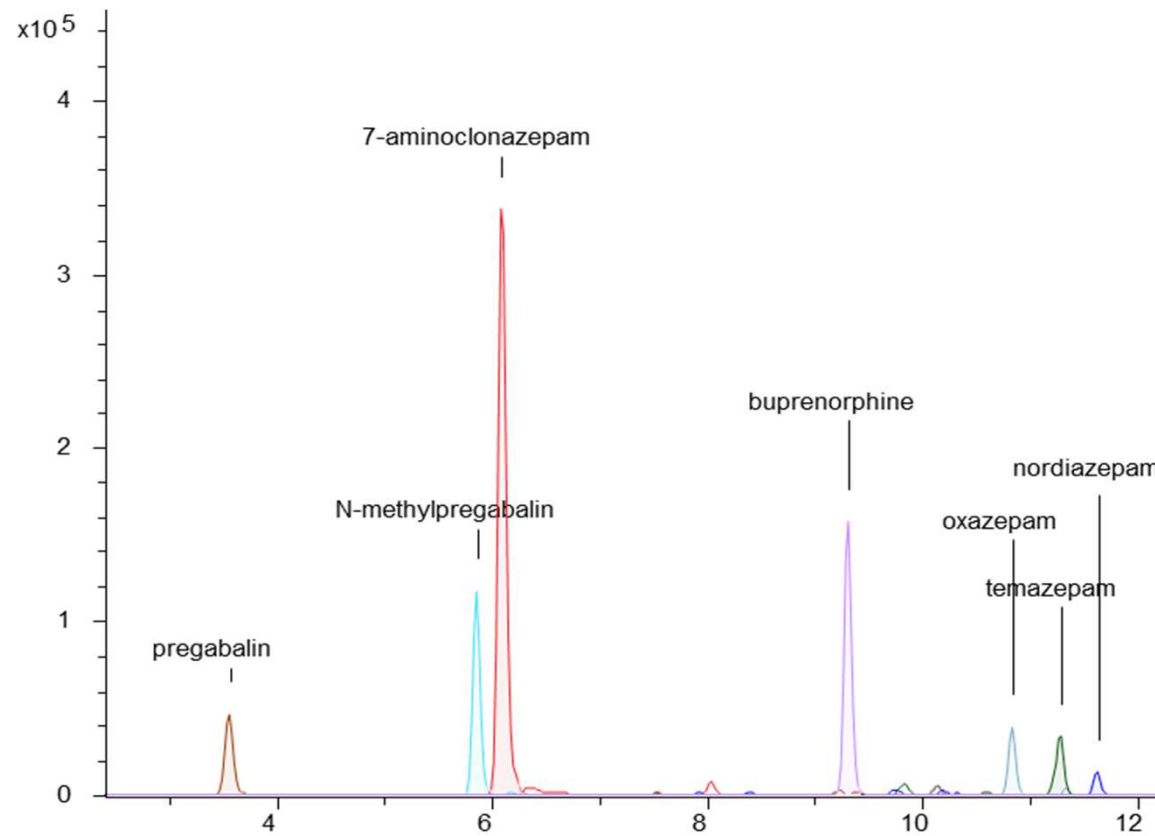


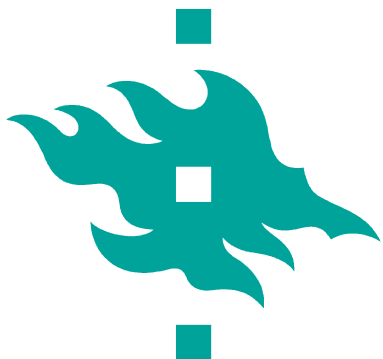
Other findings by UHPLC-HR-TOFMS

	n
Oxycodone	16
Fentanyl	3
Pregabalin	20
Zopiclone	25
Zolpidem	5
Methadone	4
Ketamine	3
ephedrine/pseudoephedrine	4
Tramadol	19
alfa-PVP	3
AMT	1



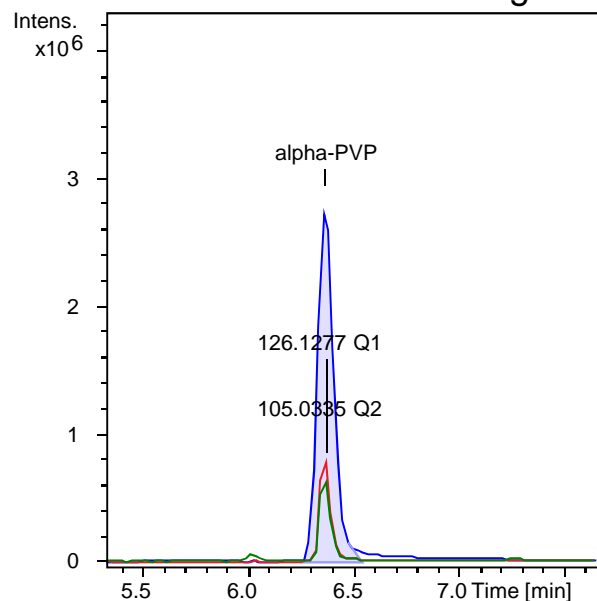
Case example



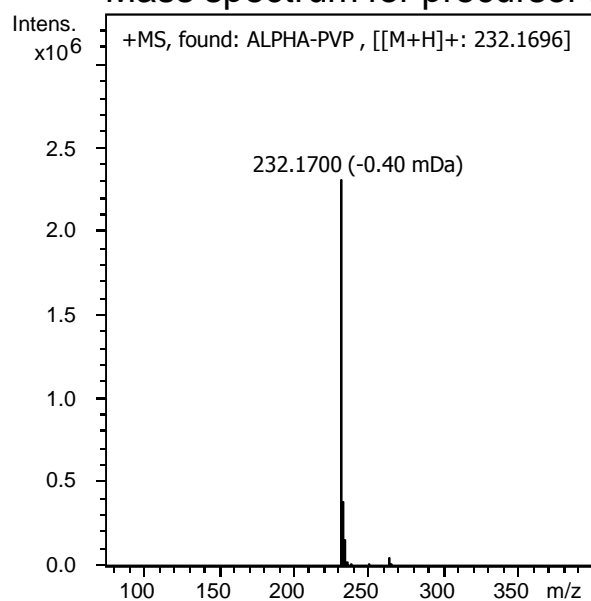


Example of a confirmation level identification of alpha-PVP

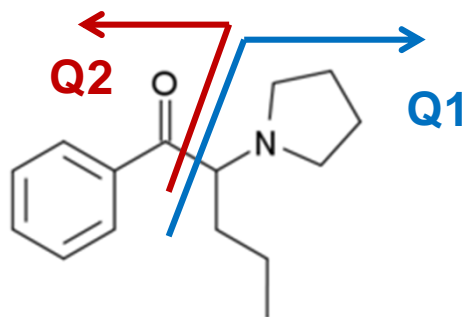
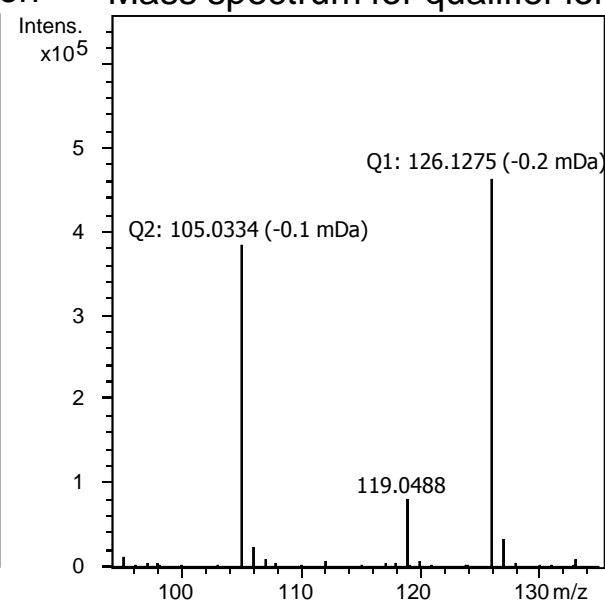
Extracted ion chromatograms

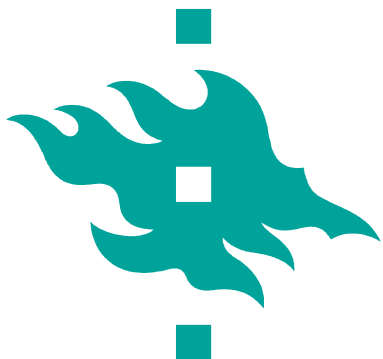


Mass spectrum for precursor ion



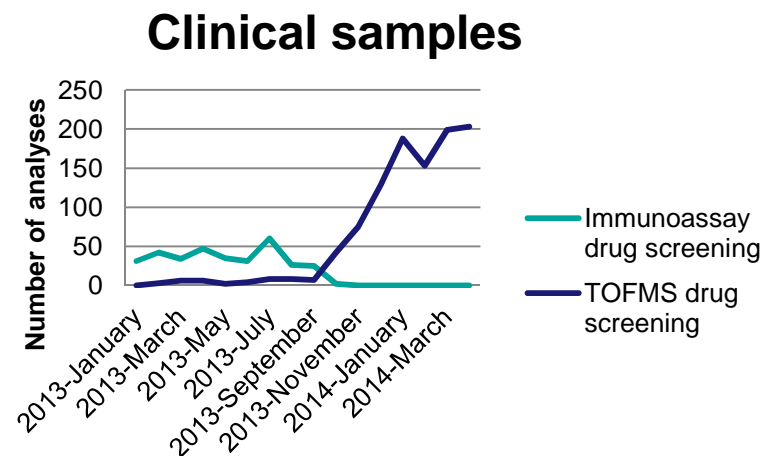
Mass spectrum for qualifier ions

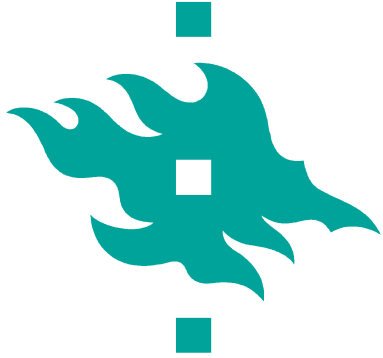




Conclusions

- UHPLC-HR-TOFMS
 - Lower number of false positives and negatives
 - The versatile analyte menu useful in the continuously changing drug scene
 - New drugs and metabolites can be easily added
 - The wider scope allowed detection of NPS and prescription drugs
 - Has replaced immunoassay drug screening in our laboratory





Thank you for your attention!