Pira International Research report

Analytical Group

Investigation into the effect of additives on migration of substances originating from colourants used in food contact plastics

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Final Report

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Contents

Glossary

APCI	Atmospheric pressure chemical ionisation
BCMA	British Colour Makers Association
BEA	Bis ethoxylated amine
C81	Chimassorb 81
CAS	Chemical Abstract Service
CEN	European Committee for Standardization
CI	Colour Index
DAD	Diode array detector
DCM	Dichloromethane
ETAD	Ecological & Toxicological Association of Dyes and Organic Pigment
EtOH	Ethanol
EU	European Union
FSA	Food Standards Agency
GCMS	Gas chromatography – mass spectrometry
GMS	Glycerol monosterate
HDPE	High density polyethylene
HPLC	High performance liquid chromatography
I1076	Irganox 1076
IPA	Isopropanol
LCMS	Liquid chromatography – mass spectrometry
LDPE	Low density polyethylene
LOD	Limit of detection
MeOH	Methanol
MPPO	Modified polyphenylene oxide
PET	Polyethyleneglycol terephthlate
PG	Pigment green
PP	Polypropylene
PR	Pigment red
PY	Pigment yellow
RT	Retention time
SB	Solvent blue
SIM	Selected ion monitoring
SR	Solvent red
SY	Solvent yellow
TI	Total immersion
TIC	Total ion chromatograms
TMP	Trimethyol propane
UOB	Uvitex OB
UV	Ultra violet

1 Summary

The aim of this project was to systematically investigate the effect of a number of common additives, particularly slip agents and anti-stats which are designed to 'bloom' to the surface of plastics, on the migration of colourants and other substances within the colourants.

Industry support was obtained from two masterbatch manufacturers, a chemical additive manufacturer, Members of the British Colour Makers Association (BCMA) and the Ecological & Toxicological Association of Dyes and Organic Pigment (ETAD). In total 24 samples of colourant were obtained from different sources in Europe, India and China. The European colourants were supplied through ETAD, the colourants from India and China were supplied by the masterbatch manufacturers and the additives were supplied by the additive manufacturer.

The colourants were screened for potential migrants using solvent extraction with dichloromethane, acetone and methanol to cover a wide range of polarities. The solvent extracts were analysed using GC/MS and LC/MS procedures.

Following this screening, 7 different colourant samples were selected for further investigation using migration testing. These comprised 2 different Pigment Red 254 colourants both from China, a Pigment Blue 15:1 from India, a Pigment Blue 15:1 from China, a Solvent Yellow 114 from China, a Solvent Blue 104 from Europe and a Pigment Green 7 from China. Selection was based upon the number of migratable substances detected in the colourant, the colourant type, the plastic in which the colourant was used and the geographical sourcing of the colourant. The plastics used in the testing were high density polyethylene, low density polyethylene, polypropylene and polyethylene terephthalate.

Plastics test samples were prepared using the selected colourants incorporating additives commonly used in plastics formulations. The additives were selected on the basis that they may influence migration and were slip agents, antistats and colourant carriers (PET only). The plastics samples were tested for migration using appropriate food simulants and test conditions. The test conditions were selected to represent the most severe conditions that the plastic would encounter in practice. The values were compared against control samples which were plastic test pieces with colourant added at the same level but containing no additive.

In total, 15 different plastic/additive/colourant blends were prepared. For each plastic there were 2 blends each with a different additive. A third blend of plastic and colourant with no additive was also prepared to act as a control sample. In all cases the colourants and additives were incorporated into the plastic at their highest level to give the worst case values.

In some cases, to give as much information as possible, more than 1 colourant was blended into a sample. It is common practice in industry to blend colourants to obtain a desired effect.

Plaques suitable for testing were prepared by injection moulding and these were subjected to migration testing. Specific migration tests were conducted using the most appropriate food simulants selected from Directive 97/48/EC. The most severe test conditions appropriate for each polymer were selected from Directive 97/48/EC. The samples were aged for at least 3 months to establish if time has an effect on the blooming of additives and consequently the migration of substances.

In general migration was found to be low with no migration of the substances from the colourant detected from LDPE. Migration using 95% EtOH was found to be more severe than iC8 for PET but was less severe than iC8 for the polyolefins. This is a recognised effect for 95% EtOH. PET is a polar polymer and is more readily attacked by the polar ethanol solution causing swelling. Isooctane being non-polar does not tend to penetrate the polymer and lead to migration. No migration was detected into the aqueous simulants 10% EtOH and 3% acetic acid.

The presence of additives in PET did not change migration to any significant effect. However, the additives used in this polymer were not intended to bloom to the surface. Long term storage did increase migration of SY114 by a factor of at least 4 fold in all samples including the control sample.

For HDPE, in general, migration was not affected by the additive, however, for 1 compound which is still not identified migration increased by a factor of up 3 over the control sample when additive was present. It was not possible to identify this compound but the mass spectra indicate it is a chlorinated compound. This may in turn mean it is polar and could therefore partition to a greater extent into the additives. As the additives bloom to the surface with time this compound could be concentrated near the surface of the plastic giving rise to increased migration.

For PP which was blended with S005981/18 a PB 15:1, 3 of the 10 components found in the colourant were found to migrate. The substances that did migrate were in all cases long chain fatty acid esters. The presence of both additives increased migration both when the samples were tested on receipt and when tested after storage. The additives are long chain compounds with a polar end group that encourages them to bloom from the plastic. The long chain fatty acid ester may be soluble in the additive chain which as it blooms to the surface will carry the substance from the colourant with it and promote migration. This effect appears to occur quite rapidly with long term storage having little effect on migration. Some minor variations were noted after long term storage but these differences could be attributed to experimental error.

2 Background

EU harmonised legislation covering colourants used in food contact materials is expected in the future and consideration could be given to the Council of Europe Resolution in which colourants are controlled by their purity and the degree that colourant is transferred in a colour 'bleed' test. However, there is only limited published work on the migration of substances from colourants, except for primary aromatic amines.

FSA Project A03045 reported on the migration of colourants and colourant impurities into food simulants. A03045 was a first step to identify potential migrants and evaluate the propensity for migration of identified substances into food. Plastic test samples were prepared specially for this work and, in general, low levels of migration were found. One variable that was not investigated was the effect of other additives that are usually present in plastics on migration, in particular some additives such as slip agents and antistats that 'bloom' to the surface of plastic materials and articles to prevent them sticking together. This was a deliberate decision to avoid over complicating the analyses and to permit colourant related migrants to be more readily identified. However, additives such as slip agents and antistats have a low solubility in the polymer and fulfil their function by blooming to the surface in a way that cannot be predicted by Fickian diffusion theory. In the process of 'blooming' to the surface, it is possible that they may also exaggerate the migration of other substances present, including those originating from colourants. There is no published data describing the synergistic effect of slip additives and antistats on the migration of other substances present in plastics. It is important to establish if colourant migration from plastics is affected by other additives, as this has direct relevance to consumer safety. The results could also be useful in a wider context to migrants

not originating from colourants to establish if refinements are needed in migration modeling, which is now an accepted tool for compliance testing.

3 Objectives

- In collaboration with industry, the most commonly used colourants with food contact applications will be selected. These colourants will be obtained from a number of different sources

- Using solvent extraction followed by GCMS and LCMS the colourants will be evaluated for the presence of impurities and potential migrants.

- Colourants will be selected for further investigations. In collaboration with industry suitable colourant/additive/plastic combinations will be selected for migration testing and the appropriate test conditions agreed.

- Test samples for migration evaluation will be obtained with the help of industry. They will be prepared using processes that are consistent with materials found in the market place. Test samples containing colourants without other additives will also be prepared to act as control samples.

- Specific migration data will be obtained on the prepared test samples into food simulants both on receipt and after storage for approximately 3 months.

- Migration data will be compared to evaluate the effect of the additives and storage on specific migration.

- Report on the effect of additives studied on the migration of colourants from plastics.

4 Selected Colourants

Following discussions with masterbatch producers, additive manufacturers and industry trade associations a number of colourants were identified for use in the project. The selection criteria were :-

- The colourants must be used in the UK market

- They should represent the most commonly used food contact colourants

- The colourants should be used in the more commonly used plastics

At the outset of the project a meeting was held with industry representatives from BCMA, masterbatch producer, plastic additive manufacturers and representatives from the FSA. With the help of industry the colourants were to be obtained from manufacturers in Europe, India and China and those used in the project have been given below:

Table 1 List of Colourants

Sample No.	Colourants Type	Origin	Description
S005981/1	PR 254	China	Diketopyrrolopyrrole
S005981/2	PR 254	China	Diketopyrrolopyrrole
S005981/3	SR 135	China	Perinone
S005981/4	PG 7	India	Phthalocyanine
S005981/5	PB15:1	India	Phthalocyanine
S005981/6	SY 114	China	Quinophthalone
S005981/7	SB 104	China	Anthraquinone
S005981/8	SB 104	China	Anthraquinone
S005981/9	PR 144	Europe	Hydroxyquinophthalone
S005981/10	PR 254	Europe	Diketopyrrolopyrrole
S005981/11	SB 104	Europe	Anthraquinone
S005981/12	PG 7	Europe	Phthalocyanine
S005981/13	PY 183	Europe	Azo calcium lake
S005981/14	PB15:1	Europe	Phthalocyanine
S005981/15	PG 7	India	Phthalocyanine
S005981/16	PB 15:3	India	Phthalocyanine
S005981/17	PG 7	China	Phthalocyanine
S005981/18	PB15:1	China	Phthalocyanine
S005981/19	PG 7	China	Phthalocyanine
S005981/20	SR 135	China	Perinone
S005981/21	SR 135	Europe	Perinone
S005981/22	SY 114	Europe	Quinophthalone
S005981/23	SR 135	Europe	Perinone
S005981/24	SY 114	Europe	Quinophthalone

Following screening 7 colourants were selected for further investigation. To give maximum data 2 colourants were blended together in HDPE and PET. These were samples S005981/1 and 2, 2 Pigment Red 254 samples used in HDPE, and samples S005981/6 and 11, the 2 colourants used in PET. The blending of these

colourants in the same plastic sample was suggested by industry who informed us, at a meeting to discuss the project, that this was standard industry practice and was often carried out to obtain a desired effect.

Selection of additives

Additives that are commonly blended into plastic that are widely used for food contact application were selected for use in the study. Additives that are designed to bloom to the surface of the plastic and possibly enhance migration were sought as these would potentially give the biggest effect.

The additives selected for this study were erucamide, a common slip agent used in polyolefins and an antistatic agent of which was a 2:1 blend of Glycerol monostearate (GMS) and Bis ethoxylated amine (BEA).

To cover PET, another plastic widely used in food contact application, 2 colour carriers were selected. Although these will not bloom to the surface they could influence migration. Carrier 1 was non-polar and the other, carrier 2, was polar. Due to confidentiality restraints it was not possible to disclose the identity of the carriers, it was recognised that this placed an unavoidable limitation on the data produced in the study.

5 Experimental work

5.1 Identification of potential migrants

In the initial investigations the colourants were screened for substances that could potentially migrate when the colourant was blended in a plastic together with an additive.

Samples of colourant listed in Table 1 were extracted, sequentially, with 3 different solvents. The solvents were selected to cover a wide range of polarities so that compounds with differing solubility characteristics and polarities would be extracted. The extraction protocol was not intended to be a quantitative exercise but was used to identify potential migrating species.

5.1.1 Solvent extraction procedures

The following procedure was used to extract the colourants. The solvents used, in order, were

1. Dichloromethane (DCM)

2. Acetone
3. Methanol (MeOH)

Procedure

1. Weigh 1g of colourant into a 40 ml screw top septum sealed vial

2. Add 20 ml of DCM

3. Shake and put in ultrasonic bath for 15 minutes

4. Remove vials from bath and centrifuge for 30 minutes at 3000 rpm

5. Decant supernatant liquid into clean 40 ml vial.

6. Evaporate solvent using either a steam bath or by gently heating the solution under a stream of nitrogen being careful to remove vials immediately all solvent has been evaporated.

7 Dissolve residue in 5 mls of isopropanol (IPA), using an ultrasonic bath if necessary, and analyse using GCMS and LC/MS

8. Extract the remaining colourant which had been recovered after removal of DCM (see step 5) with acetone by repeating steps 2 to 7.

9. Extract the remaining colourant which had been recovered after removal of DCM and acetone (see step 5) with MeOH by repeating steps 2 to 7.

During the extraction procedure it was found that all three samples of solvent blue 104, was totally soluble in DCM. As a consequence it was dissolved in DCM and 0.5 ml of the solution was diluted to 25 mls with IPA to give a solution of approximately 1mg/ml. Acetone and MeOH were not used for this sample.

5.1.2 Analysis of extract

The extracts obtained above were analysed by GCMS and LCMS using the conditions given below

Conditions

GCMS

Column:	5% Phenylmethyl siloxane 30m × 0.25mm 0.25μm film thickness
Injection volume	1μl Splitless
Injector Temperature	280°C
Detector Temperature	280°C
Temperature Programme	40°C hold for 5 mins 10°C/min to 320°C hold for 30 minutes
Detector	MS Scanning from 28 to 1000 mass units
Carrier gas	Helium at 7.7 psi

LC/DAD/MS

Column:	25cm × 4.0 mm Glass lined Wakosil C_{18} 5 μm
Detectors:	Diode array detector(DAD), data acquired for 190 - 800 nm, chromatogram present for 220 nm. MS mulitimode (both ES and APCI) scanning over mass range of 50 to 1350 allowing both positive and negative ions to be detected
Injection volume:	20μl

Mobile phase:

Time Mins	Water %	Acetonitrile %	IPA %
0	90	9	1
2	90	9	1
20	0	90	10
50	0	90	10
51	90	9	1
56	End analysis		

5.2 Preparation of Samples for migration evaluation

Based on the findings from the solvent extraction tests described above, 7 colourants were selected for blending into polymers to make migration test samples. The selection criteria for the colourants were as follows

Potential migrants were identified in the colourant

Colourants were selected from Chinese, Indian and European sources

Colourants were selected so that all major food contact plastics would be tested in the study.

The colourants selected are listed below:

Table 2 List of Colourants Selected for Blending

Project Sample No.	Colourants Type	Origin	Polymer for blending
S005981/1	Pigment Red 254	China	HDPE
S005981/2	Pigment Red 254	China	HDPE
S005981/5	Pigment Blue15:1	India	LDPE
S005981/6	Solvent Yellow 114	China	PET
S005981/11	Solvent Blue 104	Europe	PET
S005981/17	Pigment Green 7	China	PP
S005981/18	Pigment Blue15:1	China	PP

The colourants tested were typically used in specific polymers at known concentrations. Colourants selected based on the above criteria were compounded into suitable plastics at the highest appropriate concentration to give the worst case data. The additives selected for the study were used at levels representing the highest typical concentrations used in practice. Approximately 200 test plaques of each of the compounded plastics were prepared by injection moulding. These plaques, or test samples, were designed to be suitable for migration testing by total immersion procedures without cutting and therefore not exposing a cut edge. The full list of samples prepared is given below.

1 PET + 0.03% S005981/11 and 0.015% S005981/6
2 PET + 0.03% S005981/11 and 0.015% S005981/6 + 400 ppm carrier 1
3 PET + 0.03% S005981/11 and 0.015% S005981/6 + 150 ppm carrier 2
4. HDPE + 0.3% of [S500981/2(75%) and S005981/1 (25%)]
5. HDPE + 0.3% of [S500981/2(75%) and S005981/1 (25%)] + 3000ppm erucamide
6. HDPE + 0.3% of [S00005981/2(15%) and S00005981/1 (25%)] + 5000ppm blend A
7. LDPE + 0.3% S005981/5
8. LDPE + 0.3% S005981/5 + 1000 ppm erucamide
9. LDPE + 0.3% S005981/5 + 2000ppm blend A
10 PP + 0.3% S005981/17
11 PP + 0.3% S005981/17 + 5000ppm erucamide
12 PP + 0.3% S005981/17 + 5000ppm blend A

13 PP + 0.3% S005981/18
14 PP + 0.3% S005981/18 + 5000ppm erucamide
15 PP + 0.3% S005981/18 + 5000ppm blend A

The samples with no additive present were to act as controls so that the effect of the additive could be assessed.

The samples were prepared by 2 masterbatch manufacturers using their in-house moulds. One set of plaques were 39mm × 58mm × 2mm and the other was 50mm × 75mm × 2mm.

5.3 Migration Testing

Migration from the polymers was carried out into simulant B (3% Acetic Acid), Simulant C (10% ethanol) and using substitute tests for Simulant D (olive oil). Simulant A (distilled water) was not used in this study as the other aqueous simulants are generally recognized to give higher migration.

Substitute tests for simulant D are given in Commission Directive 97/48/EC (EC 97) and may be used if, for technical reasons, it is not feasible to use olive oil. The relevant substitute tests are listed in table 3.

Table 3: Conventional Conditions for substitute tests

Test Conditions with simulant D	Test Conditions with isooctane	Test Conditions with ethanol 95%	Test Conditions with MPPO*
10 d at 40°C	2 d at 20°C	10 d at 40°C	-
2 h at 70°C	0.5 h at 40°C	2.0 h at 60°C	-
2 h at 100°C	1.5 h at 60°C	3.5 h at 60°C	2 h at 100°C
2 h at 121°C	2.5 h at 60°C	4.5 h at 60°C	2 h at 121°C

* = Modified polyphenylene oxide (Tenax)

The Directive states that the highest migration values from the above be reported. The test using Modified polyphenylene oxide (MPPO) was not used in this study as past experience indicates this will only pick up material transferred by vapour phase transmission and always gives lower values than isooctane or 95% ethanol (EtOH).

It was considered not feasible to use olive oil in this study for a number of reasons. These include:-

- A complex range of potential migrants were being investigated and olive oil is a complex matrix in which to carry out an analysis. Each migrant would require a specific method to be developed. This was impractical within the scope of this project.

- It was the aim of the study to achieve a limit of detection of 50 µg/kg or better. To aid sensitivity of the experiment it is standard protocol to reduce the volume of iC8 and 95% EtOH by evaporation and prepare the migration residue using a smaller volume of solvent. Clearly it was not possible to reduce the volume of olive oil by this procedure.

- The primary aim of the project was to evaluate the effect of the additive on migration and this would be more readily achieved using the substitute simulants.

An accepted procedure to improve analytical sensitivity is to increase the ratio of surface area of plastic to volume of simulant used. In Directive 2002/72/EC (as amended) a standard ratio of 6dm^2 for every litre (kilogram) of simulant is assumed when assigning migration limits. In the tests for this study a ratio of up to 12dm^2 per litre of simulant was employed in order to concentrate potential migrants and thus improve the limit of detection of the methods.

Each migration test was carried out in triplicate. As the plaques were 2 mm thick both sides of the sample could be used in the migration calculation. Testing was carried out by total immersion without cutting the samples.

Worst case test conditions were selected to represent the most severe conditions likely to be encountered by the polymers. These are listed in table 4.

Table 4 Migration conditions

Plastic sample no.	Polymer	iC8	95% EtOH	10% EtOH	3% HAC
1 to 3	PET	0.5h/40°C + 2d/20°C	2.0h/60°C+10d/40°C	2h/70°C + 10d/40°C	2h/70°C + 10d/40°C
4 to 6	HDPE	1.5h/60°C + 2d/20°C	3.5h/60°C+10d/40°C	2h/100°C + 10d/40°C	2h/100°C + 10d/40°C
7 to 9	LDPE	0.5h/40°C + 2d/20°C	2.0h/60°C+10d/40°C	2h/70°C + 10d/40°C	2h/70°C + 10d/40°C
10 to 15	PP	2.5h/60°C + 2d/20°C	4.5h/60°C+10d/40°C	2h/121°C + 10d/40°C	2h/121°C + 10d/40°C
HDPE Blank	HDPE	1.5h/60°C + 2d/20°C	3.5h/60°C+10d/40°C	2h/100°C + 10d/40°C	2h/100°C + 10d/40°C
LDPE Blank	LDPE	0.5h/40°C + 2d/20°C	2.0h/60°C+10d/40°C	2h/70°C + 10d/40°C	2h/70°C + 10d/40°C
PP Blank	PP	2.5h/60°C + 2d/20°C	4.5h/60°C+10d/40°C	2h/121°C + 10d/40°C	2h/121°C + 10d/40°C
PET Blank	PET	0.5h/40°C + 2d/20°C	2.0h/60°C+10d/40°C	2h/70°C + 10d/40°C	2h/70°C + 10d/40°C

Migration testing was conducted in incubators where the temperature was continually logged, or in an autoclave where the temperature was monitored using a calibrated thermocouple. For the short term, high temperature tests, the temperature of a sample of blank simulant was monitored and timing began when the simulant reached the test temperature.

5.3.1 Analysis of Simulants

The simulant was gently evaporated to dryness using either a water bath or hotplate and redissolved in 5 mls of propan-2-ol (IPA) which is a suitable solvent for injection for GC analysis. GCMS was the preferred method of analysis for the simulants. It was not possible to identify the extracted substances by LCMS and the GCMS was more sensitive. It was decided to concentrate on this technique for the analysis of the food simulants.

At the initial stage, no information was available with respect to which of the potential migrants identified from the colourant extract would migrate from the plastic into the food simulants. As there were a lot of substances that could potentially migrate it was necessary to analyse the simulant concentrates using techniques that would detect as many as possible of the compounds that may be present. In particular this required that the GCMS was operated in the SCAN mode, spectra were acquired by scanning from 10 to 1000 mass units.

GCMS

Column:	5% Phenylmethyl siloxane 30m × 0.25mm 0.25μm film thickness
Injection volume	1μl Splitless
Injector Temperature	280°C
Detector Temperature	280°C
Temperature Programme	40°C hold for 5 mins 10°C/min to 320°C hold for 30 minutes
Detector	MS Scanning from 28 to 1000 mass units
Carrier gas	Helium at 7.7 psi

From the data obtained by GCMS analysis of the solvent extracts of the colourants, major ions for the possible migrants were obtained. Using these ions reconstructed ion chromatograms were obtained from the GCMS scanned chromatograms of the migration simulants to identify migrating substances. The samples were then reanalysed using Selected Ion Monitoring (SIM) mode to improve sensitivity and reduce background interference. The ions selected for

the SIM depended on the colourant and the major ions found in the substances detected in the colourant extract.

6. Validation

To evaluate the chromatographic performance of the systems a blend of plastic additives were used. These were Chimasorb 81 (2-hydroxy-4-(octyloxy)benzophenone), Irganox 1076 (octadecyl 3-(3,5-di-tert-butyl-4-hydroxyphenyl)propionate). Uvitex OB (2,5-bis(5-tert-butyl-benzoxazol-2-yl)thiophene and Irgaphos 168 (tris(2,4-di-tert-butylphenyl)phosphite). Experience has shown that in particular Chimasorb 81, a benzophenone, is extremely sensitive to active sites in the GC and will tail very readily if contamination is present. Whenever this happened the GCMS liner was changed and, if required, the front of the column removed. Analysis of the samples was only started when the chromatography of the standards was satisfactory.

6.1 Estimation of limits of detection in migration simulant samples

For GCMS in SCAN mode

When analysing samples using the GCMS in scan mode it was necessary to evaluate the performance of the instrument to ensure there was sufficient sensitivity to detect all potential compounds that may be present. However, as the potential migrants covered a range of polarities and molecular weights it was decided to verify the performance of the instrument using solutions of a representative set of standard additives prepared in IPA, the same solvent used to dissolve the samples. Once again Chimassorb 81 (C81) Irganox 1076 (I1076), Irgaphos 168 (I168) and Uvitex OB (UOB) were selected because they are common additives often found in plastics. Irganox 1010 (I1010) was also included in the blend but was found not to elute from the GCMS.

Using the total ion chromatograms (TIC) for the analysis of the standards, limits of detection for the GCMS were calculated for test compounds. These were calculated using the ion given in Table 5 abstracted from the TIC and based on a 3:1 signal to noise ratio. These values take into account the sample concentration step and the standard EU ratio of plastic surface area to volume of simulant of 6dm^2/ l(kg). The values are given in table 5.

Table 5: Estimated Limits of Detection for GCMS

Standard	Typical Retention Time (min)	Ion m/z	Limit of detection (μg/kg)
C81	29.1	213	32
I 168	33.2	441	4
I1076	34.5	530	10
UOB	37.1	430	8

For LCMS

Analysing samples using the LCMS the performance of the instrument was evaluated using the same mixture of compounds that were used to test the GCMS performance. It is possible to run the LCMS in a multi media mode in which both electro spray (ES) and atmospheric pressure chemical ionization (APCI) process are monitored simultaneously. it is also possible to scan the spectra in both the negative ion and positive ion mode. This will reduce sensitivity of the detector but as the compounds being extracted from the colourant are unknown it was decided to adopt this procedure so that as far as possible all compounds will be detected

In addition to the mass spectrometer the LC eluent was also monitored using a UV diode array detector (DAD). Limits of detection for the LCMS were calculated for test compounds based on a 3:1 signal to noise ratio for all detection modes. These values take into account the sample concentration step and the standard EU ratio of plastic surface area to volume of simulant of 6dm^2/ l(kg). The values are given in table 6.

Table 6: Estimated Limits of Detection for LC

Standard	Typical Retention Time (min)	Limit of detection (μg/kg)		
		DAD 220 nm	ES+APCI negative mode	ES+APCI positive mode
C81	24.1	1	N/D	130
UOB	27.2	2	N/D	20
I1010	27.9	3	250	N/D
I1076	45.9	11	450	N/D
I 168	47.1	8	N/D	90

N/D = Not detected indicating that some compounds will ionise in the positive mode and some using negative scan. Using the multi-mode procedure all standards compounds have been detected.

From the above it can be seen that the LC/UV is the most sensitive detector, however, it does require that the eluting species have UV chromophore before they can be detected

6.2 Calibration of GCMS

For some migrants detected a range of standards were prepared covering the concentration of the sample. It was not possible to do this in all cases as the identities of the substance(s) could not be determined. Additionally, it was not essential to identify and quantify all migrants since the effect of the additives on migration could be determined by comparing peak areas for samples with and without additives.

For substances where calibration was carried out, a limit of detection (LOD) of less than 50 ppb and correlation coefficients better than 0.996 were sought. Example of calibration curves for all analyses are given in appendix 1. In these graphs the concentration is expressed in terms of μg/ml of substance in the concentrated migration solution. To determine migration the values are calculated back to the standard EU ratio of area of plastic sample to volume of simulant which is accepted to be 6dm^2 to 1 litre of simulant or 1 kg of simulant assuming a density of 1. Migration is therefore reported in terms of μg/kg. The LOD for SY114 was found to be 20 μg/kg and that for SB 104 was found to be 10 to 30 μg/kg.

7. Results and Discussion

7.1 Solvent Extraction

The purpose of the solvent extraction of the colourant was not to quantify potential migrants but to identify and characterise as many compounds as possible present in the colourants. The concentrated extract was analysed by GCMS in order to identify peaks where possible using the Wiley7n library. All of the solutions were also analysed using LCMS. Although it was not possible to identify unknown compounds using this technique by comparing to library matches it may show evidence of compounds too polar to be analysed by GCMS

and highlight differences between the colourants and the effect of the presence or absence of additives

The observations from the findings of the solvent extraction exercise are discussed below.

The only colourant found to be soluble in any of the solvents was sample Solvent Blue 104 (samples S009581/7, 8 and 11) which is intended for use in PET only. This was readily soluble in DCM and was not extracted using ether acetone or methanol.

For all of the colourants evaluated no additional peaks were detected in the acetone and MeOH extracts by ether technique, as a result the findings from the DCM extracts only are presented. GCMS and LCMS traces of the DCM extract and spectra of peaks, together with their identification where available are given in appendix 2.

Tables 7 to 13 summarises the findings from the solvent extraction screening procedure. These findings are from the extraction of the colourants only and were not migration data, therefore, there will not be effects from any additives present. Chromatograms and spectra for all the colorants are given in appendix 2

Table 7 Summary of Findings for PR 254

Sample No.	Colourants Type	DCM Extract
S005981/1	PR 254 M. Wt 357 China	GCMS: 5 peaks, 3 unidentified 1,3,5-Trazine, 2,4-bis(4-chlorophenyl)-6-phenyl- 1,3,5-Trazine, 2,4,6-tris(4-chlorophenyl) LC/DAD: 3 peaks at 23.9, 26.1 & 27.7 no chromophore in the visible spectrum LCMS –ve: 3 peaks detected LCMS +ve: 2 peaks detected
S005981/2	PR 254 M. Wt 357 China	GCMS: 3 peaks, 2 unidentified 3-Chlorobenzamide LC/DAD: 2 peaks at 10.9 & 23.9 no chromophore in the visible spectrum LCMS –ve: No peaks detected LCMS +ve: 1 peak
S005981/10	PR 254 M. Wt 357 Europe	GCMS: 3 peaks, 1 unidentified 4-chlorobenzonitrile 3-chlorobenzamide LC/DAD: 3 peaks, 1 at 20.2 with chromophore in the visible spectrum LCMS –ve: LCMS +ve: 3 peaks detected

Table 8 Summary of Findings for SR 135

Sample No.	Colourants Type	DCM Extract
S005981/3	SR 135 M. Wt 408 China	GCMS: 2 peaks, 1 unidentified Dichlorobenzene Peak 2 37.30 highest ion at 408 LC/DAD: 7 peaks detected. Peaks at 18.9, 23.6, and 24.6 with chromophore in the visible spectrum LCMS –ve: 1 peak detected Peak 1 20.3, highest ion 381 LCMS +ve: 2 minor peaks detected
S005981/20	SR 135 M. Wt 408 Other	GCMS: 4 peaks, 2 unidentified Oleamide SR135? LC/DAD: 7 peaks 2 with chromophore in the visible spectrum LCMS –ve: 2 Peaks detected LCMS +ve: no significant peaks detected
S005981/21	SR 135 M. Wt 408 Previous study	GCMS: 4 Peaks detected 3 unidentified Dichlorobenzene LC/DAD: I peak 20.1 with strong chromophore in the visible spectrum LCMS –ve: 2 minor peaks detected LCMS +ve: 1 Peak detected
S005981/23	SR 135 M. Wt 408 Europe	GCMS: No peaks detected LC/DAD: 7 peaks 24.3 with chromophore in the visible spectrum LCMS –ve: 1 peak detected LCMS +ve: No peaks detected

Table 9 Summary of Findings for PG 7

Sample No.	Colourants Type	DCM Extract
S005981/4	PG 7 M. Wt 1030-1130 India	GCMS: 4 peaks, Dodecanoic acid Tetradecanoic acid Hexadecanoic acid Oleic acid LC/DAD: 6 Major peaks with no chromophore in the visible spectrum LCMS –ve: 2 peaks detected LCMS +ve: 1 peak detected
S005981/12	PG 7 M. Wt 1030-1130 Europe	GCMS: I peak 3,4,5,6-Tetrachlorophthalimide LC/DAD: 1 peak with no chromophore in the visible spectrum LCMS –ve: I peak detected LCMS +ve: no peaks detected
S005981/15	PG 7 M. Wt 1030-1130 India	GCMS: Complex chromatogram with 7 major peaks 5 unidentified DIBP Oleic acid LC/DAD: 3 peaks with no chromophore in the visible spectrum LCMS –ve: 1 peak detected LCMS +ve: 1 peak detected
S005981/17	PG 7 M. Wt 1030-1130 China	GCMS: Complex chromatogram with 9 major peaks 3 unidentified 1,3-Dichlorobenzene Isolongifolene Simonelite Resin acid? 13-isopropylpodocarp acid Resin acid? LC/DAD: No peaks with chromophore in the visible spectrum LCMS –ve: 3 peaks detected LCMS +ve: 1 peak detected
S005981/19	PG 7 M. Wt 1030-1130 China	GCMS: Chromatogram with 9 peaks 5 unidentified Decahydro-4,8,80trimethyl-9-methylene-1,4-methaneazulene Hexchlorobenzene Phenatherenecarbaldehyde Dehydroabietic acid LC/DAD: 3 peaks with no chromophore in the visible spectrum LCMS –ve: 2 peaks detected LCMS +ve: 1 peak detected

Table 10 Summary of Findings for PB15:1 (15:3)

Sample No.	Colourants Type	DCM Extract
S005981/5	PB15:1 M. Wt 576 India	GCMS: Complex chromatogram with at least 9 major peaks 1 undentified 1-dodecene 1-tetradecene 1-chlorododecane Hexadecane 1-chlorotetradecane Octadecane 1-chlorohexadecane Phthalimidomethylphthalimide LC/DAD: Complex chromatogram of at least 5 major peaks none with chromophore in the visible spectrum LCMS –ve: no peaks detected LCMS +ve: 2 peaks detected
S005981/14	PB15:1 M. Wt 576 Europe	GCMS: 3 minor peaks 1 unidentified Phthalic anhydride Isoindole LC/DAD: 5 peaks none with chromophore in the visible spectrum LCMS –ve: 1 peak detected LCMS +ve: No peaks detected
S005981/16	PB 15:3 M. Wt 576 India	GCMS: Complex chromatogram with at least 8 major peaks 7 unidentified DIBP LC/DAD: Very confused and busy chromatogram no chromophore in the visible spectrum LCMS –ve: Very confused and busy chromatogram LCMS +ve: Very confused and busy chromatogram 1 peak detected
S005981/18	PB15:1 M. Wt 576 China	GCMS: Complex chromatogram with at least 10 major peaks 1 unidentified 1-dodecene Cyclodecane Palmitic acid Stearic acid Dodecanol? Phthalimidomethylphthalimide Hexadecanoic acid, dodecyl ester Lauryl stearate Myristyl stearate LC/DAD: Complex chromatogram with 1 Major peak with no chromophore in the visible spectrum LCMS –ve: 1 peak detected LCMS +ve: 1 peak detected

Table 11 Summary of Findings for SY 114

Sample No.	Colourants Type	DCM Extract
S005981/6	SY 114 M. Wt 289 China	GCMS: 3 peaks 1 unidentified Isoindole Probably SY114 LC/DAD: 1 Major peak Peak 1 20.2, Strong chromophore in the visible spectrum (SY114?) LCMS –ve: 1 peak detected (probably SY 114) LCMS +ve: 1 peak detected (probably SY114)
S005981/22	SY 114 M. Wt 289 Previous Study	GCMS: 4 peaks, 3 unidentified Probably SY 114 LC/DAD: 1 major peak at 20.2 with chromophore in the visible spectrum LCMS –ve: 1 peak detected (probably SY 114) LCMS +ve: 1 peak detected (probably SY 114)1
S005981/24	SY 114 M. Wt 289 Europe	GCMS: 1 peak Probably SY114 LC/DAD: 1 peak at 19.8 with chromophore in the visible spectrum LCMS –ve: 1 peak detected (probably SY 114) LCMS +ve: 1 peak detected (probably SY 114)

Table 12 Summary of Findings for SB 104

Sample No.	Colourants Type	DCM Extract
S005981/7	SB 104 M. Wt 474 China	GCMS: 1 peak SB 104 LC/DAD: 1 peak 28.2 strong chromophore in the visible spectrum LCMS –ve: 1 peak detected (probably SB 104) LCMS +ve: 1 peak detected (probably SB 1041
S005981/8	SB 104 M. Wt 474 China	GCMS: 1 peak SB 104 LC/DAD: 1 peak 28.2 strong chromophore in the visible spectrum LCMS –ve: 1 peak detected (probably SB 104) LCMS +ve: 1 peak detected (probably SB 104
S005981/11	SB 104 M. Wt 474 Europe	GCMS: 1 peak SB 104 LC/DAD: 1 peak 28.2 strong chromophore in the visible spectrum LCMS –ve: 1 peak detected (probably SB 104) LCMS +ve: 1 peak detected (probably SB 104)

Table 13 Summary of Findings for PR 144 and PY 183

Sample No.	Colourants Type	DCM Extract
S005981/9	PR 144 M. Wt 829 Europe	GCMS: No significant peaks LC/DAD: 4 Major peaks all with chromophore in the visible spectrum LCMS –ve: no peaks detected LCMS +ve: 1 peak detected
S005981/13	PY 183 M. Wt 545 Europe	GCMS: I peak unidentified LC/DAD: no peaks detected LCMS –ve: no peaks detected LCMS +ve: I peak detected

It was found that, in general, the samples sourced from Europe contain less extractable material detected using chromatographic methods than those obtained from India or China. It was also found that samples obtained from different sources in India and China differed significantly. Examples of this were Pigment Green 7 and Pigment Blue 15:1.

In Pigment Blue 15:1 a substance was identified by industry as being phthalimidomethylphthalimide. This was found in samples of S005981/5 and 18 and is a common by-product in the manufacture of phthalimido copper phthalocyanine.

In the previous study nearly all the samples were obtained from Europe whilst in this work most of the samples were sourced from India and China. As has been mentioned before, the extractable materials from both these sources can vary significantly even when obtained from the same country, however, there were some similarities between this study and the previous investigation. For example an unidentified peak detected in samples of colourant PR 254 was found in the previous investigation.

Other compounds common to both investigations include tetrachlorophthalimide which was found in samples of PG 7, phthalic anhydride was found in PB15:1 and both solvent SY 114 and SB104 were detected in both studies.

7.2 Migration Testing

Due to time and expenditure constraints it was not possible to test all the colourants initially screened for potential migrants. As a result from the 24 colourants initially screened 7 were selected for further investigation. Selection was discussed with industry representatives and was based upon the plastics in

which the colourants were used. The colourants in which the highest number of impurities were found were also selected. In addition the colourant origin was considered so that all regions were represented in the migration experiments. Because of the higher level of potential migrants found in the samples from India and China sample from these countries comprise the bulk of the samples tested for migration.

No samples were selected from SR 135, PR144 and PY 183 as these had the lowest level of migratable substances.

The colourants selected and the substances that may migrate are tabulated below. The identification and retention times are based on the GCMS analysis only as, as has been discussed previously; it is not possible to identify unknown compounds based solely on the LCMS data.

Table 14: List of colourants tested for migration and substances in the colourants

Sample No.	Colourants Type	Origin	Tentative Peak Identification from GCMS (RT are indicative only and can vary to small extent)*
S005981/1	PR 254	China	Peak 1 RT 28.6 mins Unknown ** Peak 2 RT 28.9 mins Unknown Peak 3 RT 29.8 mins Unknown Peak 4 RT 32.6 mins 2,4-bis(4-chlorophenyl)-6-phenyl-1,3,5 Triazine Peak 5 RT 34.0 mins 2,4,6-tris(4-chlorophenyl)1,3,5 Triazine
S005981/2	PR 254	China	Peak 1 RT 17.4 mins 2-chlorobenzamide Peak 2 RT 28.4 mins Unknown ** Peak 3 RT 34.9 mins Unknown
S005981/5	PB15:1	India	Peak 1 RT 13.9 mins 1-dodecene Peak 2 RT 16.8 mins 1-tetradecene Peak 3 RT 17.82 mins 1-chlorododecane Peak 4 RT 19.3 mins hexadecane Peak 5 RT 20.3 mins 1-chlorotetradecane Peak 6 RT 21.6 mins Octadecane Peak 7 RT 22.4 mins 1-chlorohexadecane Peak 8 RT 30.4 mins phthalimidomethylphthalimide*** Peak 9 RT 31.4 mins Unknown
S005981/6	SY 114	China	Peak 1 RT 17.0 mins Isoindole Peak 2 RT 20.6 mins Unknown Peak 3 RT 32.3 mins Probably SY114
S005981/11	SB 104	Europe	Peak 1 RT 43.6 mins Probably SB104
S005981/17	PG 7	China	Peak 1 RT 11.2 mins 1,3-Dichlorobenzene Peak 2 RT 16.9 mins Isolongifolene Peak 3 RT 23.7 mins Unknown Peak 4 RT 23.97 mins Unknown Peak 5 RT 24.04 mins simonelite Peak 6 RT 25.1 mins Unknown Peak 7 RT 26.6 mins Resin acid? Peak 8 RT 27.5 mins 13-isopropylpodocarp acid Peak 9 RT 27.8 mins Resin acid?
S005981/18	PB15:1	China	Peak 1 RT 13.7 mins 1-dodecene Peak 2 RT 17.5 mins Cyclodecane Peak 3 RT 23.1 mins Palmitic acid Peak 4 RT 24.9 mins stearic acid Peak 5 RT 27.5 mins Unknown Peak 6 RT 29.2 mins dodecanol? Peak 7 RT 30.1 mins phthalimidomethylphthalimide *** Peak 8 RT 30.9 mins Hexadecanoic acid, dodecyl ester Peak 9 RT 32.2 mins lauryl stearate Peak 10 RT 33.4 mins myristyl stearate

* Note: Retention Times (RT) are intended to be indicative only. These can vary to small extent depending on the age and condition of the chromatographic column. Because of this these retention times may differ to small degree from those given in appendix 2

** Same compound *** Same compound

To date it has not been possible to identify the GCMS peaks 1, 2 and 3 for colourant S005981/1. The colourant is 3,6-Bis(4-chlorophenyl)-2,5-dihydro-pyrrolo[3,4-c]pyrrole-1,4-dione, has a molecular weight of 357. The spectra for all 3 compounds are consistent with the presence of chlorine atoms. The spectra for peak 1 is similar to peak 3 save for the fact that peak 3 has an ion at m/z 356, which would be consistent to a molecular ion for the colourant. Both compounds have a base ion of m/z 314. Peak 2 has a base ion of m/z 328 which is also the highest ion in the spectra which suggests that it could be the same compound as peak 1 with the addition of CH_2.

In the LCMS analysis of the DCM extract from this colourant similar compounds are detected in the ES+APCI positive scan mode. These give protonated $[m+H]^+$ ions of m/z 315-319, m/z 329-333 and m/z 357-361 which suggests that the base peaks detected in the GCMS may also be the molecular ions. The compound with the base ion of m/z 315 is also present in the extract for S005981/2 mirroring the findings from the GCMS analysis.

Test samples were prepared by blending the colourants into selected plastics with and without additives, the samples where no additive was blended were to act as control materials. Migration was tested using the most severe conditions to which the plastics would be subjected, see Table 4. The simulants 3% acetic acid, 10% ethanol and the substitute tests for simulant D were used in the study.

Migration was tested immediately upon receipt of the samples from the manufacturers which was within 48 hours of production. To evaluate the effect of additives blooming to the surface and increasing migration of the components associated with the colourants the polyolefin plastic test samples were stored, in the dark at ambient conditions for 3-3.5 months and migration tests were repeated using identical conditions. The additives in the samples of PET were not designed to bloom to the surface of the plastic so the length of storage should make little difference to the level of migration. However, to maximise any storage effect these were stored for 5 months before they were retested.

Using the analytical conditions given above the migration simulants were analysed. From the initial screening exercise it was possible to identify ions in the target compounds. In addition to using the GCMS in SCAN mode the method was modified to increase sensitivity of the analysis. All migration samples were reanalysed with the GCMS SIM mode, based on the ions for the target compounds. The simulants were not analysed using LC/DAD/MS as very little

additional information was obtained on the colourant extracts using this procedure.

To increase the ratio of surface area of plastic to volume of simulant (thereby increasing sensitivity of the final analysis) each migration test was carried out using three plaques and each migration test was carried out in triplicate. As the plaques were 2 mm thick both sides of the sample could be used in the migration calculation. Testing was carried out by total immersion without cutting the samples.

As stated previously, test conditions were selected that would represent the most severe conditions of use likely to be encountered.

The findings from the migration test are discussed below.

Plastic Samples 1 to 3 PET

These were samples of PET containing the colourants S005981/11 (SB104 from Europe) and S005981/6 (SY 114 from China). The 2 colourants were blended at their maximum levels together to get as much information as possible. Plastic sample 1 contained no additive, plastic sample 2 contained non-polar colourant carrier 1 and plastic sample 3 contained the polar colourant carrier 2.

Colourant S005981/11 was soluble in DCM and when analysed by GCMS the chromatogram had single peak with a retention time of 43.6 minutes and an ion at m/z 474 which was attributable to the colourant. LC/DAD/MS was not used for the analysis as no additional peaks were detected.

Colourant S005981/6 was slightly soluble in DCM and the colourant extract gave 3 peaks. Peak 1 eluted with a retention time of 17.0 minutes and was identified as 2-chlorobenzamide, peak 2 eluted with retention of 20.6 minutes with a maximum ion of m/z 219, peak 3 eluting at 32.3 minutes with an ion at m/z 289 and was possibly attributable to the colourant. LC/DAD/MS was not used for the analysis as no additional peaks were detected using this technique. Examples of chromatograms and spectra of both colourants are given in appendix 2

The migration solutions were analysed by GCMS. In some of the migration solutions from S005981/6, 1 compound only was found to migrate. This was peak 3 which was attributable to SY 114. No other compounds were detected.

Migration of SB 104 into 95% EtOH was found from the samples of PET containing S005981/11.

Solutions of the colourants were used as calibration standards. As colourant sample S005981/11 had no other peaks in the chromatogram it was assumed to be >98% pure. The area of the peak attributable to the colourant S005981/6 was >96% of the total peak are in the chromatogram. Using these standards migration of the colourants was determined and the values are given below. No correction was made for the purity of the colourants in the sample. The results are given in tables 15 and 16.

Table 15: Migration of SB 104, S005981/11, from PET (mean of 3 determinations)

	Mean Migration Values μg/kg					
	Plastic 1 + No additive		Plastic 2 + Non-polar carrier		Plastic 3 + Polar carrier	
	On Receipt	After Storage	On Receipt	After Storage	On Receipt	After Storage
iC8	<10	<30	<10	<30	<10	<30
95% EtOH	40	51	45	58	60	49
10% EtOH	<10	<30	<10	<30	<10	<30
3% AA	<10	<30	<10	<30	<10	<30

NOTE: The limit of detection (LOD) for SB 104 changed between the analysis on receipt (10 μg/kg) and after storage (30 μg/kg). It should be noted that the migration measurements were made after a 5 month interval. The reason for the small variation of sensitivity could be due to changes of performance of the detector, column or injection system. However, both LOD's would be acceptable when submitting an EU dossier where the LOD required is usually < 50 μg/kg.

Table 16: Migration of SY 114, S005981/6, from PET (mean of 3 determinations)

	Mean Migration Values μg/kg					
	Plastic 1 + No additive		Plastic 2 + Non-polar carrier		Plastic 3 + Polar carrier	
	On Receipt	After Storage	On Receipt	After Storage	On Receipt	After Storage
iC8	25*	<20	<20	<20	<20	<20
95% EtOH	25	98	25	121	20	90
10% EtOH	<20	<20	<20	<20	<20	<20
3% AA	<20	<20	<20	<20	<20	<20

* mean of 2 results only

LOD for SY114 = 20 μg/kg both when analysed on receipt and after storage.

To evaluate the effect of the additive, the ratio of migration from sample 1 with that from the samples with additive was calculated and are given in table 17. If the ratio is >1 then migration has increased and if this ratio is < 1 then migration has decreased.

Table 17: Comparison of migration Results for PET samples 1 to 3 (mean of 3 determinations)

		Ratio of Mean Migration in sample /Mean Migration from sample 1					
	R/Time	Plastic 1 + No additive		Plastic 2 + Non-polar carrier		Plastic 3 + Polar carrier	
iC8		On Receipt	After Storage	On Receipt	After Storage	On Receipt	After Storage
Peak 3 S005981/6	32.3	1	N/D	N/D	N/D	N/D	N/D
95% EtOH							
Peak 3 S005981/6	32.3	1	1	1.0	1.2	0.8	0.9
Peak S005981/11	43.6	1	1	1.1	1.3	1.5	1.0

Numbering in red indicates increase of migration on storage

Numbering in blue indicates decrease of migration on storage

N/D = Not Detected

From the triplicate analyses the precision of the analyses was calculated for each sample for both SB104 and SY114. From these precision calculations the mean Relative standard deviation (RSD) was calculated and found to be

RSD SB 104	5.2%
RSD SY 114	8.2%

These standard deviation values include analytical errors, any variation originating from the migration test and variations arising from the production of the samples.

Migration into the aqueous simulants was in all cases below the limit of detection. Migration into isooctane was in almost all cases was not detected. Migration into 95% Ethanol was the most severe which is generally accepted for this polymer.

Migration of SB 104 changed marginally during storage. The effect of the additives on migration was also marginal with the ratio of areas when compared to the plastic with no additive ranges from 1.0 to 1.5.

Migration of SY 114 was found to change on storage in all cases increasing in one case from 25 μg/kg to 121 μg/kg. This increase occurred both where additive was present and where it was not. In addition, SY 114 was the more polar of the 2 colourants being tested. The polar carrier appear to reduces migration of the polar colourant to small degree when compared to the non-polar carrier. This may be explained if the carrier helps to incorporate the colourant into the plastic matrix more readily.

Plastics 4 to 6 HDPE

These were samples of HDPE containing the colourants S005981/1 (PR 254 from China) and S005981/2 (also PR254 from China). The 2 colourants were blended at their maximum level together to provide as much information as possible on the effects of additives on migration. Plastic sample 4 contained no additive, plastic sample 5 contained erucamide at 3000 ppm and plastic sample 6 contained 5000 ppm of a 2:1 blend of Glycerol monostearate (GMS) and Bis ethoxylated amine (BEA).

Colourant S005981/1. When evaluating this colourant at least 5 substances were found in this colourant. It was possible to tentatively identify 2 of these compounds but to date the remaining 3 are still unknown.

Colourant S005981/2. At least 3 potential migrants were found. It was possible to tentatively identify one of these compounds but to date the remaining 2 are still unknown. One of these peaks was present in S005981/1 therefore the migration of 7 different compounds is being investigated. These are:-

Peak 1	From S005981/2, retention time 17.4 minutes, 2-chlorobenzamide
Peak 2	From both S005981/1 and 2, retention tine 28.6 minutes, unknown
Peak 3	From S005981/1, retention time 28.9 minutes, unknown
Peak 4	From S005981/1, retention time 29.8 minutes, unknown
Peak 5	From S005981/1, retention time 32.6 minutes, 2,4-bis(4-chlorophenyl)-6-phenyl-1,3,5 Triazine
Peak 6	From S005981/1, retention time 34.0 minutes 2,4,6-tris(4-chlorophenyl)1,3,5 Triazine
Peak 7	From S005981/2, retention time 34.9 minutes, unknown

Chromatograms and spectra of the impurities are given in appendix 2

It was not been possible to find a source of the triazines for calibration purposes and as the identity of the other peaks was unknown peak areas of the migrating compounds have been used to compare migration between the 3 samples. This has been used to evaluate the effect of additives and storage on migration. To compare migration for the different samples, the ratios of the mean peak areas of samples with additive against the mean peak area of the sample with no additive were calculated. If this ratio is >1 then migration has increased and if this ratio is < 1 then migration has decreased. The results are tabulated below.

Table 18 Migration Results for HDPE samples 4 to 6 Mean Peak areas

		Mean areas					
	R/Time	Plastic sample 4 No additive		Plastic sample 5 + erucamide		Plastic sample 6 + GMS/BEA blend	
iC8		On Receipt	After Storage	On Receipt	After Storage	On Receipt	After Storage
Peak 1	17.4	N/D	N/D	N/D	N/D	N/D	N/D
Peak 2	28.6	3294651	453296	3616128	521775	3497652	546099
Peak 3	28.9	45532	3514	64666	5890	59129	6514
Peak 4	29.8	1564379	226531	1662371	263991	1631979	247596
Peak 5	32.6	204406	11883	221742	12141	195619	12989
Peak 6	34	492341	30505	543877	30864	557452	37270
Peak 7	34.9	N/D	N/D	N/D	N/D	N/D	N/D
95% EtOH							
Peak 1	17.4	N/D	N/D	N/D	N/D	N/D	N/D
Peak 2	28.6	2332663	826645	2832680	1071437	2954263	1116118
Peak 3	28.9	34700	5922	40221	13668	27250	15966
Peak 4	29.8	831859	281732	942389	372524	1091020	359537
Peak 5	32.6	89860	22531	116508	25413	162370	28178
Peak 6	34	245694	47912	264278	61055	316433	68962
Peak 7	34.9	N/D	N/D	N/D	N/D	N/D	N/D

N/D = Not Detected. No migration was found into 3% acetic acid, 10% ethanol

Note: It can be observed that the areas differ markedly before and after storage. This can be attributed to the fact that the samples were analysed at different times using different instruments. The instrument performance standards used to validate the GCMS were analysed at the same time and reflect these changes in response. The signal noise also changed between the 2 instruments so the LOD was not affected. The areas were not corrected for these changes of response as it will not influence the comparison of migration results reported in table 19

Table 19: Comparison of migration Results for HDPE samples 4 to 6 (mean of 3 determinations)

		Ratio of Mean areas from sample/Mean areas from sample 4					
	R/Time	Plastic sample 4 No additive		Plastic sample 5 + erucamide		Plastic sample 6 + GMS/BEA blend	
iC8		On Receipt	After Storage	On Receipt	After Storage	On Receipt	After Storage
Peak 1	17.4	N/D	N/D	N/D	N/D	N/D	N/D
Peak 2	28.6	1	1	1.1	1.2	1.1	1.2
Peak 3	28.9	1	1	1.4	1.7	1.3	1.9
Peak 4	29.8	1	1	1.1	1.2	1.0	1.1
Peak 5	32.6	1	1	1.1	1.0	1.0	1.1
Peak 6	34	1	1	1.1	1.0	1.1	1.2
Peak 7	34.9	N/D	N/D	N/D	N/D	N/D	N/D
95% EtOH							
Peak 1	17.4	N/D	N/D	N/D	N/D	N/D	N/D
Peak 2	28.6	1	1	1.2	1.3	1.3	1.4
Peak 3	28.9	1	1	1.2	2.3	0.8	2.7
Peak 4	29.8	1	1	1.1	1.3	1.3	1.3
Peak 5	32.6	1	1	1.3	1.1	1.8	1.3
Peak 6	34	1	1	1.1	1.3	1.3	1.4
Peak 7	34.9	N/D	N/D	N/D	N/D	N/D	N/D

Numbering in red indicates increase of migration on storage

Numbering in blue indicates decrease of migration on storage

N/D = Not Detected. No migration was found into 3% acetic acid, 10% ethanol

From the triplicate analyses the precision of the analyses was calculated for each sample for all substances detected. From these precision calculations the mean Relative standard deviation (RSD) was calculated and found to be:

RSD Peak 2	10.0%
RSD Peak 3	17.8%
RSD Peak 4	6.5%
RSD Peak 5	10.7%
RSD Peak 6	7.2%

These standard deviation values include analytical errors, any variation originating from the migration test and variations arising from the production of the samples.

Of the 7 impurities in the colourants 5 are found to migrate. In general, migration was higher into iC8 than into 95%EtOH.

The additives enhance migration to a small degree and in some cases there is some further increase detected on storage. In particular for peak 3, retention time 28.9 mins where the peak area ratio has increased for isooctane and 95%EtOH for both samples 5 and 6. For 1 case, peak 5 migration into 95% EtOH from sample 6, a decrease was noted on storage

Compound 3 has the highest error of analysis therefore some of this change could be attributable to variations in production of the sample and analytical error, however, the variations are too large for this be the only cause. It has not been possible to identify this compound, even after help from industry but based on the mass spectra it is a chlorinated compound. This may in turn mean it is polar and could therefore partition to a greater extent in the additives. As the additives bloom to the surface with time this compound could be concentrated near the surface of the plastic giving rise to increased migration. This peak was shown to have increased migration after storage with both additives more so than any other compound.

The compounds that were analysed as instrument performance standards it would be expected that the limit of detection for the instrument would be significantly better than 32 μg/kg.

Plastics 7 to 9 LDPE

These were samples of LDPE containing colourant S005981/5, PB15:1. Plastic sample 7 contained no additive, plastic sample 8 contained erucamide at 1000 ppm and plastic sample 9 contained 2000 ppm of a 2:1 blend of Glycerol monostearate (GMS) and Bis ethoxylated amine (BEA).

There were 10 impurities found in this colourant and these are listed below.

Peak 1	RT 13.9 mins 1-dodecene
Peak 2	RT 16.8 mins 1-tetradecene
Peak 3	RT 17.82 mins 1-chlorododecane
Peak 4	RT 17.88 mins 1-decene?

Peak 5	RT 19.3 mins hexadecane
Peak 6	RT 20.3 mins 1-chlorotetradecane
Peak 7	RT 21.6 mins Octadecane
Peak 8	RT 22.4 mins 1-chlorohexadecane
Peak 9	RT 30.4 mins phthalimidomethylphthalimide
Peak 10	RT 31.4 mins Unknown

Many of these are long chain hydrocarbon or simple chlorinated long chain hydrocarbons. It has not been possible to detect migration of these from LDPE into any of the food simulants.

Chromatograms and spectra of the impurities are given in appendix 2.

Plastics10 to 12 PP

These were samples of PP containing colourant S005981/17, PG 7. The colourant was blended at its maximum level (0.3%) to provide as much information as possible on the effects of additives on migration. Plastic sample 10 contained no additive, plastic sample 11 contained erucamide at 5000 ppm and plastic sample 12 contained 5000 ppm of a 2:1 blend of Glycerol monostearate (GMS) and Bis ethoxylated amine (BEA).

Colourant S005981/17. When evaluating this colourant at least 9 impurities were found. It has not yet been possible to identify some of these compounds.

Peak 1	RT 11.2 min 1,3-Dichlorobenzene
Peak 2	RT 16.9 min Isolongifolene
Peak 3	RT 23.7 min Unknown
Peak 4	RT 23.97 min Unknown
Peak 5	RT 24.04 min simonelite
Peak 6	RT 25.1 min Unknown
Peak 7	RT 26.6 min Resin acids
Peak 8	RT 27.5 min 13-isopropylpodocarpa-8,11,13-trien-15-oic acid
Peak 9	RT 27.8 min resin acids

Of these low migration was found for the compound with RT 16.9 into isooctane and tentatively identified as isolongifolene. This was found to migrate from sample 11, the plastic with erucamide, into isooctane only. Migration was found from the sample tested on receipt but not from the sample tested after storage. Chromatogram and spectra of the impurities are given in appendix 2. No other migration was found into any other simulant.

Plastics 13 to 15 PP

These were samples of PP containing the colourant S005981/18 (PB15:1 from China). The colourant was blended at its maximum level (0.3%) to provide as much information as possible on the effects of additives on migration. Plastic sample 13 contained no additive, plastic sample 14 contained erucamide at 5000 ppm and plastic sample 15 contained 5000 ppm of a 2:1 blend of Glycerol monostearate (GMS) and Bis ethoxylated amine (BEA).

Colourant S005981/18. When evaluating this colourant at least 10 impurities were found. It has not yet been possible to identify some of these compounds.

Peak 1	RT 13.7 min, 1-dodecene
Peak 2	RT 17.5 min, 1-chlorodecane
Peak 3	RT 23.1 min, palmitic acid
Peak 4	RT 24.9 min, stearic acid
Peak 5	RT 27.5 min, unknown
Peak 6	RT 29.2 min, dodecanol
Peak 7	RT 30.1 min, phthalimidomethylphthalimide
Peak 8	RT 30.9 min, hexadecanoic acid dodecyl ester
Peak 9	RT 32.2 min, lauryl stearate
Peak 10	RT 33.4 min, myristal stearate

Chromatograms and spectra of the impurities are given in appendix 2

The compounds listed as peaks 8, 9 and 10 were found to migrate and peak areas of the migrating compounds have been used to compare migration between the 3 samples. The compounds listed as peaks 1 to 7 did not migrate and for clarity are not considered.

Peak areas of the eluting species have been recorded and the mean peak areas for the triplicate measurements calculated. The mean peak areas from the samples with additives are compared to the mean peak area of the sample with no additive and the ratios for the areas have been determined. If the ratio is >1 then migration has been enhanced and if the ratio is < 1 migration has been reduced when compared to migration of the sample with no additive. The ratios are given in table 21.

Table 20: Migration Results for PP samples 13 to 15 Mean Peak areas

		Mean areas					
	R/Time	Plastic sample 13 No additive		Plastic sample 14 + erucamide		Plastic sample 15 + GMS/BEA blend	
iC8		On Receipt	After Storage	On Receipt	After Storage	On Receipt	After Storage
Peak 8	30.9	3485432	2022618	15868093	6995600	5148792	2766191
Peak 9	32.2	6205367	3278169	11336255	5467640	10719021	5142772
Peak 10	33.4	1483237	742354	3904448	1327504	2665056	1121638
95% EtOH							
Peak 8	30.9	740727	435907	961481	651386	1462390	622829
Peak 9	32.2	1245179	576495	2396193	1175258	2513703	1000175
Peak 10	33.4	N/D	52274	N/D	78002	290795	202194

- N/D = Not Detected

Table 21: Comparison of migration from PP Sample 13 to 15 (mean of 3 determinations)

		Ratio of mean areas from samples/Mean areas from sample 13					
	R/Time	Plastic sample 13 No additive		Plastic sample 14 + erucamide		Plastic sample 15 + GMS/BEA blend	
iC8		On Receipt	After Storage	On Receipt	After Storage	On Receipt	After Storage
Peak 8	30.9	1	1	4.6	3.5	1.5	1.4
Peak 9	32.2	1	1	1.8	1.7	1.7	1.6
Peak 10	33.4	1	1	2.6	1.8	1.8	1.5
95% EtOH							
Peak 8	30.9	1	1	1.3	1.5	2.0	1.4
Peak 9	32.2	1	1	1.9	2.0	2.0	1.7
Peak 10	33.4	N/D	1	N/D	1.5	N/A*	3.9

Numbering in red indicates increase of migration on storage

Numbering in blue indicates decrease of migration on storage

- N/A = Not applicable: Migration was found for this sample, however, as no migration found in sample 13 it was not possible to obtain a ratio by dividing by 0
- N/D = Not Detected

From the triplicate analyses the precision of the analyses was calculated for each sample for all substances detected. From these precision calculations the mean Relative standard deviation (RSD) was calculated and found to be

RSD Peak 8	14.5%
RSD Peak 9	9.6%
RSD Peak 10	11.8%

These standard deviation values include analytical errors, any variation originating from the migration test and variations arising from the production of the samples.

Of the impurities in the colourant, 3 were found to migrate into iC8 and 95% EtOH. No migration was found into the aqueous simulants. The substances that did migrate were in all cases long chain fatty acid esters.

The additives enhanced migration of these compounds into both simulants. The additives are long chain compounds with a polar end group that encourages them to bloom from the plastic. The long chain fatty acid ester may be soluble in the additive chain which as it blooms to the surface will carry the substance from the colourant with it and promote migration. In most cases this effect appears to occur quite rapidly with long term storage having little effect on migration. Some minor variations were noted after long term storage. All but 2 of these differences could be attributed to experimental error. These were for the migration of peak 10 into isooctane from sample 14 and peak 8 from sample 15 into 95%EtOH. In both these cases a minor drop in migration was observed for these 2 compounds. The reason for this drop is unclear but after taking into account the error the drop maybe considered to be relatively minor.

Based on a number of compounds that were analysed as instrument performance standards it would be expected that the Limit of Detection for the instrument would be significantly better than 32 μg/kg.

8. Conclusions

In this project 24 colourants have been screened for impurities. The screening was achieved using solvent extraction procedures followed by GCMS and LCMS analysis. The solvent extraction procedure was intended to be severe so that all possible potential migrants were detected.

As found in the previous project those produced in China and India appear to contain more potential migrants than those originating from Europe.

With help from industry, prior knowledge of the colourants and using the Wiley7n search library, it was possible to identify many of the components in the colourants, however, it has not been possible to identify all the contaminants.

Of the colourants screened 7 were selected for further investigation. Again with help from industry, selection was based on the number of contaminants found in the initial colourant screening, colourant type and polymer in which the colourant is used. Using these colourants, up to 15 plastics/additive/colourant blends were prepared by manufacturers and tested for migration immediately on receipt and after storage for at least 3 months.

Of the colourants investigated for migration under the analytical test conditions used it was possible it identify 75% of the potential migrants present.

In general, migration was found to be low with no migration found from LDPE. Migration using 95% EtOH was found to be more severe than iC8 for PET but is less severe than iC8 for the polyolefins. This is a recognised effect of 95% EtOH. PET is a polar polymer and is more readily swollen by the polar ethanol solution. Isooctane being non-polar does not tend to penetrate the polymer and thereby promote migration. No migration was detected into the aqueous simulants 10% EtOH and 3% acetic acid from any of the test samples.

The presence of additives in PET did not change migration of colourant associated substances to any significant effect, however, the additives used in this polymer were not intended to bloom to the surface. Long term storage did increase migration of SY114 by a factor of at least 4 fold in all samples including the control sample.

For HDPE in general migration was not affected by the additive, however, for 1 compound which is still not identified migration increased by a factor of 2.7 over the control sample when the antistatic additive blend was present.

For PP which was blended with S005981/18, a PB 15:1, 3 of the 10 components found in the colourant were found to migrate. The presence of both additives increased migration both when the samples were tested on receipt and when tested after storage. This finding could hold true for other migrants not related to colourants. Therefore, prediction of migration using migration modeling may give underestimations when additives that are designed to bloom to the surface are present.

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