SUPERCRITICAL FLUID EXTRACTION OF POTENTIAL MIGRANTS FROM PAPER AND BOARD INTENDED TO USE AS FOOD PACKAGING MATERIALS

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ABSTRACT

The optimization of Supercritical Fluid Extraction (SFE) using CO_2 for the extraction of contaminants in 15 samples of recycled Paper and board (P&B) has been studied. An experimental design has been used for simultaneous optimization of the variables involved in both the extraction step and the collection of the extract. Methanol was used as modifier. Several plastisizers such as Diethyl phthalate, Diisobutyl phthalate, Di-n-butyl phthalate, Dioctyl adipate and Diethylhexyl phthalate (from 2 to 100 μg/ g of paper) have been found in the recycled P&B samples. A discriminate analysis applied to all results obtained allow to classify the samples in three different groups according to both the content of recycled pulp (0 %, 10–30 % and > 80 % of recycled pulp), the sample thickness (from < 300 to > 600 μm) and the surface treatment of the paper. The analytical behavior as well as the results are discussed.

INTRODUCTION

Paper and board (P&B) are mainly composed by cellulose, hemicellulose and lignin and also contain some additives as well as coating materials, which provide specific properties to the final material. The use of P&B as food packaging material is well extended in the Food Industry, as primary packaging in direct contact with dried foodstuffs and as secondary packaging too.

But the materials used in contact with food have to accomplish a series of conditions to guaranty their safety in use *(1)*. Several regulations exist *(2)* to control the presence and concentration of potential migrants in order to protect the consumer health.

Because of the environmental pressure, recycled P&B is being more and more used in a wide variety of applications, and the food packaging is one of the ongoing approach which could be explored. Recently, the Council of Europe has proposed to limit the concentration of several potential migrants in recycled paper and board intended to use as food packaging materials and the proposed limits depend on the type of food. Without doubt, the use of any recycled materials in contact with food involves the previous analysis of the materials, in order to know the potential migrants which could be transferred to the food *(3)*.

However, one of the critical points in the analysis of migrants is the extraction of the paper samples. Most of the published papers about the analysis of components in paper and board employ the solvent extraction with ethanol *(4)* or toluene, both by direct immersion or using the Accelerated Solvent Extraction procedure (ASE) *(5)*.

One attractive approach in this context is the use of Supercritical Fluid Extraction (SFE) with CO_2 as supercritical fluid, which can provide a fast and efficient extraction of a wide range of compounds. SFE has been shown as an excellent extraction system of different matrices *(6-16),* but it has not been explored yet for paper and board.

This paper shows the behavior of SFE for the extraction of several paper and board samples of different composition, including virgin and recycled fiber. Many variables are involved when working with SFE and crossed interactions between the variables are frequent *(17-19)*. For this reason, an experimental design able to optimize simultaneously all the variables involved either in the extraction step or in the collection of the extract has been applied *(20)*. On the other hand, paper samples are complex matrices and a high variety of compounds can be extracted *(21,22)*. To evaluate the analytical power of SFE in this type of samples as well as the relationship between the extracted compounds and the pattern recognition, a discriminate analysis has been applied to the final results.

The analytical results are compared with those obtained by solvent extraction using ethanol, according to the CEN method *(4)*. The relationship between the SFE ability and the rate of recycled pulp of the samples is discussed too.

EXPERIMENTAL SECTION

Apparatus

A Prepmaster (Suprex Corp. Pittsburgh, PA) stand-alone Supercritical Fluid Extraction system, supplied by Varian and equipped with the Accutrap module was used for all the extraction/collection experiments. The cell was filled in sandwich mode, using glass wool on both the top and the bottom and anhydrous sodium sulfate was added as dryer agent. The Accutrap module was equipped with a cartridge of microspheres of silanized glass (80/100 mesh, Suprex) inserted in a cryogenic module (cooled with CO_2). A pump was used to elute the cartridge with the appropriate solvent.

A Gas Chromatography (GC) Hewlett-Packard 6890, coupled to a mass selective (MS) detector Hewlett-Packard 5973 with a GC capillary column SGL-5 (60 m x 0.25 mm I.D. and 0.25 µm film thickness) were used. The analyses were carried out in both SCAN and SIM mode. He C-50 (Carburos Metálicos, Spain) was used as carrier gas.

Reagents and solutions

Diethyl phthalate (DEP), Diisopropylnaphthalenes (DiPN's), Diisobutyl phthalate (DiBP), Di-n-butyl phthalate (DBP), Dioctyl adipate (DOA) and Diethylhexyl phthalate (DEHP) (Aldrich) were used as chromatographic standards for quantitation. Standard solutions of these standards in methanol at the appropriate concentration were used for calibration. All the standards and solutions were gravimetrically controlled. Standard addition procedure was used for the quantitative analysis of the 15 samples. Three replicates of each sample were analyzed.

Analytical procedure

All the analyses were done at constant flow of 1 mL/min. 1 µL of extract was injected in splitless mode, using an Hewlett Packard-Autosampler 7673 GC. The GC program used was as follows: injection temperature 260 ºC, oven temperature: initial temperature 65 ºC for 2 min and then, 5 ºC/min rate to 300 ºC and held for 6 min. MS conditions were as follows: mass range from 35 to 700 m/z (SCAN mode). The following characteristic masses were selected (SIM mode): 149 m/z (diethyl phthalate, diisobutyl phthalate, Di-n-butyl phthalate and diethylhexyl phthalate), 197 m/z (diisopropylnaphthalenes) and 129 m/z (dioctyl adipate).

Extraction procedure with ethanol

Small pieces of 1 x 1 cm size of each paper sample were exactly weighed and placed in a vial of 20 mL, and 2 mL of Ethanol (1.5 g) were added to the vial. The vial was closed and shacked for 30 min at room temperature. Then, these extracts were directly analyzed by GC-MS.

Supercritical fluid extraction procedure

The optimum conditions for collection and extraction procedures are listed in Table 1. Supercritical fluid extraction analysis does not require previous digestion, dissolution or special treatment of the samples. Each paper sample was extracted using the optimum conditions. The SFE extracts were directly analyzed by GC/MS.

Samples

Table 2 lists the characteristics of the 15 samples analyzed. Paper European Companies supplied all of them and they have been coded to avoid the pattern identification.

RESULTS AND DISCUSSION

Experimental design

The optimization of an analytical procedure usually involves a series of experiments in which the variables affecting the process are systematically studied one by one. However, when the number of variables increases, the required number of experiments is very high. Besides that, the mutual interaction within the variables is not clear and additional experimental work is required. A good system to minimize the total number of experiments is the use of a mathematical model which is called "experimental design", which consists of considering simultaneously all the variables which influence the process, each one having different range of application.

Among the different mathematical procedures, the Factorial Design is one of the well-known procedures. The total number of experiments is expressed by $N = n^K$, where "n" is the number of considered levels and "K" is the number of factors (variables) involved in the study. The extreme values, which are defined by (+) and (-) correspond to the maximum and minimum values of the fixed interval for each factor. Two levels were designed for the study and consequently, $N = 2^K$.

Also the variables can be divided in two different categories: qualitative which are discrete variables, and the quantitative ones, which have continuous variation in the selected interval. The system of coordinates is dimensionless being (+1) and (-1) the highest and lowest values respectively and being "cero" the central coordinate in all cases. The central point corresponds to the coordinate origin and then the matrix of the experimental design is prepared taking into account all the variables involved in the process. It is possible that the interaction between factors were more important than the factors themselves. For this reason, the terms of interaction have to be included in the model. Also other terms (square and cubic terms) as well as those more complex terms could be considered too.

Two different experimental designs were applied to optimize the SFE procedure, one for the extraction itself and another for the collection of the extract. To reduce the total number of experiments, a fractional factorial design was applied in each case, using the computer program Modde 4.0 for Windows.

The variables, which influence the extraction step, can be classified according to the process as follows:

(a) Variables which affect the supercritical fluid: nature of modifier; nature of supercritical fluid and how to use it (dynamic or static mode); percentage of modifier to be used and flow of supercritical fluid.

(b) Variables which concern to the solid sample: sample weight; size of the sample and how to fill the sample cell (sandwich mode, glass wool, etc.).

(c) Variables affecting the extraction conditions: extraction mode (static or dynamic); time of each extraction step (in both dynamic and static mode); extraction temperature and pressure.

A series of potential variables, were initially fixed, as well as those affecting the collection module, to optimize the extraction step. It is worth to point out that the optimization procedure was carried out in this case using real paper samples as starting matrix. The common system is to use standards or reference materials for optimization purposes, but in the case of paper samples no reference materials are available, and the use of pure standards in absence of the paper matrix is not representative enough. On the other hand, the influence of the matrix in SFE is probably higher than in the other analytical procedures. For this reason, it was considered to be more appropriate to work with real paper samples from the beginning. The maximum amount of extracted compounds was used as criterium for optimization. Table 3 shows the matrix used to optimize the extraction step.

The final extract was adsorbed on an inert trap consisting of a cartridge filled with microspheres of sinalized glass. Once the extraction was finished, the cartridge was eluted with a solvent. The following factors were optimized in the collection step:

(a) Those affecting the solvent; such as the nature of solvents; flow through the cartridge; final volume (weight) of solvent and the number of washing steps after each sample extraction.

(b) Factors which influence the process: restrictor temperature; temperature of the cartridge during the adsorption step (adsorption temperature); and temperature of the cartridge during the elution (desorption temperature).

As in the previous step, some of these variables were fixed and the rest were optimized, as shows Table 3.

The optimum values obtained for the global process (extraction + collection) are shown in Table 1. In these conditions the maximum amount of compounds was extracted from the paper samples.

Qualitative analysis

Fifteen different samples of paper and board were analyzed by SFE-GC-MS using the optimized procedure above described. Figure 1 shows the chromatograms of one virgin sample (R11) and one 100% recycled sample (R1), which can be considered as representative of those virgin and recycled samples respectively, analyzed by SFE and liquid extraction with ethanol.

As can be seen, the compounds extracted at higher concentration were the plastisizers (phthalates). Comparing the SFE extraction with the liquid extraction using ethanol applied to the same sample (Figure 1), similar qualitative analysis was obtained. Looking through the chromatograms of all the samples extracted, it can be seen that the behavior of the recycled samples was very similar within the group, while the concentration of all the analyzed compounds was much lower in the virgin samples. This fact agrees with the idea of a stronger penetration of the supercritical fluid into the recycled pulp. Table 4 lists the quantitative results obtained by SFE-GC-MS analysis. It was expected too that the content of plasticizers and other contaminants were much lower in virgin samples, and the experimental data confirm that.

According to the Table 2, recycled paper samples could be classified in three different groups:

- Virgin samples.

- Those samples containing between 10 and 30% of recycled pulp.
- Samples containing more than 80% of recycled pulp.

The qualitative profile of the samples corresponds to these groups, with similar contaminants within each group of samples.

Quantitative analysis

Table 4 shows the quantitative results obtained by SFE-GC-MS. Similar values were obtained from ethanol extraction followed by GC-MS, with the exception of samples R8, R13 and R15 in which ethanol extraction provided higher values of some compounds. Table 5 shows the quantitative results obtained by EtOH-GC-MS.

As these samples belong the group of recycled pulp it seems that a more polar extraction agent is required with recycled fibers. It is clear from Tables 4 and 5 that the recycled samples have a higher concentration of contaminants.

To distinguish from the data the relationship between the characteristics of the samples and the quantitative data of contaminants, a discriminate analysis has been applied to all the data. The software SPSS was used to classify the groups, and different statistical functions such as the Fisher function allowed the classification.

Using all the analytical data from SFE-GC-MS analysis and EtOH-GC-MS, the Figure 3 a) was obtained. As can be seen, three different groups are well classified, which correspond to the three groups of samples above mentioned.

All the data either from ethanol extraction or from SFE gave the same profile. Other characteristics such as the "grammage" and the surface treatment of the samples could influence the extraction of contaminants and consequently, their migration. Taking into account the analytical data and the grammage and applying again the SPSS statistical program, Figure 3 b) was obtained. A clear distribution in three different groups was found, which correspond to the three different levels of "grammage", lower than 100 g/m^2, between 100 and 300 g/m^2 and higher than 300 g/m^2. Similar groups were obtained from both ethanol and SFE extraction.

Concerning the surface treatment, the Figure 3 c) shows three groups again, although in this case the number of samples with surface treatment is very short. According to these results it is clear that the characteristics of the samples play an important role in the extraction of contaminants from the paper samples, which means that they likely affect the migration ability to both food or food simulants in contact with the paper. If paper and board are intended to be used as food packaging materials, the migration values are very important and the usual migration units are expressed on mg/dm^2. If the quantitative data are converted to the migration units, the samples are grouped again in three different classes which correspond to the recycled, partially recycled and virgin samples. The "grammage" has also a strong influence, which means that the migration is also affected by the sample thickness. These results suggest that migration values can be compared only if the sample thickness is similar within the series of samples.

Concerning the potential migration, it can be said that SFE is a good technique to evaluate it, assuming 100 % of migration, as the efficiency of this extraction technique is very good.

CONCLUSIONS

Several interesting conclusions can be emphasized from the study carried out:

1- SFE is a good procedure to extract the contaminants present in paper samples. This extraction technique easy to use and with minimum handling and time consuming, has been shown very efficient for paper analysis.

2- The lower amount of organic solvents (< 2 mL) required in SFE for an efficient extraction of contaminants from paper samples, compared with the liquid extraction, confirms SFE as a safer and environmental friendly technique.

3- The SFE procedure should be optimized following an experimental design, so that all the variables involved in the process as well as the interaction between the different variables, were optimized at the same time.

4- The collection system of the final extract from SFE is very critical, and a polar solvent such as methanol is required to elute the more polar compounds. This way, the SFE extraction provides similar results to those obtained by liquid extraction using ethanol.

5- The characteristics of the paper samples affect the content of contaminants in the samples and such contaminants are potential migrants. SFE provides a good way to evaluate the potential migration of chemicals to food and, assuming 100 % of migration, additional migration test could be avoided.

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Table 1. Optimum values for the global process SFE.

Extraction step						
Step	*Mode*	*CO_2 flow*	*Time*	*Modifier*	*P (atm)*	*T (ºC)*
Eq[1]	St[2]	0		no	425	120
1	Dy[3]	0	5 min	no	425	120
2	Dy[3]	1 mL/min	5 min	10%	425	120
Collection step						
Solvent volume		*Trap flow*		*T restrictor*	*T ads[4]*	*T des[5]*
1.2 mL		1 mL/min		85 ºC	0 ºC	50 ºC

[1] Eq = equilibrium step, [2] St = static step, [3] Dy = dynamic step, [4] T ads = adsorption temperature and [5] T des = desorption temperature.

Table 2. Characteristics of the paper and board samples analyzed.

Code	Type	Pulp	% Recycled	Surface treatment	"Grammage" (g/m^2)	Thickness (µm)
R1	Testliner	Recycled	100	no	128	191
R2	Liner	Unbl. Kraft + recycled	10-30	no	178	234
R3	Liner	Unbl. Kraft + recycled	10-30	Dispersion	145	188
R4	Liner	Unbl. Kraft + recycled	10-30	LDPE/PET	180	214
R5	Fluting	NSSC + recycled	30	no	107	209
R6	MG-paper	Bleached Kraft	0	no	71.5	82
R7	Toilet tissue	Recycled	100	no	33.5	145
R8	Kitchen towel	Recycled	100	no	46.7	188
R9	Liquid board triplex	Bl. Kraft + CTMP	0	no	273	486
R10	Liquid board triplex	Bl. Kraft + CTMP	0	LDPE	314	508
R11	Liquid board triplex	Bl. Kraft + CTMP	0	no	267	478
R12	Liquid board triplex	Bl. Kraft + CTMP	0	PE/Al/PE	358	549
R13	White lined chipboard	Recycled (GD2)	80	clay coating	497	719
R14	White lined chipboard	Recycled (GD2)	80	PE coating	522	739
R15	Chipboard	Recycled	100	no	406	576

Unbl. Kraft = unbleached Kraft; Bl. Kraft = bleached Kraft; CTMP = chemi-thermomechanical pulp, and NSSC = neutral sulphite semi-chemical pulp and GD2 = multiply packaging boards (2 layers)

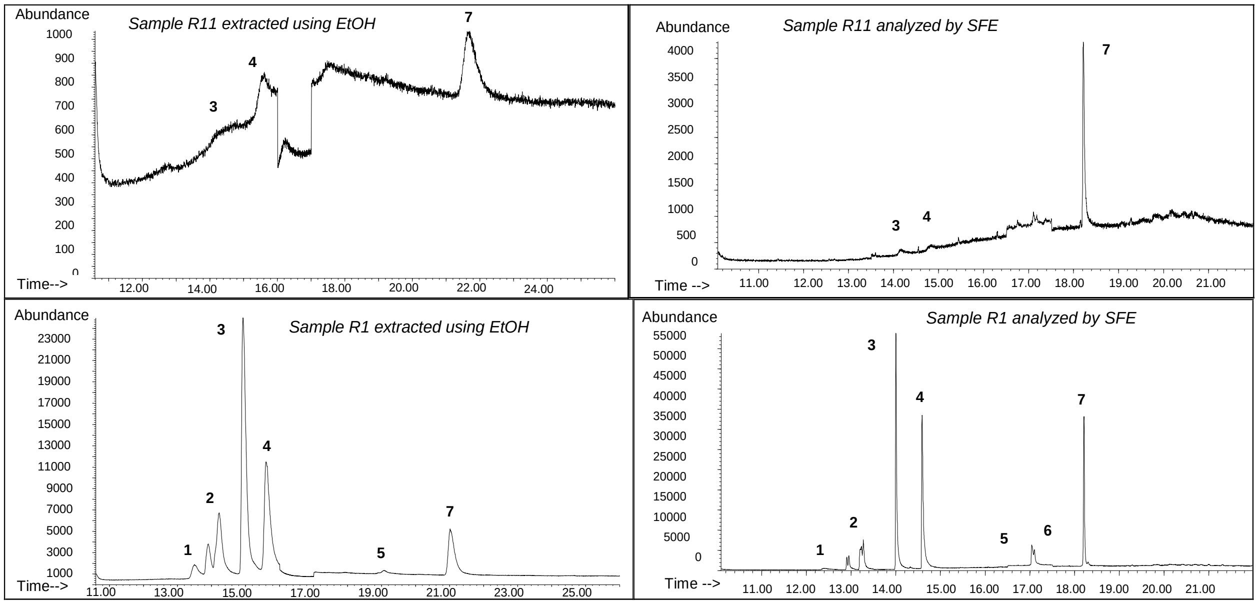
Table 3. Matrix used to optimize the extraction and collection steps.

EXTRACTION STEP optimization								
Variables optimized	*Pressure (atm)*		*Temperature (ºC)*		*SF Flow (mL/min)*		*Modifier (%)*	
	Range	Optimum	Range	Optimum	Range	Optimum	Range	Optimum
	200-425	425	50-120	120	1-3	1	0-10	10
Variables fixed	Nature of supercritical fluid						CO_2	
	Nature of modifier						Methanol	
	How to use modifier (mode)						Dynamic	
	Sample weight						1 g	
	Extraction cell volume						5 mL	
	How to introduce sample						Sandwich	
	Static step time						5 min	
	Dynamic step time						5 min	
Collection step	*Restrictor T*[1]		*Tads*[2]	*Tdes*[3]	*Solvent*		*Flow*	*Volume*
	85 ºC		- 20 ºC	40 ºC	methanol		1 mL/min	1.2 mL

COLLECTION STEP optimization				
Variables optimized	*T adsorption (ºC)*		*T desorption (ºC)*	
	Range	Optimum	Range	Optimum
	- 30 - 0	0	10 - 50	50
Variables fixed	Solvent flow		1 mL/min collection step	
			2 mL/min clean-up step	
	Solvent volume		1,2 mL	
	Whashing step number		2	
	Restrictor temperature		85 ºC	
Extraction step	*Oven temperature*	*Pressure*	*SF flow*	*% Modifier*
	120 ºC	425 atm	1 mL/min	10

[1] Restrictor temperature (ºC), [2] Adsorption temperature (ºC) and [3] Desorption temperature (ºC).

Figure 1 a) Chromatograms virgin paper (R11) and 1 b) Chromatograms recycled paper (R1) analyzed by SFE and liquid extraction.



(1) Diethyl phthalate; (2) Diisopropylnaphthalenes-isomers; (3) Diisobutyl phthalate; (4) Di-n-butyl phthalate; (5) Butyl benzyl phthalate; (6) Dioctyl adipate and (7) Diethyl hexyl phthalate.

Table 4. Quantitative results obtained by SFE-GC-MS analysis and using the standard addition procedure. Values expressed as µg/g paper.

Compounds	*R1*	*R2*	*R3*	*R4*	*R5*	*R6*	*R7*	*R8*	*R9*	*R10*	*R11*	*R12*	*R13*	*R14*	*R15*
DEP	5.0	0.7	0.9	0.3	-	-	0.9	-	-	-	0.5	-	1.1	0.7	-
DiPN's	17.4	12.4	12.2	0.3	4.3	-	1.1	2.4	-	-	-	0.2	5.1	6.4	49.0
DiBP	18.1	32.5	27.0	1.4	32.5	1.6	3.4	1.3	-	-	1.2	0.4	9.0	14.3	16.0
DBP	14.4	7.4	7.9	0.5	4.5	0.6	2.3	0.7	0.4	-	0.4	0.3	3.7	5.5	7.8
DOA	5.3	1.7	1.2	0.8	-	-	194.8	7.4	0.1	-	0.2	< 0.1	2.2	3.5	2.2
DEHP	16.6	4.3	10.0	5.7	3.2	0.3	6.2	4.5	0.4	0.7	1.2	0.3	5.2	10.0	10.5

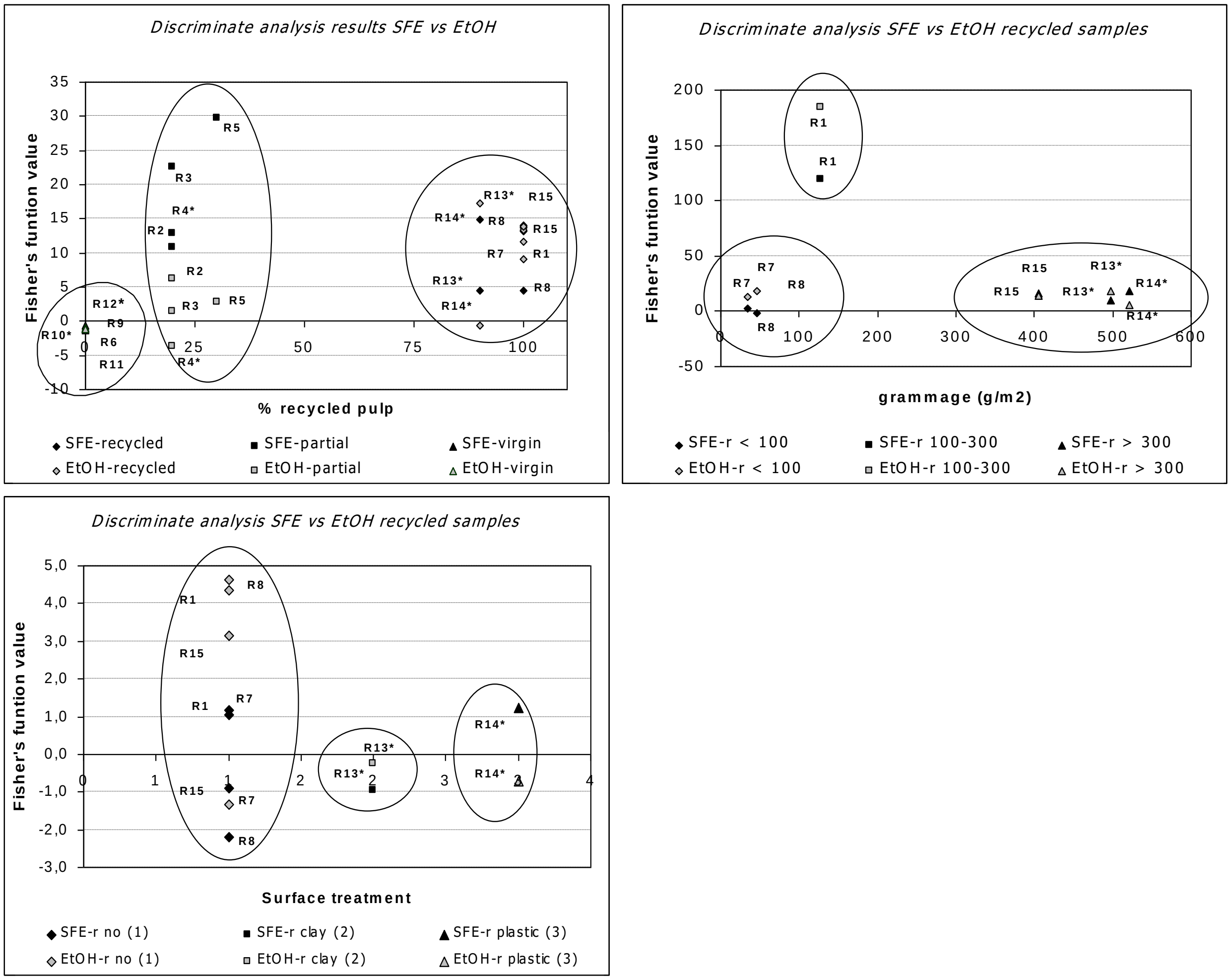
DEP = Diethyl phthalate; DiPN's = Diisopropylnaphthalenes; DiBP = Diisobutyl phthalate; DBP = Di-n-butyl phthalate; DOA = Dioctyl adipate and DEHP = Diethyl hexyl phthalate.

Table 5. Quantitative results obtained by EtOH-GC-MS analysis. Values expressed as µg/g paper.

Compounds	*R1*	*R2*	*R3*	*R4*	*R5*	*R6*	*R7*	*R8*	*R9*	*R10*	*R11*	*R12*	*R13*	*R14*	*R15*
DEP	3.0	0.6	5.2	0.4	1.4	1.3	3.0	12.5	-	-	-	-	0.9	0.9	1.4
DiPN's	22.3	14.1	4.4	-	2.8	-	-	3.4	-	-	-	-	11.9	9.2	94.9
DiBP	37.5	48.6	16.7	0.6	26.1	1.2	6.0	8.2	0.6	0.3	0.3	0.4	15.7	11.1	75.0
DBP	19.6	15.5	10.1	0.6	6.1	1.5	4.6	4.5	0.5	-	0.4	0.4	5.7	4.2	24.3
DOA	3.6	2.3	0.9	-	1.3	3.7	132.8	69.7	-	-	-	-	2.6	1.6	7.1
DEHP	32.0	20.1	11.8	0.6	5.8	3.2	4.0	3.4	0.5	0.7	1.6	0.4	21.9	11.5	61.1

DEP = Diethyl phthalate; DiPN's = Diisopropylnaphthalenes; DiBP = Diisobutyl phthalate; DBP = Di-n-butyl phthalate; DOA = Dioctyl adipate and DEHP = Diethyl hexyl phthalate.

Figure 3 a) b) and c). Discriminate analysis using all the analytical data from SFE-GC/MS and EtOH-GC/MS taking into account the % recycled pulp, the "grammage" and the thickness.



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