POTENTIAL HAZARDS FOR THE USE OF SOME SOIL CONSERVATION PRACTICES: THE PRESENCE OF LINEAR ALKYLBENZENESULFONATES AND THEIR CARBOXILIC DEGRATION PRODUCTS IN AMENDED SOILS: A CASE STUDY

V. Andreu[A]*, J.L.Rubio*[A] *and Y. Picó*[B]
[A] Centro de Investigaciones sobre Desertificación-CIDE, Camí de la Marjal s/n, Apdo. Oficial, 46470 Albal, Valencia, Spain.
[B] Laboratori de Bromatologia i Toxicología, Facultat de Farmacia, Universitat de Valencia, Av. Vicent Andrés Estellés s/n, 46100 Burjassot, Valencia, Spain.

Abstract
Linear alkylbenzene sulfonates have been determined in samples of a Mediterranean forest soil, classified as Rendzic Leptosol, undisturbed and amended with three rates of composted sludges (0, 5 and 10 % w/w). Linear alkylbenzene sulfonates and their main metabolites, the sulfophenylcarboxilic acids, were extracted by sonication with a mixture of methanol-water (50:50 v/v) followed by solid phase extraction with octadecyl bonded-silica, and determined by liquid chromatography-mass spectrometry. Recoveries ranged from 38 to 103 % and the linear range of the method from 0.008 to 4.6 mg/kg with a limit of detection between 1 and 800 µg/kg extracting 5 g of soil. Data showed that the linear alkylbenzene sulfonates and sulfophenylundecanoic acid were present in untreated samples in levels from 0.2 to 1.04 mg/kg. However, samples treated with sewage sludges reached levels of these compounds between 5 and 10 times higher than those untreated. Sulfophenyloctanoic and sulfophenylhexanoic acids were also detected in the samples treated with 10 % of sludges. Quantities of surfactants found depended on the amount of sewage sludge added and the different metabolites detected in treated and untreated samples confirm that LAS undergo degradation in soil.

Additional Keywords: soil, pollution, surfactants, amended, sludges, conservation

Introduction
The use of sludges as organic amendment in the restoration of degraded soils is under debate since unknown contaminants, present in these products, may lead to a wide range of problems that could cause adverse effects in the human population (Scott and Jones 2000; Mulligan *et al.* 2001; Knepper *et al.* 2003).

Linear alkylbenzenesulfonates (LASs) are the most commonly used anionic surfactants that can pollute soil through the application of organic amendments. LASs biodegradation intermediates are sulfophenyl carboxylic acids (SPCs) (Scott and Jones 2000). An important potential toxic factor that must be taken into consideration is that other organic contaminants retained in soil are mobilized by the surfactants; there is the potential for downward or horizontal movement of the contaminants and subsequent groundwater contamination (Cho *et al*. 2002). In addition, some surfactants themselves are suspected endocrine disruptors (Knepper *et al.* 2003). However, limited studies have been performed over a long period of time to determine the levels of residual contamination that are achievable at full scale (Andreu *et al*. 2004).

Routine determination of LAS and SPCs in soils requires multiple steps that involve solvent extraction and solid phase cleanup. Chromatographic analysis of surfactants and their metabolites is typically solved by liquid chromatography coupled to mass spectrometry, which provided sufficient specificity for testifying beyond all doubt the presence of traces in complex matrices (Di Corcia 1998; Riu et al. 2001; Knepper et al. 2003; Andreu and Picó 2004).

This work describes a method that achieves fast, selective and sensitive quantification of LASs (C_{10}-C_{13}) and SPCs (C_2-C_6, C_8 and C_{11}) by liquid chromatography-mass spectrometry in soil extracts obtained by methanol-water extraction and solid phase cleanup. LASs and SPCs were monitored in samples of a Mediterranean forest soil, classified as Rendzic Leptosol, undisturbed and amended with three rates of composted sludges (0, 5 and 10 % w/w).

Materials and Methods
LASs were kindly provided by Petresa (Madrid, Spain) in a mixture with the following percentage of each homologous C_{10} (3.9%), C_{11} 37.4%), C_{12} (34.4%), C_{13} (21.3%) and C_{14} (0.2%). Phenylacetic, 2-phenylpropionic, 4-phenylbutiric, 5-phenylvaleric, 6-phenylhexanoic and 8-phenyloctanoic acids were obtained from Promochem

(Barcelona, Spain) and phenylundecanoic acid (mixture of isomers) from Acros (Geent, Belgium). Sulphuric acid, diethyl ether, sodium hydroxide, hydrochloric acid and 2-propanol were from Panreac (Barcelona, Spain). The SPCs were synthesized by sulfonation of the corresponding phenylalkanoic acids according to the procedure reported by Taylor and Nickless. Methanol (Merck, Darmstatdt, Germany) and the deionized water, obtained with a MilliQ system, were filtered through a Nylon filter of 0.45 µm Scharlau (Barcelona, Spain). The solid phase C_{18} used was acquired from Analysis Vínicos (Tomelloso, Spain).

Field Sites
Eight soil samples, of the superficial horizon (A), were taken from a burned forest area that was prepared for reforestation with a previous amendment of sludges, four of them corresponding to a zone amended with sludges in a ratio of 5 % w/w (S1) and the other four in a zone amended in a ratio of 10% w/w (S2). Four more samples were taken from a zone of the same area not affected by fire (S0). The soil belongs to Rendzic Leptosol type (FAO, 1988). This soil showed a high content in total carbonates (45.2), pH of 7.1 and sandy-loam texture (Ingelmo et al., 2003). Once in the laboratory, they were dried at room temperature, sieved through a 2 mm mesh and homogenized. The standard analytical methods were applied for the determination of the most important physical and chemical characteristics in these samples.

Analysis
Five grams of soil were extracted three times with 20 mL of methanol-water (50:50) during 20 minutes in an ultrasonic bath. The methanolic extracts were evaporated to dryness in a rotavapor to 40 ºC and 337 mbar. The dry residue was redissolved in 100 ml of hot water, acidified to pH 3, salt was added to a concentration of 0.7 M, then it is passed through a column that contained 500 mg of C_{18} previously activated with 10 ml of methanol and 10 ml of water. The retained surfactants were eluted with 10 ml methanol that was evaporated in an N_2 stream to a volume of 1 ml.

The chromatographic separation was carried out with a column Phenomenex (150 x 4.6 mm), using a gradient methanol-water and tributylamine as contraion at flow rate of 0.4 mL/mm. The detection was carried out using a mass spectrometer HP1100 equipped with an electrospay source. The conditions of the source were: voltage of the capillary, 3500 V; fragmentor 140 V; temperature and flow of the drying gas, 350 °C and 3 l/min, respectively. The EM was used in a negative ionization way (NI).

Results and Discussion
Recovery and repeatability were examined on six replicate extractions and analysis of LAS and SPC spiked soils at limit of quantification (LOQ) and ten times the LOQ level. Spiked samples were prepared by addition of 1 ml LAS and SPC standard solutions at appropriate concentrations to 5 g of soil in a 100 mL flask. The fortified samples were let to stand at room temperature for 3 hrs to achieve solvent evaporation. Results are listed in Table 1.

Table 1. Recoveries (%) and repeatability (RSD, %) for LAS and SPCs at the LOQ and 10 times the LOQ levels

Compound	Concentration, mg/kg	Recovery, %	RSD, %	Concentration, mg/kg	Recovery, %	RSD, %
C_2-SPC	--	--	--	--	--	--
C_3-SPC	0.8	76	12	8	84	8
C_4-SPC	0.6	41	19	6	38	13
C_5-SPC	0.6	97	13	6	85	9
C_6-SPC	0.4	84	16	4	103	10
C_8-SPC	0.2	84	14	2	76	9
C_{11}-SPC	0.2	94	15	2	88	8
C_{10}-LAS	0.001	83	16	0.01	90	12
C_{11}-LAS	0.001	82	17	0.01	88	9
C_{12}-LAS	0.001	87	9	0.01	83	5
C_{13}-LAS	0.01	91	10	0.1	89	10

The extraction with methanol and C_{18} achieved high recoveries for the compounds studied. Recovery and repeatability were examined on five replicate extraction and analysis of LAS-spiked soil samples. Recoveries were 38-103 %, with the exception of C_2 SPC that was not recovered. The average recovery of the overall method was 91 %, which is adequate for the demands of environmental studies.

High repeatability of the proposed extraction and analytical method was obtained. The relative standard deviation (RSD) of the overall method was < 19 %, from 5 to 19 %, average 12 %. LOQs between 1 and 800 µg/kg were obtained analyzing 5 g of soil depending of the LAS or SPC homologue. These results show that the proposed method can be used for the determination of LAS in soil samples with high accuracy. Calibration graph 0.008-4.6 mg/kg were linear over the whole range. The linear regression correlation coefficients were better than 0.997.

Figure 1 shows chromatograms of standard LASs and SFCs solutions, LAS and SFCs –spiked and unspiked soil treated with sludges at 5 % (w/w). Comparison of these chromatograms indicates that the cleanup process and LC separation were excellent.

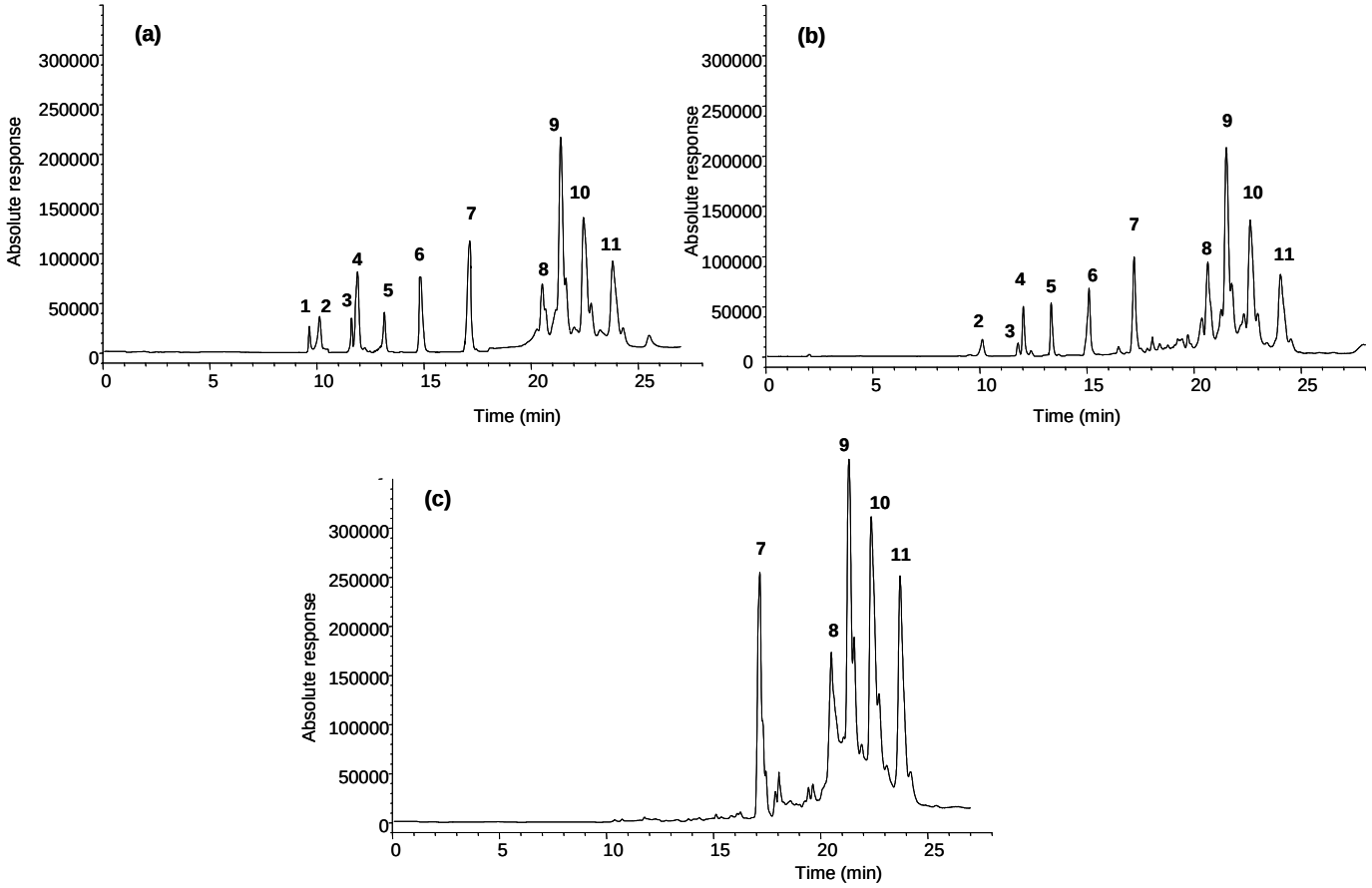

Figure 1. Chromatograms of (a) standard LASs and SFCs solution (at 10 times the LOQ), (b) LASs and SFCs-spiked soil (at 10 times LOQ), and (c) un-spiked soil treated with 5 % (w/w) sludges. Peak identification and selected ions were: 1=C_2SPC (*m/z* 215), 2= C_3SPC (*m/z* 229), 3= C_4SPC (*m/z* 243), 4= C_5SPC (*m/z* 257), 5= C_6SPC (*m/z* 271), 6= C_8SPC (*m/z* 299), 7= C_{11}SPC (*m/z* 341), 8= C_{10}LAS (*m/z* 297), 2= C_{11}LAS (*m/z* 311), 2= C_{12}LAS (*m/z* 325), 2= C_{13}LAS (*m/z* 339)

The results obtained when analyzing the samples without amendment and the samples treated with sludges are summarized in Table 2. The total concentrations of LAS in soils ranged from 2.82 to 27.04 mg/kg and of SPCs from 0.20 to 1.92 mg/kg.

These levels show that a substantial amount of LAS remains in soils, even in samples that have not been treated with sludges and confirm the higher concentrations in treated soils compared with those not amended. LASs content is related to the percentage of sludge added to the sample. Higher LAS levels were found in those samples treated with 10 g of sludges (27.04 mg/kg), in which some SPCs metabolites (from C_6 to C_{11}) were also detected

with a total content of 1.92 mg/kg. In the other samples, the only metabolite detected was the C_{11} SPC at very low levels. A study on LAS and SPCs concentrations in sewage sludges (Riu *et al*. 2001) demonstrated that large amounts of LASs and no SPC were present. That could be explained by the higher polarity of SPCs that diminish their affinity for particulate matter. However, in the present study, samples contain some of the studied SPCs. Probably because once removed from the anaerobic environment of sludge digestion and/or storage, bacteria begin to metabolise LAS.

Table 2. Concentrations (RSD %, *n* = 5) of the individual and total LASs and SPCs in soil samples

Samples	LASs (mg/kg)					SPCs (mg/kg)			
	C_{13}	C_{11}	C_{12}	C_{13}	Total	C_{11}	C_8	C_6	Total
S0	1.04 (17)	0.59 (16)	0.44 (18)	0.75 (15)	2.82	0.20 (15)	--	--	0.20
S1	6.42 (5)	2.25 (7)	1.06 (13)	2.51 (12)	12.24	0.58 (12)	--	--	0.58
S2	13.88 (19)	4.49 (19)	2.06 (18)	6.12 (14)	27.04	1.02 (17)	0.57 (10)	0.33 (13)	1.92

Conclusions

The proposed operating procedure represents a marked improvement on LC-MS applications, since it achieves high sensitivity with an unequivocal detection and confirmation of different compound structures by isolating the corresponding deprotonated molecule. In this way, this methodology validates its effectiveness showing the reliability of this new approach to determine LASs and SPCs homologues in complex environmental matrices.

LASs are present in the studied soils although they have not been treated with organic amendments. However, a correlation exists among the treatment with muds and the levels of LASs present in the samples.

Results from the current study will be useful in predicting the fate and transport of other organic contaminants, such as agrochemical or aromatic polycyclic hydrocarbons, in soils treated with sewage sludges where surfactants are present. Additionally, the results have relevance to soil remediation studies, in which surfactants could be added to mobilize contaminants or increase contaminant bioavailability.

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