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APEOs in leather processing – A survey

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Abstract

The application of alkylphenol ethoxylates (APEOs) in leather processing has been limited by restrictions such as European 2003/53/EC and 2009/563/EC Directives due to their potential to form estrogenic substances. Thus, a routine analytical method is desirable to screening these compounds in leather-making auxiliaries. However, there is still no standard method for analyzing these pollutants. This investigation illustrated the problems posed by using the conventional techniques for directly analyzing APEOs, and presented a newly developed procedure based on a special cleavage reaction with aluminum triiodide (AlI_3) to equivalently converting APEOs to the parent alkylphenol (AP). The established method was applied for screening APEOs residuals in the chemicals for socking, liming, degreasing, retanning, fatliquoring and finishing in a tannery. The results revealed the significance of auxiliaries contaminated by APEOs, as well as the possibility using the developed procedure to perform efficient screening and monitoring programs for APEOs in leather processing.

Keywords: Leather auxiliary; Alkylphenol; Alkyphenol ethoxylates; Analysis

1. Introduction

In recent years, ecological and toxicological demands are playing an increasingly important role in leather manufacture, and some chemicals have been limited for leather processing use. Alkylphenol ethoxylates (APEOs), the widely used industrial surfactants, have been listed as restricted substances by legislation due to their potential to form estrogenic substances as alkylphenol (AP) and mono- or di- ethoxylated alkylphenols [1,2]. In most cases, APEOs refer exclusively to nonylphenol ethoxylates (NPEOs) and octylphenol ethoxylates (OPEOs) because both are by far the most commonly used and encompass more than 98% of APEOs market. The most influential legislations relating to APEOs are the European Union (EU) Directive 2003/53/EC and 2009/563/EC which especially restrict the use of NPEOs in substance or preparations for leather processing. Additionally, the two legislations provide a basis for many Eco-labels and brands to introduce their own limits for the presence of APEOn in the consumer products made of leather.

Although the Directive 2003/53/EC has been followed for many years, APEOs are still possibly applied in the tannery because of their low cost and excellent characteristics. APEOs and products based on APEOs are possibly employed in the whole leather-making process because APEOs are very versatile chemicals in the auxiliary formulation. Despite that APEOs substitutes as alkyl ethoxylates (AEOs) were widely applied and led to use of APEOs decreased significantly, APEOs were still

detected in leather with high level [3]. These facts indicate a challenge to the leather industry and its suppliers. Thus, for pollution source control, analytical methods for monitoring APEOs in leather processing auxiliaries should be developed.

Numerous journal articles regarding the determination of APEOs have been published, but there is currently no officially authorized method for the routine analysis of APEOs. Methods found in the literatures used for the determination of these substances are very diverse, and the results are far ranging and difficult to compare due to variability in sample handling, analysis and detection limits [4-6]. Generally, determination of APEOs was conducted by chromatographic procedures such as liquid chromatography (LC) coupled with fluorescence detection [7-9], while one of the main problems is the limited selectivity inherent in the methods [4]. Improved analytical procedures for the selective determination of these surfactants require the use of mass (LC-MS) or tandem mass spectrometry (LC-MS/MS) [10-11]. The application of LC-MS or LC-MS/MS for the direct determination of APEOs provides a single-step analysis and seems to be a suitable analytical strategy [6]. However, LC technique can not solve the chromatographic separation of APEOs due to their extreme ranges in polarity and surfactivity based on the ethoxylate (EO) chain length which leads to the “coeluting” in one broad peak [12,13]. This problem is particularly important when simultaneously dealing NPEOs and OPEOs that can be found in APEOs-related samples. Therefore, rather than developing exotic and expensive analytical techniques for APEOs that exhibit extremes in polarity and volatility, a procedure relatively independent of EO chain length should be established to greatly simplify the analysis. One approach is to remove all the EO chains in APEOs, leaving the alkylphenol (AP) that can be easily analyzed by the general analytical tool, because the analysis of AP itself is a simple extension of the GC-MS technique described in ISO 18857-1 or EPA Method 625. Unfortunately, few literatures reported an available technique although the concept has been recognized and expressed as “total potential alkylphenols” [14-15].

In this investigation, an analytical method was investigated based on the newly developed cleavage technique for screening APEOs in various leather-making auxiliaries. This procedure involved aluminum triiodide (AlI_3) cleavage reaction combined with GC-MS detecting the yielded AP. The calculation of APEOs follows the yielded AP contents using a 1:1 molar conversion by normalization to $APEO_9$. The method developed here provides a cost effective, rugged and sensitive means of analysis using equipment that is readily found in most laboratories, allowing one to easily screen APEOs in leather processing auxiliaries.

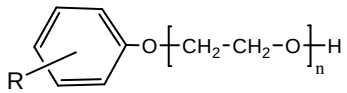
2. Experimental

2.1. Reagents and solutions

Eight kinds of APEOs identified by EO unit number and commercial name, were selected in this study, as shown in Table 1. Analytical standards of nonylphenol (NP, CAS No. 84852-15-3, technical mixture), octylphenol (OP, CAS No. 140-66-9) were purchased from Dr. Ehrenstorfer (Germany). Acetonitrile (ACN), hexane and methanol of HPLC grade were supplied by Merck (Germany). Organic free water was obtained in the laboratory using a Milli-Q system (Millipore). All other reagents used were of reagent grade.

Stock solutions of each APEOs, as well as NP and OP, were separately prepared in ACN at a concentration of 1.0 mg/mL. These stock solutions were diluted to appropriate concentrations using ACN. All solutions were stored in darkness at 4 °C.

Table 1 APEOs used in this study: commercial name, alkyl chain, average number of EO units (n, Avg) and appearance.



No.	Commercial name	Alkyl chain / EO units (n, Avg.)	Appearance
1	Igepal-210	R = Nonyl, n ≈ 1.5	Pale yellow liquid
2	Tergitol™ NP-9	R = Nonyl, n ≈ 9	Yellow liquid
3	Tergitol™ NP-15	R = Nonyl, n ≈ 15	Waxy white solid
4	Tergitol™ NP-30	R = Nonyl, n ≈ 30	Waxy white solid
5	Triton™ X-15	R = Octyl, n ≈ 1.5	Yellow liquid
6	Triton™ X-45	R = Octyl, n ≈ 4.5	Yellow liquid
7	Triton™ X-100	R = Octyl, n ≈ 9	Pale yellow liquid
8	Triton™ X-165 [a)]	R = Octyl, n ≈ 16	Pale yellow liquid

[a)] with purity 70% (w/w).

2.2. Basic cleavage reaction

On the basis of chelation mechanism, the cleavage of Aryl-O-alkyl ether by AlI_3 was suggested to be a stepwise process [16]. The reaction is illustrated by the following scheme where Ar-O-R is APEOs. The reaction has been approved to be fast in ACN solution [3].

$$\text{Ar-O-R} \xrightarrow{AlI_3} \text{Ar-O-}AlI_2 + \text{R-I} \qquad (1)$$

$$\text{Ar-O-}AlI_2 \xrightarrow{H_2O\,/\,H^+} \text{Ar-OH} \qquad (2)$$

2.3. Preparation of the cleavage agent

AlI_3 is commercially available, and it can also be easily prepared in the laboratory by simply mixing aluminum metal and iodine in ACN [3]. The effective mass of the white cleavage agent is determined by weight analysis and the regent is recommended to more than 2 g for the following use.

2.4. Sample test

2.4.1. Preparation of samples

The leather processing auxiliaries selected here are employed in soaking, liming, degreasing, retanning, fatliquoring, dyeing and finishing. For standard experiments, a sample of liquid auxiliary (1.0 g) was diluted with 2×10 mL ACN and then anhydrated over anhydrous calcium sulphate (~1 g) bed. After that, the extracts were filtered through a 0.45 μm membrane filter and filled to 25.0 mL with ACN. Considering that fat/oil is commonly the strong matrix which can affect the instrumental analysis and the cleavage reaction, sample extracts were divided into two groups as follows:

Group A: without fat/oil contained auxiliaries (except fatliquor).

Group B: only fatliquors.

2.4.2. Cleavage procedure

Cleavage of APEOs was accomplished according to the previous work [3]. Briefly, 10.0 mL of the sample extracts were pipetted into the flask containing 2 g AlI_3 in ACN. The contents were then treated under refluxing at 90 ^{o}C for 10 min to complete the cleavage reaction. When analysis was required, the solution was subjected to the following work-up.

2.4.3. Work-up prior to analysis

Water was dropped into the reaction mixture until no boiling occurred. Then, another 30 mL of water was added and the mixture was acidified with 1 mL HCl (0.1 M). The yielded AP was isolated with 2×10 mL hexane extractions. The hexane extracts were then de-iodinated with 2 mL $Na_2S_2O_3$ (saturated), washed with 2×30 mL distilled water, dehydrated with anhydrous Na_2SO_4, and finally filled to 25.0 mL. The solution was ready for GC-MS analysis described below.

2.5. Calibration curves

The calibration curve was made by plotting six pairs of the amounts of given $APEO_9$ and the contents of the yielded AP according to the described procedure above. The $APEO_9$ (Tergitol NP-9 and Triton X-100) was chosen as calibration standards because APEOs for cleaning products usually have an approximately Poisson Distribution of nine-mole ethoxylates. The pairs were fitted by linearship according to the following formula: $C_i = k\,A_i + p$ where C_i corresponds to the given APEOs content; A_i corresponds to the area response of isolated AP by GC-MS which represents the contents; p corresponds to the ordinate intercept of the linear curve and k is the slope. The correlation coefficient r^2 was calculated from the linear regression to evaluate the dependence of the yielded AP amounts on the given APEOs contents.

2.6. Instrumental analysis

Agilent 7890A GC equipped with 5975 MS operated in the positive electron impact mode (EI+) was used for analysis. The GC separation was performed on a fused silica capillary column (DB-5 ms; film thickness, 0.25 μm; 30 m * 0.25 m i.d. (J&W Science)). The GC-MS conditions were as follows:

carrier gas, helium; injector temperature, 250 °C; and transfer line temperature, 280 °C. The ion source temperature was adjusted to 150 °C. For analysis, 1 μL splitless injections with purge time 0.8 min were made by the autosampler. GC conditions were programmed as follows: initial oven temperature 80 °C, held for 1 min, followed by a temperature ramp of 10 °C/min to 280 °C, and held for 10 min.

For the identification of NP and OP, mass spectrums of the available authentic standards at the same retention times were used, as well as comparison with mass spectrums in other publications [17,18]. Confirmation of analytes was carried out in full scan mode, and data were acquired from m/z 50 to 300. For the quantitation purposes, chromatograms were registered using selective ion monitoring (SIM) of the main characteristic fragment ions. The selected ions for OP were at m/z 135 and 206, and for NP at m/z 107, 121, 135 and 149.

3. Results and Discussion

3.1. Cleavage of APEOs

The generality of AlI_3 cleaving APEOs to yield AP was first investigated with the APEOs as substrates (see Table 1). The results revealed that NPEOs and OPEOs were successfully converted to their corresponding NP and OP, and no ethoxylated AP ($APEO_1$ or $APEO_2$) were detected, as listed in Table 2. The representative chromatograms and mass spectrums of the yielded NP and OP were presented in Figure 1 and Figure 2.

Table 2 Cleavage of APEOs by AlI_3 and the calibration data [a)]

No.	Substrate / APEOs	Detected product	Calibration data		
			$p\times10^{-2}$	$k\times10^{-4}$	r^2
1	Igepal-210 ($NPEO_{1.5}$)	NP	-2.43	4.19	0.992
2	Tergitol™ NP-9 ($NPEO_9$)	NP	2.41	9.39	0.993
3	Tergitol™ NP-15 ($NPEO_{15}$)	NP	4.63	12.5	0.990
4	Tergitol™ NP-30 ($NPEO_{30}$)	NP	-4.52	21.5	0.991
5	Triton™ X-15 ($OPEO_{1.5}$)	OP	-1.07	3.18	0.998
6	Triton™ X-45 ($OPEO_{4.5}$)	OP	-0.43	4.73	0.997
7	Triton™ X-100 ($OPEO_9$)	OP	1.66	6.93	0.998
8	Triton™ X-165 ($OPEO_{16}$)	OP	1.06	9.67	0.996

[a)] The experiments were carried out with 10.0 mL of APEOs in ACN which substitute the sample extracts. Content range of APEOs: 10 ~ 300 μg for all the substrates. All calibration curves were fitted by linership.

The correlation coefficients r^2 were calculated from the liner calibration curve by testing six levels of increasing concentrations of each APEOs, as summarized in Table 2. All the calibration curves were fitted well by linear regression with r^2 more than 0.99, indicating the dependence of the yielded AP on the given APEOs in a wide range of concentrations for all those compounds, as well as the stability of AP during the cleavage process. At the same time, the values of k, which related to the number of EO units in APEOs, were varied with different NPEOs and OPEOs. These indicated the general use of the special cleavage technique for analyzing these substances.

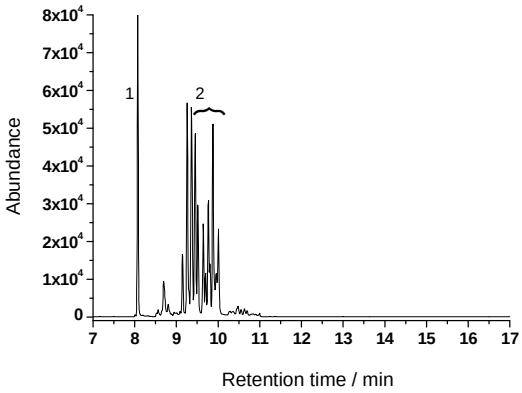


Figure 1. Representative GC-MS/SIM chromatogram of NP (peak 1, 8.086 min) and OP (peak 2, 9.17 -10.05 min) originated from the parent NPEOs and OPEOs by AlI_3 cleavage reaction under the optimized conditions.

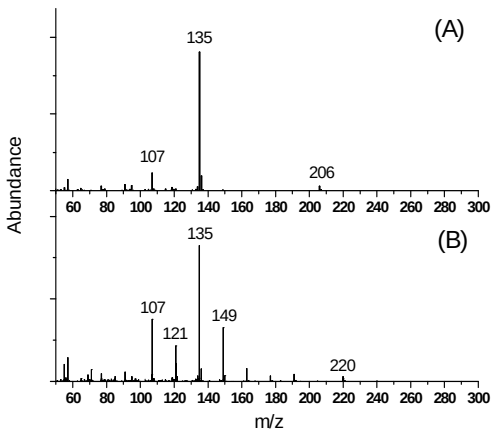


Figure 2. Mass spectrum (EI) corresponding to Figure 1. (A) For OP and (B) for NP.

3.2. Matrix effect studies

Commercial leather-making auxiliaries are commonly complex mixture of organics and in-organics which might disturb the cleavage process. For liquid products, water is the main component. Considering the easy hydrolysis of AlI_3 in water and lead to the extra consumption of AlI_3, these sample extracts were dehydrated with $CaSO_4$ prior to cleavage. The impact of matrix effects on the present cleavage results was examined by spiking experiments with Tergitol NP-9 and Triton X-100 as surrogate standards. These sample extracts have been screened according to the described procedure, and no AP was detected.

Table 3 shows the test results of samples in Group A which consist of ACN-extract from these auxiliaries except fatliquors. Regardless of the different component in these auxiliaries, all the $APEO_9$ ($NPEO_9$ and $OPEO_9$) were converted into their parent AP, with recoveries ranging from 80~95%.

These figures mean no disturbance of matrix on the cleavage process, as well as on the instrumental analysis.

Table 3 Recovery and detection limit of $APEO_9$ in Group A

Auxiliary agent [a)]	Recovery (%)		Detection limit (mg/kg)	
	$NPEO_9$	$OPEO_9$	$NPEO_9$	$OPEO_9$
Soaking	95.1	92.3	80	40
Liming	83.4	93.1	90	40
Degreasing	92.7	88.7	80	40
Retanning	84.8	92.8	100	50
Liquid dyestuff	80.6	90.4	80	40
Pigment	85.5	88.3	100	50

[a)] Only one product for the type was tested, and each test was performed in duplicate. 100 μL of $APEO_9$ in ACN (1.0 mg/mL) was added to the sample extract (10.0 mL).

The presence of protic impurites, exemplified by fat/oil, in Group B, does not exert a bad influence on the completion of cleavage (see Table 4). The three recoveries of $NPEO_9$ more than 100% lie in the GC-MS spectra of multi-peak pattern (see Fig. 1) because of the branched nonyl- side chain. The data indicates the matrix effect on the instrumental analysis of NP, but the negative influence is not obvious. For $OPEO_9$, the recoveries ranged from 80~96% which are consistent with the results in Table 3.

Table 4 Recovery and detection limit of $APEO_9$ in Group B

Fatliquor [a)]	Recovery (%)		Detection limit (mg/kg)	
	$NPEO_9$	$OPEO_9$	$NPEO_9$	$OPEO_9$
Sulfitated fish oil	108.1	96.3	100	40
Chlorinated paraffin	88.8	84.1	100	40
Alkyl alcohol phosphate	105.6	90.2	120	50
Sulfitated rape oil	82.3	80.4	100	50
Sulfated castor oil	110.6	93.7	120	50
Sulfated neatsfoot oil	85.5	82.3	120	50

[a)] Only one product for the type was used, and each test was performed in duplicate. 100 μL of $APEO_9$ in ACN (1000 mg/L) was added to the sample extract (10.0 mL).

The detection limits estimated by the signal-to-noise of 3:1, as well as weight of the sample and volume of the extracts, range from 80~100 mg/kg for $NPEO_9$ and 40~50 mg/kg for $OPEO_9$ in Group A (see Table 3). The values increase to 100~120 mg/kg for $NPEO_9$ in the fatliquors (see Table 4) mainly due to the matrix effect (fat/oil). These detection limits easily attain the purpose of monitoring NPEOs in leather auxiliaries restricted by EU Directive 2003/53/EC (<1000 mg/kg). Besides, the detection limits can be further improved to ~5 mg/kg by evaporating the final extracts volume. However, for the purpose of convenience, the solvent removal process was not applied in the analysis.

3.3. Analysis of leather auxiliaries

The developed procedure was applied for screening APEOs in the leather auxiliaries which were all obtained from one tannery. These samples were considered to contain APEOs due to the fact that the finished leathers were found seriously contaminated by APEOs (1500 mg/kg) in our previous study [3]. The total of 23 auxiliaries were analyzed and three were found to contain APEOs, as shown in Table 5. The detected concentrations ranged from 10^4 to 10^5 mg/kg for $NPEO_9$ and 1200 mg/kg for $OPEO_9$. The highest concentration of NPEOs (10^5 mg/kg) was in a degreasing agent.

Table 5 APEOs found in leather auxiliaries

Auxiliary agent	Sample Number (n)	Number of APEOs detected		Conc. range (mg/kg)	
		NPEOs	OPEOs	$NPEO_9$	$OPEO_9$
Soaking	2	0	0	/	/
Liming	2	0	0	/	/
Degreasing	2	1	0	~10^5	/
Retanning	4	1	0	~10^4	/
Fatliquoring [a)]	8	1	1	~10^4	1200
Liquid dyestuff	3	0	0	/	/
Pigment	2	0	0	/	/

[a)] one sample contains both NPEOs and OPEOs.

Up until now, the survey of APEOs in leather processing in the tannery, could be ceased. Although the pollution sources were found, it was still not possible to identify an exact correlation between the levels of APEOs measured in chemical products and the APEOs content of the leather manufactured with them, and this problem would become even more complicated when several auxiliaries contained APEOs, as presented in Table 5. In this way, the best soluton is to phase out APEOs from all of the auxiliaries by the established method.

4. Conclusions

To summarize, the successful conversion of APEOs in leather auxiliaries to the parent AP, was achieved by using AlI_3 as cleavage agent. The advantages are: (1) complete de-ethoxylation, (2) rapid treatment (~10 min), (3) easy operation with simple apparatuses (GC), and (4) inexpensive chemicals. Thus, this method can be adopted as the on-site screening method for laboratory-scale APEOs -containing chemicals used for leather making process.

The established procedure above has been arranged and submitted to IULTCS (IUC) for circulation (Draft - CEN/TC 289/WG1 & 2 & 3: N 222), and expected for inter-lab experiment and validation.

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