Surface Activated Chemical Ionization in the analysis of addicted drugs in human hairs by acid hydrolysis and sample dilution

Keywords: Cocaine, hairs, no sample preparation, high sensitivity

Background

Drugs abuse is a serious issue in our modern society and a sensible approach to detect it is necessary. Hair analysis can show the presence of addict drugs in the body even after many days since the time of assumption. Actually, solid phase extraction (SPE) approaches are necessary due to the low legally mandated cut-off to be detected for the analysis to be considered valid; In this work a new liquid ionization source, *Surface-Activated Chemical Ionization* (SACI), has been coupled to Brukler Daltonics' Esquire 3000 mass spectrometer to analyze. Cocaine and Benzoyletgonine extracted from human hair without using SPE but simply diluting the 10 times the extracted solution.

Surface Activated Chemical Ionization

Surface-Activated Chemical Ionization technology (SACI) [Fig 1]. constitutes a significant improvement with respect to Atmospheric Pressure Chemical Ionization sources (APCI), ElectroSpray Ionization sources (ESI) and advanced heated Ionization sources (H-ESI, Turbo-V). In particular, this new technology introduces two key innovations to upgrade current ionization sources:

1. Insertion of a metallic surface in the ionization chamber, allowing for a better ion focalization and hence for an increase in ionization efficiency [Fig 2].
2. Application of a low electric potential (usually 100 - 400 V) to this surface, causing theionization of the neutral molecules of the analyte, instead of the high potential (usually 2.000 – 5.000 V) applied to corona discharge needle that gives rise to ionization of the analyte molecules in APCI.

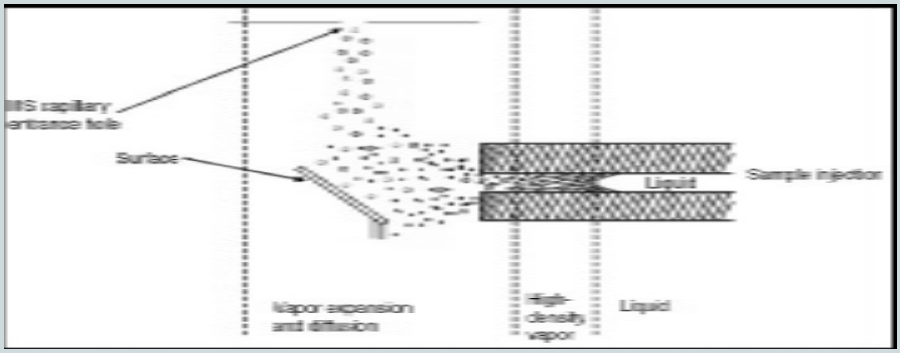


Figure 2 : SACI ionization and focalization mechanisms

Experimental conditions and settings

Internal Standard: Benzoyletgonine D3

Analyte:Cocaine and Benzoyletgonine

Calibration Curves: 0.005-5 ng/mL using Blood (R2 = 0.9856)

Sample: Hair

Sample Preparation and Analytical Conditions: Sample preparation 30 mg of hair sample were treated, for 2 hour, with 10 mL of HCl 0.1 M (acid hydrolisis)containing Benzoyletgonine D3 (20 ng/mL). 100 µL of surnatant were diluted with 900 µL of pure water so to achieve a 1:10 dilution factor for each compounds. Volumes of 20 µL per sample were injected. LC-MS/MS conditions C18 50 × 4.6 mm, 1.8 µm column was used. The chromatographic analysis was performed under gradient conditions.

The mobile phases were: (A) H2O + 0.05% HCOOH and (B) CH3CN + 0.01% HCOOH. 5% of B was maintained for 3 min a gradient was used passing from 5% of B to 25% of B in 3 min. At this eluent composition the analyte eluted. 25% of B was maintained for 5 min and in 3 min the 70% B was reached so as to eliminate wash the columns. This gradient was optimized to avoids the co-elution of analytes and matrix compounds. These conditions were maintained for 2 min and after 1 min the initial conditions were reached. Volumes of 20 µL per sample were injected.

Instruments Employed:

- Esquire 3000, Bruker Daltonics

Experimental Results

1.Mass spectra and Chromatograms

MS/MS spectra obtained with the HCT ultra mass spectrometer

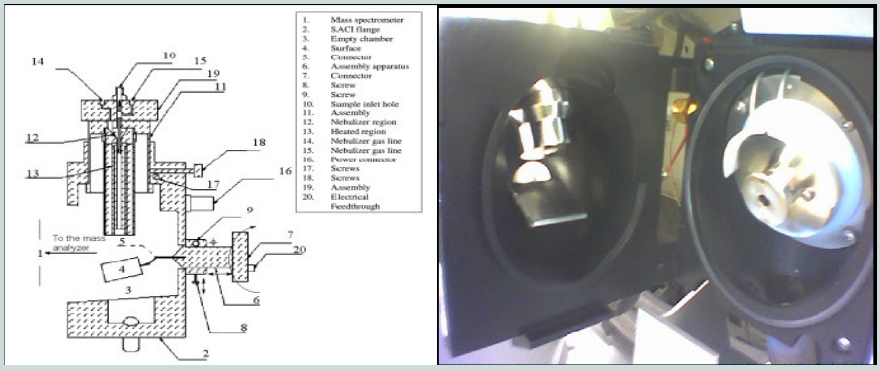


Figure 1 :SACI source .

coupled with SACI ionization technique allow to detect an extremely clear signal.

the low spectrum chemical noise makes possible to obtain high sensitivity.

SACI allows to clearly detect trace of these compounds in extracted hair solution obtained by acid hydrolysis, an application that by now can be considered unique in the market.

The analytical approach and the instruments employed allowed to achieve a significant area reproducibility and stability of LCMS/MS approach.

In particular, the use of SACI ionization, coupled with high flow chromatography, allowed to almost eliminate matrix effect, even avoiding Solid Phase Extraction (SPE).

2. Quantitative data

Bruker Daltonics' HCT ultra coupled with SACI ionization

technique allows the achievement of quantitative parameters well below legally mandated minima, thus enabling the use of this approach as a routine application for the screening of drugs in human hair

Conclusions

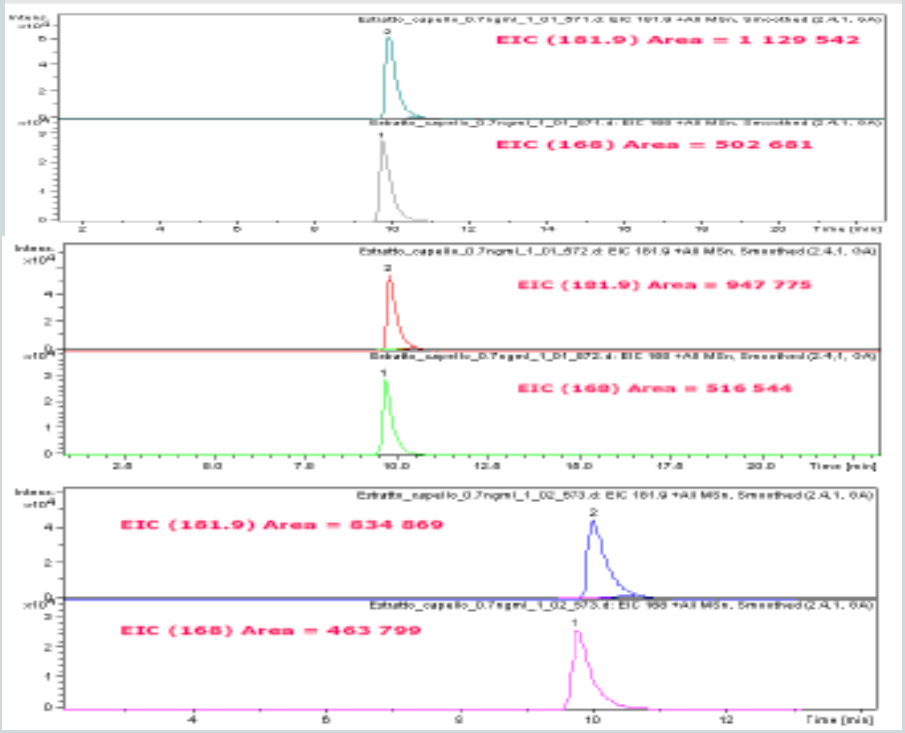
The follow benefits of SACI were observed:

High sensitivity by LC-MS/MS analysis;

Limit of quantitation below legally mandated limits;

Due to the high sensitivity of method steps of sample preconcentration or purification (solid phase extraction) are bypassed, thus cutting costs per analysis by a factor of 40%.

Bruker Daltonics S.r.l.
Via Cluentina 26/R, 62010 Macerata
Tel. 0733 283141
Fax 0733 292885
E-mail: bruker@bdal.it



Figures 3: LC-MS/MS ion extraction chromatograms and chromatographic peak areas obtained by three consecutive injection of human hairs. %area error ..... .

LC-MS/MS analysis of Benzoyletgonine (m/z transition 290→168) Concentration law limit to be detected 0.05ng/mg of hairs		LC-MS/MS analysis of cocaine (m/z transition 304→182) Concentration law limit to be detected 0.5ng/ mg of hairs	
Analytical Parameter	ng/(mg of hair)	Analytical Parameter	ng/(mg of hair)
Limit of Detection	0.002	Limit of Detection	0.016
Limit of Quantitation	0.016	Limit of Quantitation	0.15

Table 1: Limit of law to be detected and limit of detection (LOD) and limit of quantitation (LOQ) of the developed LC-MS/MS method for a) Cocaine and b) Benzoyletgonine.t

Acknowledgements and References

Acknowledgements

1) Bruker Daltonics S.r.l., Macerata, Italy

2) Bruker Daltonik GmbH, Bremen, Germany

References

Cristoni S. "Surface Actiavted Chemical Ionization" Enciclopedia of Mass Spectrometry In Press

Cristoni S, Bernardi LR, Guidugli F, Tubaro M, Traldi P. "The role of different phenomena in surface-activated chemical ionization (SACI) performance" J Mass Spectrom. 2005 Dec;40(12):1550-7.

Cristoni S, Bernardi LR, Biunno I, Tubaro M, Guidugli F. "Surface-activated no-discharge atmospheric pressure chemical ionization" Rapid Commun Mass Spectrom. 2003;17(17):1973-81.