Safety Assessment: Outcome of the Assessment of 2-Hydroxyethyl Methacrylate Phosphate as a Monomer for use in the Manufacture of Plastic Food Contact Materials and Articles

Reference number RP1190

Assessment finalised: 21/06/2024

Following the publication of the finalised safety assessment, an agreement was reached with the applicant to change the name of the petitioned substance from 2-hydroxyethyl methacrylate phosphate to phosphoric acid, mixed esters with 2-hydroxyethyl methacrylate. Whilst this has not been updated in the published assessment, these two names can be used interchangeably and does not impact the assessment outcome.

Contents

1. Summary

An application was submitted to the Food Standards Agency (FSA) and Food Standards Scotland (FSS) in January 2021 by Keller and Heckman LLP for the authorisation of 2-hydroxyethyl methacrylate phosphate (HEMAP) as a monomer in a commercial product for use in the manufacture of kitchen countertops and sinks that are intended for contact with all types of food.

All components of the commercial product are listed in assimilated Regulation EU No 10/2011 on plastic materials and articles intended to come into contact with food. The application and the following assessment are for HEMAP only, not the commercial product.

Satisfactory information regarding the identity of substance, physical and chemical properties, intended application of substance, data on migration of substance and toxicological data were submitted.

To support the FSA and FSS in evaluating the dossier, the Joint Expert Group on Food Contact Materials (FCMJEG) were asked to review the dossier submitted by the Applicant and the subsequent additional information requested. The FCMJEG concluded that the there was no concern to human safety from the use of HEMAP in the commercial product to be used in the manufacture of kitchen countertops and sinks. The Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment (COT) also reviewed the FCMJEG safety assessment agreeing with the conclusions of the FCMJEG.

The views of the FCMJEG and COT have been taken into account in this safety assessment which represents the opinion of the FSA and FSS on the authorisation of HEMAP as a monomer for use in the manufacture of kitchen countertops and sinks.

2. Introduction

The FSA and FSS have undertaken a risk assessment for the authorisation of HEMAP as a monomer for use in the manufacture of kitchen countertops and sinks.

The dossier was evaluated on behalf of the FSA and FSS by the FCMJEG. In line with assimilated Regulations EU No 10/2011 and EC No 1935/2004 the assessment has considered the aspects of the monomer such as physical and chemical properties, migration data and toxicological data and its conditions of use, which has formed the basis and structure for the assessment.

With thanks to members of the FCMJEG during the course of the assessment who were: Dr Stuart Adams, Dr Emma Bradley, Dr Gill Clare, Dr Sibylle Ermler, Dr Natalia Falagan, Dr Jenny Odum and Dr Michael Walker.

This document outlines the conclusions of the FCMJEG assessment on the safety of the proposed use of HEMAP in kitchen countertop and sinks.

3. Assessment

3.1 Identity of the substance

3.1.1 Starting substances

2-hydroxyethyl methacrylate phosphate (HEMAP) is manufactured by mixing multiple starting substances, including 2-hydroxyethylmethacrylate (HEMA).

The following substances (that make up HEMAP) are formed as successive mono-, di-, and tri-hydroxyethyl methacrylate esters, on the phosphate or diphosphate cores:

- Mono-substituted phosphoric acid
- Di-substituted phosphoric acid
- Tri-substituted phosphoric acid
- Substituted diphosphates.

Reviewing the information provided, the FCMJEG agreed that no by-products were anticipated to form in the production of HEMAP but that any impurities/substances detected were residuals of the starting substances.

3.1.2 Physical and chemical properties

As HEMAP is never separated from the commercial product, the physical and chemical properties are the commercial product and not HEMAP itself.

The final commercial product has a freeze and melting point below -80°C, and a decomposition temperature of 225°C and is not soluble in water but has good solubility in ethanol and olive oil (greater than 1% at 25°C).

3.1.3 Food contact material

HEMAP is used as a minor monomer in the production of the polymer fraction of acrylic counter tops and sinks. The maximum percentage in formulation is 0.35% of HEMAP.

As such, HEMAP can come into contact with all types of food prepared in kitchens resulting in the potential for migration from the countertops manufactured with the substance.

Considering all available information, the FCMJEG agreed that under the foreseeable conditions of use, the substance was not expected to be reactive once incorporated into the countertop. Furthermore, no hydrolysis, unintentional decomposition/transformation or interactions with food substances were expected.

In addition, the FCMJEG agreed with the Applicant that under anticipated use, it is expected that the countertops and sinks would be cleaned before each use, including initial use.

3.2 Data on migration of substance

3.2.1 Contact time and temperature

The migration test samples were sheets of solid surface acrylic countertops that were fabricated both with and without the petitioned monomer (i.e. HEMAP).

The duration of direct contact with food is not expected to exceed several hours (48 hours as a worse case) at room temperature (RT, approximately 20-25° C), or a short time (e.g., 1 hour) at high temperature (up to 70ºC), followed by cooling and additional contact time at RT, unlikely to be exceeding 2 hours. Only repeated use applications are considered. Although some foods, e.g., certain raw and uncut fruits, may on occasion be in contact with a countertop over a longer time period, they do not present an extractive medium into which the components of the countertop are readily susceptible to migration.

Hot baked goods or cooked meats are not typically expected to be placed in direct contact with acrylic countertops. However, this exposure scenario was used for the migration study and therefore was considered to represent a reasonable worst-case scenario by the Applicant. The Applicant assumed the standard contact area to be 6 dm^2 per kg food. Fatty foods present an extractive medium into which the countertop components may migrate. Under normal conditions of use, fatty foods would only be expected to be left on countertops or sinks for very short periods of time prior to consumption. In practice, foodstuffs are generally in or on another article (e.g., fruit basket or chopping board). The FCMJEG agreed with the Applicant's conclusion that these conditions of use would be highly exaggerative and would therefore be reflective of a conservative model of the intended use.

The assimilated Regulation EU No 10/2011 requires that compliance for repeated-use food-contact articles like countertops shall be confirmed on the basis of the level of the migration found in the third extraction carried out on a single sample using a fresh portion of simulant for each exposure.

For this application, single extractions were conducted for 10 days (240 hours) at 40°C using 10% ethanol and 3% acetic acid as the aqueous and acidic food simulants respectively. A single extraction for 2 days at 20°C using 95% ethanol was used as an alternative to vegetable oil. The FCMJEG agreed with the Applicant's assessment, that although three successive exposures were not carried out, the exposure conditions applied here were more severe and exaggerative of the intended use.

3.2.2 Analytical method

The precision of the test method was determined and the recovery was acceptable for all food simulants with mean recovery and coefficient of variation of:

- 10% ethanol: 98% (Bias 2%) CV 3%
- 3% acetic acid: 96% (Bias 4%) CV 4%
- 95% ethanol: 99% (Bias 1%) CV 3%

3.2.3 Specific migration

The Applicant used the standard assumption that 1 kg food is in contact with 6 dm^2 of FCM. An overview of the obtained migration results in the different food stimulants (µg/6dm^2) is given in Table 1.

Table 1. Specific migration in food simulants

	10% Ethanol (µg/ 6 dm^2)	3% Acetic acid (µg/ 6 dm^2)	95% Ethanol (µg/ 6 dm^2)
Commercial product blank	<4.0	<3.2	<2.76
Commercial product + HEMAP	4.20	24.8	2.81

3.2.4 Overall migration

Overall migration was performed with 10% ethanol and 3% acetic acid as food simulants for 10 days at 40°C. Overall migration was also performed with 50% ethanol, isooctane and vegetable oil for 10 days at 40°C and 95% ethanol for 2 days at 20°C. As a substantial amount of olive oil was absorbed by the samples and as such it was not considered to be suitable for the determination of the overall migration, 50% ethanol, 95% ethanol and isooctane were included as outlined in assimilated Regulation EU No 10/2011.

All experiments were carried out in triplicate with migration cells to obtain single side contact, with the exception of isooctane and olive oil where total immersion was performed.

Table 2 provides the results for overall migration testing. The original test report noted that a substantial amount of olive oil was absorbed by the samples.

Table 2: Overall migration into food simulants

	10% Ethanol (mg/dm^2)	3% Acetic acid (mg/dm^2)	50% Ethanol (mg/dm^2)	95% Ethanol (mg/dm^2)	Isooctane (mg/dm^2)
Commercial mixture + HEMAP	0.9 ± 0.2	2.9 ± 0.4	1.3 ± 0.2	0.3 ± 0.0	0.6 ± 0.4

3.2.5 Oligomers and reaction products

Volatiles

Commercial samples with and without HEMAP were screened for the presence of volatile, semi-volatile and non-volatile oligomers and reaction products.

The migration results for volatiles showed one peak in the chromatogram of the sample containing HEMAP, which was not seen in control samples. The peak was tentatively identified as one of the starting substances, given that it was present only in the test sample containing HEMAP and was present in HEMAP itself.

The migration of semi-volatile and non-volatile reaction and breakdown products was determined in 3% acetic acid, 10% ethanol for 10 days at 40°C, and 95% ethanol for 2 days at 20°C.

No non-volatile reaction and breakdown products related to HEMAP were detected in the food simulants above the limit of detection of 2 µg/kg.

For semi-volatiles (non-polar and polar) the total ion chromatograms (TIC) in each simulant obtained for the blank (simulant) and samples without HEMAP were compared to the TIC from samples manufactured with the mixture containing HEMAP. All peaks detected in the chromatograms from the food simulants exposed to the test samples with HEMAP, were also present in the samples without HEMAP. Hence, no peak was detected that could be related to use of 2-hydroxyethyl methacrylate in the countertop test samples.

In 10% ethanol, one of the oligomers containing propanoic acid was also tentatively identified, together with other peaks with lower intensities.

However, no additional peaks were observed for the samples with HEMAP when compared to the sample without HEMAP, nor in any of the blank food simulant samples.

Base-peak chromatograms are not specific enough to detect peaks with low intensities. Therefore, extracted ion chromatograms of the exact masses calculated for the theoretical oligomers and reaction products were used to screen for their presence.

Several peaks were present, with some being identified as the main components of HEMAP.

The FCMJEG concluded that under the conditions of the manufacture process of the solid surface countertop, the starting substances have little reactivity except towards free radical addition to the growing polymer chain.

3.3 Data on residual content of substance in the FCM

HEMAP is intended to be used as a minor monomer (0.35%) during the manufacture of acrylic countertops and sinks and therefore is intended to react into the polymer backbone. Therefore, the Applicant considered HEMAP not to be available to migrate. Equally the Applicant considered HEMAP to have multiple reactive functional groups making it therefore less likely to remain unreacted. Therefore, an actual residual content was not determined. Instead, specific migration was determined.

The FCMJEG agreed with the above considerations taken by the Applicant, specifically that migration was low and all of the HEMAP was incorporated into the backbone.

3.4 Toxicological data

As HEMAP is never separated from the final commercial product, the toxicological data presented below are for the final product and not HEMAP itself.

3.4.1 Genotoxicity

Bacterial reverse mutation test (Ames Test)

The study was conducted in accordance with test guideline OECD No. 471, using the plate incorporation method.

Salmonella typhimurium strains TA1535, TA1537, TA98, and TA100 and *Escherichia coli* strain WP2 uvrA were exposed to the test substance + HEMAP at concentrations ranging from 47 – 5000 µg/plate with or without S9-mix (post-mitochondrial supernatant from liver of rats treated with an enzyme inducer) for metabolic activation.

Clear and colourless stock solutions of the test substance (+ HEMAP) were prepared at concentrations of 50 mg/ml and 30 mg/ml in dimethyl sulfoxide (DMSO) for Experiments 1 and 2. In all tests performed, the mean numbers of His+ and Trp+ revertant colonies of the negative controls were within the acceptable range and the positive controls gave the expected increase. Therefore, the experiments were considered valid.

In Experiment 1, toxicity was observed at concentrations ≥ 1667 µg/plate in all Salmonella strains in the absence of S9-mix. Toxicity was observed at and above 750 µg/plate in strain TA1537, in the absence of S9-mix. A 1.9-fold increase in revertant colonies was noted with *S. typhimurium* TA1537 at 185 µg/plate in the absence of S9-mix. To determine whether this result was biologically relevant, a repeat assay (Experiment 2) was performed with *S. typhimurium* TA1537 without S9 at concentrations of 0, 47, 94, 188, 375, 750, 1500, or 3000 µg/plate. The highest concentration tested in this repeat assay was limited by toxicity observed in the initial assay. In the repeat assay, cytotoxicity was noted at ≥ 750 µg/plate.

The test substance (+ HEMAP) induced less than a 2-fold or dose-related increase in revertant colonies compared with that of the solvent control; because the increase noted in the initial assay was not reproducible, it was not considered biologically relevant.

Under the conditions of this study, the final product, containing HEMAP was negative/not mutagenic in the bacterial reverse mutation test (Ames test).

In vitro mammalian test

The study was conducted in accordance with test guideline OECD No. 487.

Cultured human lymphocytes were exposed *in vitro* to the test substance (+HEMAP) in the presence or absence of S9-mix for metabolic activation. Negative and positive controls were run concurrently. DMSO was chosen as the solvent as the culture medium was deemed unsuitable, due to the formation of fine oil droplets.

In Experiment 1, cells were exposed to the test substance for 4 hours, followed by a 20-hour recovery period at concentrations of 15.6, 31.3, 62.5, 125, 250, 500, 750, 1000, 1500 and 2000 µg/mL with and without S9-mix.

In Experiments 2 and 3, cells were exposed to the test substance for 20 hours with no recovery period at concentrations of 15.6 - 2000 µg/mL (Experiment 2, same as in Experiment 1).

In Experiment 3, cells were exposed to the test substance for 20 hours with no recovery period at concentrations of 50, 100, 200, 300, 375, 425, 500, 600 700 and 800 µg/mL µg/mL, without S9-mix.

The number of binucleated cells containing micronuclei in negative/solvent controls were within the acceptable range and treatment with positive controls resulted in statically significant increases in the number of binucleated cells containing micronuclei, when compared to controls. Therefore, the experiments were considered valid.

In Experiment 1, the test substance showed concentration-dependent cytotoxicity, with severe cytotoxicity (72%) at the highest concentration tested (2000 µg/mL) in the presence of S9-mix. Concentrations of 250, 500, 1000, and 1500 µg/mL (selected for micronucleus analysis) showed cytotoxicity of 4%, 0%, 17%, and 40%, respectively. The applicant noted that this condition did not meet the aimed cytotoxicity range of the OECD guideline (55 ± 5%). CBP analysis showed a low cell density on the slides, which indicated a reduction in cell number as a result of the treatment. Therefore, a top concentration with a slightly lower cytotoxicity was selected. The lower selected concentrations (500 and 250 µg/mL) showed no or low cytotoxicity, respectively, when compared to the solvent control. At the concentration of 1000 µg/mL a single culture was used for analysis, since the other culture was considered to be an outlier based on the cytokinesis-block proliferation index (CBPI) (use of single culture is in alignment with the

OECD guideline). In the absence of S9-mix, severe cytotoxicity was noted at the two highest concentrations tested (1500 and 2000 µg/mL). Concentrations of 250, 500, and 1000 µg/mL (selected for micronucleus analysis) showed cytotoxicity of 3%, 23%, and 53%, respectively, compared to the control.

In Experiment 2, severe cytotoxicity was noted at the four highest concentrations tested (750, 1000, 1500 and 2000 µg/mL) in the absence of S9-mix. The next lower concentration (500 µg/ml) showed 64% cytotoxicity, all other concentrations (250 to 15.6 µg/mL) exhibited little to no cytotoxicity. The applicant noted that 64% cytotoxicity was above the aimed cytotoxicity stated in the OECD guideline. In addition, due to a steep concentration response curve for the test substance a concentration representing moderate cytotoxicity was not obtained. Although this experiment did not fully meet the target cytotoxicity range of the OECD guideline, three concentrations (125, 250, and 500 µg/ml) together with the solvent control and positive control were selected for analysis of micronuclei induction.

In Experiment 3, the test substance showed concentration related cytotoxicity, in the absence of S9-mix. Strong cytotoxicity was noted at the four highest concentrations tested (500, 600, 700 and 800 µg/mL). Concentrations of 100, 300, and 425 µg/mL (selected for micronucleus analysis) showed cytotoxicity of 12%, 29%, and 57%, respectively, compared to the control.

There was no statistically significant ($p > 0.05$, Chi-Square one-sided test) increase in the incidence of micronuclei-containing binucleated cells in any experiment at any concentration analysed when compared to concurrent controls. There was no concentration-related increase when assessed for statistical trend.

Under the conditions of this *in vitro* micronucleus test, the test substance (+ HEMAP) was considered neither clastogenic nor aneugenic by the Applicant.

4. Discussion

The FCMJEG considered the information on the identity of the substance, the physical and chemical properties and intended application satisfactory. Results from the overall and specific migration tests demonstrated the migration was below the overall migration limit and the specific migration of HEMAP was considered to be acceptable.

The FCMJEG discussed the assumptions made by the Applicant with regards to the worst-case exposure scenario used in the migration study and the standard contact area of food with the commercial product. The FCMJEG agreed that the worst-case exposure scenario represents a conservative model of the product's intended use. The FCMJEG also agreed with the Applicant's assumption that 1kg food is in contact with 6 dm^2 of FCM.

The results of the bacterial reverse mutation assay were negative/not mutagenic, and the *in vitro* mammalian test determined HEMAP to be neither clastogenic nor aneugenic therefore the FCMJEG considered HEMAP to be of no concern for a risk to human health.

Taking into account that the specific migration of the sum of HEMAP plus its phosphate and diphosphate esters is not expected to exceed 50 µg/kg food, the FCMJEG proposed a Specific Migration Limit (SML) of 0.05 mg/kg food for HEMAP.

5. Conclusions

Satisfactory information regarding the identity of substance, physical and chemical properties, intended application of substance, data on migration of substance and toxicological data were submitted.

The available toxicology data showed HEMAP to be negative in the *in vitro* Ames test and *in vitro* micronucleus test and therefore unlikely to be of concern for potential carcinogenic and mutagenic toxicity.

To support and advise the FSA and FSS in evaluating the dossier, the Joint Expert Group on Food Contact Materials (FCMJEG) were asked to review the dossier and any subsequent submitted information by the Applicant. The Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment (COT) also reviewed the FCMJEG advice and agreed with the conclusions of the FCMJEG.

The FSA and FSS concluded, based on the FCMJEG's advice that there was no concern to human safety from the use of HEMAP in the commercial product to be used in the

manufacture of kitchen countertops and sinks. The FSA agree with the proposed SML of 0.05 mg/kg food for HEMAP.

6. Abbreviations

CBPI – Cytokinesis block proliferation index (CBPI)

COT - The Committee on Toxicity of Chemicals in Food, Consumer Products and the Environment

DMSO - Dimethylsulfoxide

FSA – Food Standards Agency

FSS – Food Standards Scotland

FCMJEG – Joint Expert Group on Food Contact Materials

HEMAP - 2-hydroxyethyl methacrylate phosphate

mg/dm^2 – milligram per square decimetre

OECD - The Organization for Economic Cooperation and Development

ppb – parts per billion

SML – Specific migration limit

TIC – Total ion chromatogram

$\mu g/dm^2$ – microgram per square decimetre

µg/kg – microgram per kilogram

For enquiries about this publication, contact the Food Standards Agency.

 Follow us on Twitter: @foodgov

 Find us on Facebook: facebook.com/FoodStandardsAgency