

AF&PA®



AMERICAN FOREST & PAPER ASSOCIATION

GROWING WITH AMERICA SINCE 1861

RECEIVED
EPA-7000

09 AUG 15 PM 1:26



August 15, 2008

By Courier

Document Processing Center (7407M)
(Attn: TSCA Section 8(e) Coordinator)
Office of Pollution Prevention and Toxics
Environmental Protection Agency
1200 Pennsylvania Avenue, N.W.
Washington, DC 20460-0001

Contain NO CBI



Re: TSCA Section 8(e) Submission of Test Results Related to Exposure to Anthraquinone

Dear Sir or Madam:

On behalf of its member companies and in accordance with Section 8(e) of the Toxic Substances Control Act ("TSCA"), 15 U.S.C. § 2607(e), the American Forest & Paper Association, Inc. ("AF&PA")¹ hereby notifies EPA of the results of tests related to the potential for human exposure to 9,10-anthraquinone, CAS Registry No. 84-65-1 ("anthraquinone").

At the request of AF&PA and its members, the National Council for Air and Stream Improvement ("NCASI") conducted two tests relevant to the potential for human exposure to anthraquinone that may be contained in paper or paperboard. Two reports describing that testing and the results are enclosed.

AF&PA and its members make this submittal pursuant to TSCA Section 8(e) because EPA guidance can be read to require reporting of any exposure information for chemicals that have been reported to cause cancer in laboratory animals, without regard to whether, in combination with information about the cancer potency of the

¹ AF&PA is the national trade association of the forest, pulp, paper, paperboard, and wood products industry. AF&PA represents member companies engaged in the growing, harvesting, and processing of wood and wood fiber, and the manufacture of pulp, paper, and paperboard products from both virgin and recycled fiber, as well as solid wood products.

313586

chemical from animal testing, the identified exposure suggests any significant risk of cancer.² Our submittal of this information under Section 8(e), therefore, is not based on a conclusion that the information indicates an actual adverse effect from exposure to anthraquinone or an actual risk to human health, either past or present.

No claim of confidentiality for information contained in this submission is made, either under TSCA Section 14(c) or any other provision.

If you have any questions regarding this notification, please contact me at the above address or call me at 202 463-2587.

Sincerely,

A handwritten signature in cursive script that reads "John L. Festa". The signature is written in black ink and is positioned above the printed name and title.

John L. Festa, Ph.D.

Senior Scientist

Enclosures

cc: AF&PA Member Companies

² A single cancer bioassay study, conducted by the National Toxicology Program of the National Institutes of Health ("NTP"), reported that anthraquinone was carcinogenic to rats and mice. The Department of Health and Human Services currently is considering a request by Chemical Products Corporation that this NTP report be withdrawn on the grounds that the sample tested, which had been derived using a process involving oxidation of anthracene, one of several processes for the manufacture of anthraquinone, was contaminated with the mutagen 9-nitroanthracene, leading to inaccurate results and conclusions.

by
Jeffrey Louch, Ph.D.
NCASI West Coast Regional Center
Corvallis, Oregon

SPECIAL REPORT NO. 08-02
JULY 2008

THE POTENTIAL FOR MIGRATION
OF ANTHRAQUINONE FROM
UNBLEACHED LINERBOARD

NATIONAL COUNCIL FOR AIR AND STREAM IMPROVEMENT

ncasi

CC AUG 15 PM 1:27

RECEIVED

Acknowledgments

This report was prepared by Jeff Louch with the assistance of Larry LaFleur, both at NCASI's West Coast Regional Center. Anna Aviza from NCASI's Northern Regional Center prepared the final document for publication. The experimental work was executed by Dean Hoy and Ginny Allen, both Sr. Research Associates at the NCASI West Coast Regional Center, with additional support provided by Larry LaFleur and Jeff Louch. The American Forest & Paper Association's Chemical & Product Stewardship Group provided input and approval of the experimental design.

For more information about this research, contact:

Jeffrey Louch, Ph.D.
Principal Scientist
NCASI West Coast Regional Center
P.O. Box 458
Corvallis, OR 97333
(541) 752-8801
jlouch@ncasi.org

Robert Fisher, Ph.D.
Vice President, Biological and Chemical Assessment
NCASI
P.O. Box 13318
Research Triangle Park, NC 27709-3318
(919) 941-6409
rfisher@ncasi.org

For information about NCASI publications, contact:

Publications Coordinator
NCASI
P.O. Box 13318
Research Triangle Park, NC 27709-3318
(919) 941-6400
publications@ncasi.org

National Council for Air and Stream Improvement, Inc. (NCASI). 2008. *The potential for migration of anthraquinone from unbleached linerboard*. Special Report No. 08-02. Research Triangle Park, N.C.: National Council for Air and Stream Improvement, Inc.



servicing the environmental research needs of the forest products industry since 1943

PRESIDENT'S NOTE

In September of 2007, California's Office of Environmental Health Hazard Assessment (OEHHA) added anthraquinone to the Proposition 65 list of chemicals known to the state to cause cancer. As a result of this action, companies have to assess the need to label products for sale or use in California that contain anthraquinone and take appropriate action by September 28, 2008. To assist members in making this determination, the industry's product sector as represented in the American Forest & Paper Association's Chemical and Product Stewardship Group requested that NCASI conduct studies (1) to develop an analysis method for anthraquinone in product, (2) to determine the potential for migration of anthraquinone from pizza delivery boxes into pizza crust to help assess the potential for exposure via indirect ingestion, and (3) to develop a partition coefficient describing the equilibrium partitioning of anthraquinone between a board sample and simulated sweat to help assess the potential for exposure via dermal contact. This report provides the results of the work NCASI executed in response to the request for information on migration into pizza crust.

In this laboratory study, the migration of anthraquinone from an unbleached kraft linerboard sample, representing a pizza delivery box, to a sample of baked pizza dough was determined using conditions characteristic of pizza delivery, consumption, and storage. Pooling the results of replicate experiments gave a mean migration (percent migration) of $3.6 \pm 1.05\%$ (mean \pm standard deviation, $n=6$).

As per the product sector's request, NCASI has also completed development of an analytical method for measuring anthraquinone in uncoated product(s), and has developed a simulated sweat-linerboard equilibrium partition coefficient for anthraquinone. The results of these efforts are summarized in related NCASI publications.

A handwritten signature in black ink, appearing to read "Ron Yeske", is positioned above the printed name.

Ronald A. Yeske

July 2008

7 1

THE POTENTIAL FOR MIGRATION OF ANTHRAQUINONE FROM UNBLEACHED LINERBOARD

SPECIAL REPORT NO. 08-02
JULY 2008

ABSTRACT

Experiments were conducted to assess the potential of anthraquinone possibly contained in food packaging such as pizza boxes to migrate into food, specifically just-cooked pizza crust brushed with vegetable oil. Briefly, pizza dough was purchased from a local pizzeria and baked according to instructions. After baking and while still hot, heated vegetable oil was brushed on the bottom of the baked crust, the crust placed on a piece of unbleached kraft linerboard and the crust-linerboard wrapped in foil. To simulate "typical" usage, this assembly was held at nominally 170°F for one hour (delivery), followed by two hours at room temperature (initial consumption) and then 48 hours at refrigerator temperatures (storage of leftovers). Subsequently, the absolute amount (mass) of anthraquinone in an exact 3"x3" square of the crust was measured. Dividing this mass by the initial mass of anthraquinone in an exact 3"x3" piece of the contact linerboard gives the fraction of anthraquinone in the linerboard that migrated into the pizza crust during the contact period.

Replicate experiments were performed as described above. Pooling the results gave a mean migration (percent migration) of $3.6 \pm 1.05\%$ (mean \pm standard deviation, n=6).

KEYWORDS

anthraquinone, food contact substance, food packaging, migration, paper, paperboard, pizza box, Proposition 65

RELATED NCASI PUBLICATIONS

Technical Bulletin No. 953 (July 2008). *NCASI Method AQ-S108.01: Anthraquinone in uncoated papers and market pulps by Soxhlet extraction and GC/MS.*

Special Report No. 08-03 (July 2008). *Determination of the anthraquinone paper/paperboard simulated sweat partition coefficient for the estimation of potential dermal contact exposure.*

Technical Bulletin No. 831 (August 2001). *Technical support document for assessing the safety of a food contact substance.*

Technical Bulletin No. 824 (May 2001). *Evaluation of food simulating liquids for paperboard food packaging.*

CONTENTS

1.0	INTRODUCTION	1
2.0	EXPERIMENTAL.....	1
2.1	Preparation of Pizza Crust and Contact Regimen	1
2.2	Analysis of Exposure Specimen and Pizza Crust	2
3.0	RESULTS.....	4
3.1	Anthraquinone in Contact Specimen of Unbleached Kraft Linerboard.....	4
3.2	Anthraquinone in Contact and Control Pizza Crust.....	4
3.3	Anthraquinone Migration	7
	REFERENCES	8

TABLES

Table 2.1 MS Ions Used in Both Full Scan and SIM GC/MS Analysis 3

Table 3.1 Single Laboratory Method Performance of NCASI Method AQ-S108.01 in Samples 5

Table 3.2 Summary of Results from Analysis of All Samples Associated with
Migration/Contact Study..... 6

THE POTENTIAL FOR MIGRATION OF ANTHRAQUINONE FROM UNBLEACHED LINERBOARD

1.0 INTRODUCTION

In response to a rodent cancer bioassay conducted by the US National Toxicology Program (NTP) on anthraquinone (CAS No. 84-65-1), California's Office of Environmental Health Hazard Assessment (OEHHA) added anthraquinone to the Proposition 65 list of chemicals known to the state to cause cancer. This listing became effective on September 28, 2007. As a result of this action, companies will have to assess the need to label products for sale or use in California that contain anthraquinone and take appropriate action by September 2008. To assist members in making this determination, the American Forest & Paper Association Chemical and Product Stewardship Group (AF&PA C&PSG) requested that NCASI conduct studies to determine the potential for migration of anthraquinone from pizza delivery boxes into pizza.

NCASI determined that in the short time available, it would not be possible to develop analytical procedures to measure trace amounts of anthraquinone in whole pizza. AF&PA C&PSG took this under advisement and determined that an acceptable alternative to using whole pizza in an exposure experiment would be to use pizza crust with vegetable oil brushed on the bottom. The group also identified an appropriate linerboard type to be used in the experiment, as well a contact regimen for simulating typical contact between pizza crust and linerboard.

2.0 EXPERIMENTAL

2.1 Preparation of Pizza Crust and Contact Regimen

Pizza dough was purchased from a local pizzeria the evening before each experiment. A roller was used to spread 18 oz of dough into an approximate 13" x 15" rectangle on a Teflon[®] coated, heavy gauge steel cooking sheet. A fork was used to poke holes in the dough at about every 2". This prevented formation of large bubbles during baking, thus giving a more uniform, flat contact surface. The dough was baked at 400°F for fifteen minutes in accordance with cooking instructions.

Upon removal from the oven, a 9"x9" square was cut from one end of the baked crust, taking care to stay at least 3/4" away from the edge. This piece of crust was weighed and the mass was recorded. Following this, a brush was used to coat the bottom of the whole piece of crust with pre-heated (400°F) vegetable oil. The bottom was then swabbed with a cotton ball to remove excess oil, the crust was reweighed, and the mass of oil added was calculated (on a surface area normalized basis). A nominal 10 mL aliquot of this oil was archived.

Immediately following addition of the oil, the still hot crust was placed oil side down on a 9" x 9" specimen of 45 lb (basis weight) brown kraft linerboard which was itself on top of a piece of foil-wrapped cardboard. The whole assembly was wrapped in foil and placed in a pre-heated commercial pizza delivery bag, keeping the crust and linerboard between 160 and 175°F. The remainder of the baked crust was wrapped in foil and placed in the same bag along with the exposed crust. This unexposed crust was used as a control to assess blank contamination and anthraquinone recovery (via matrix spiking).

The pizza crust-linerboard assembly was left in the heated bag for one hour and was then removed and stored at room temperature for two hours. It was then refrigerated ($\approx 39^{\circ}\text{F}$) for 48 hours. This process was meant to simulate normal handling of a pizza from preparation through delivery and consumption of leftovers. The process was repeated three times to provide three separate samples.

2.2 Analysis of Exposure Specimen and Pizza Crust

2.2.1 Contact Specimen of Unbleached Kraft Linerboard

The exposure specimen used in these experiments was unbleached (brown) kraft linerboard with a basis weight of nominally 45 lb containing a known amount of anthraquinone. Anthraquinone in this linerboard was determined using NCASI Method AQ-S108.01 (NCASI forthcoming), which was developed by NCASI in late 2007 and early 2008 specifically for analyses of pulps and uncoated papers. This method is currently in the queue for publication in NCASI's *Methods Manual*, and can be obtained by contacting NCASI.

Briefly, Method AQ-S108.01 calls for samples of pulp or uncoated papers to be cut into nominal 1 x 1 cm squares, 5 g of which is then extracted using 300 mL of ethanol in a Soxhlet extractor for a minimum of 12 hours. Following extraction, the ethanol is concentrated to approximately 1 mL of hexane, which is then subjected to silica gel cleanup. The analytical cut from this column cleanup is concentrated to 1 mL hexane and analyzed by full scan gas chromatography/mass spectrometry (GC/MS). Prior to extraction, samples are fortified with 5 μg 2-chloroanthraquinone as a recovery surrogate. In addition, immediately preceding silica gel cleanup, the hexane extracts are fortified with 25 μg d_{10} -benzophenone as a cleanup recovery surrogate. Assessing recoveries of these two surrogates allows for differentiating problems originating with the extraction and initial concentration steps vs. subsequent cleanup and final concentration.

Immediately prior to GC/MS analysis, final extracts are fortified with 25 μg of d_{10} -anthracene as an injection internal standard, and all quantifications are made vs. this internal standard. All internal standard quantifications use the average relative response factor (RRf) developed from a seven point calibration spanning the range from 0.2 $\mu\text{g/g}$ to 20 $\mu\text{g/g}$ (ppm, assuming an exact 5 g sample).

In this study, all analyses were performed using an HP 5890/5988A GC/MS with a Phenomenex ZB-5 GC column (30 m, 0.25 mm id, 0.25 μm , Phenomenex #7HG-G002-11). In accordance with Method AQ-S108.01, the injection port was held at 270°C and the MS interface at 290°C. The GC temperature program was 50°C (1 min hold), to 160°C at 20°C/min, to 250°C at 8°C/min, and to 300°C at 25°C/min with a final 4 min hold (helium carrier, 7 psi constant head pressure, 35 cm/sec @ 50°C). Analyses of the linerboard were performed in the full scan mode (42 to 500 amu). Table 2.1 lists the relevant mass fragments (ions) and ion abundance criteria.

Table 2.1 MS Ions Used in Both Full Scan and SIM GC/MS Analysis

Compound	Quantification Ion (m/z)	Qualitative Ions (m/z)	Ion Abundance Criteria ^a
Anthraquinone	208	180, 152	±20%
2-Chloroanthraquinone	242	214, 151	±20%
d ₁₀ -Benzophenone	110	82, 192	±20%
d ₁₀ -Anthracene	188	94, 80	±20%

^a criteria relative to mean abundance ratios established from analysis of standards under the same MS tune (e.g., if mean from analysis of standards is 50% abundance, acceptance window is 30 to 70%)

2.2.2 Pizza Crust

On completion of the contact/storage program, the contact dough was placed oiled (linerboard contact) side up on a 9" x 9" piece of solvent-rinsed glass, and 3" x 3" pieces were cut from the center using razor knives (with solvent-rinsed steel blades) and 3" x 3" pieces of solvent-rinsed glass as templates (thus obtaining multiple 3" x 3" squares). Each 3" x 3" pizza crust sample was transferred to a solvent-rinsed Petri dish and cut into small pieces (approximately 1 x 1 cm). These pieces were then transferred to a clean Soxhlet thimble containing a glass wool plug. The 3" x 3" piece of glass used to cut the dough, the razor knife, and the Petri dish were rinsed with ethanol into a beaker. This ethanol, along with any crumbs of pizza crust, was added to the Soxhlet extractor body holding the associated sample. Thus, ethanol rinses of the 3" x 3" glass template, the razor knife, and the Petri dish were incorporated in the analysis.

Pieces (3" x 3") of unexposed, un-oiled control crust were obtained as described. Once loaded into the Soxhlet thimble, a portion of the archived oil was added to the sample to simulate the oil brushed onto the bottom of the contact crust (Section 2.1).

Once in the Soxhlet thimbles, analysis of the crust samples was based on Method AQ-S108.01. The primary modifications were use of GC/MS select ion monitoring (SIM) over a calibration range spanning 20 to 2500 pg/ μ L (extract concentration), and addition of a gel permeation chromatographic (GPC; USEPA 1994) cleanup prior to the silica gel cleanup used for the linerboard.

Thus, the crust samples were fortified with 250 ng 2-chloroanthraquinone and extracted for >12 hours using 300 mL ethanol in a Soxhlet extractor. Following extraction, ethanol extracts (in the Soxhlet round bottom flasks) were concentrated to the oil using rotary evaporation (Buchi RE-11 Rotavapor, 45°C and 25 mm Hg vacuum). The oil was taken up into dichloromethane (DCM) and transferred to a tared Kuderna Danish (KD) concentrator tube, and the extract was taken back to the oil phase by KD concentration (95°C water bath). The KD concentrator tube was reweighed to determine the exact mass of oil present. The oil was then diluted into a volume of DCM allowing the whole extract to be processed through the low pressure GPC system in the minimum number of injections (0.5 g oil maximum in 3 mL DCM injections). The analytic cut from this cleanup was determined via profiling and corresponded to the period between the leading edge of the methoxychlor and perylene peaks in the GPC calibration standard (USEPA 1994). The cuts from the multiple (3-4) GPC injections were collected in a KD and concentrated to between 1 and 2 mL hexane. This hexane was fortified with 250 ng d₁₀-benzophenone as a cleanup recovery surrogate, and the extract then subjected to the silica gel cleanup specified in Method AQ-S108.01 (i.e., the same cleanup used on the linerboard sample). The analytical cut from the silica gel column was concentrated to nominally 1 mL, spiked with d₁₀-

anthracene as the internal standard, and analyzed by SIM mode GC/MS using the same GC column and temperature program used in analyses of the linerboard. The ions listed in Table 2.1 were monitored and, again, all quantifications used the average RRFs from a seven point calibration curve (20 to 2500 pg/ μ L extract concentration).

3.0 RESULTS

3.1 Anthraquinone in Contact Specimen of Unbleached Kraft Linerboard

Table 3.1 is taken directly from Method AQ-S108.01 (where it is Table 3), and summarizes surrogate recovery and matrix spike recovery results from analysis of multiple pulp and paper samples, including results obtained from analysis of the contact specimen. Duplicate analyses of the contact specimen gave 4.22 and 4.22 μ g/g, respectively (giving a relative percent difference of 0%). A single matrix spike experiment performed at 15.65 μ g/g gave 98.7% recovery, while the mean (n=3) recoveries of the recovery (2-chloroanthraquinone) and cleanup (d_{10} -benzophenone) surrogates were $94.6 \pm 3.82\%$ and $99.5 \pm 1.79\%$, respectively. Matrix spike and surrogate recovery results from the multiple analyses of this sample support the mean sample result of 4.22 μ g/g (on an as-received basis) as being both precise and unbiased.

Based on in-house measurements at the time of analysis, the contact specimen had a density of 0.143 g per square inch, corresponding to a calculated basis weight of 45.56 lb (lb per 1000 ft²). Using the measured density, a 3"x3" square of this linerboard weighs 1.29 g, and this mass of the sample contains 5.43 μ g of anthraquinone (again, the duplicate analyses gave a relative percent difference of 0%).

3.2 Anthraquinone in Contact and Control Pizza Crust

As noted, three separate migration/contact experiments were performed using three separate batches of pizza dough/crust. Duplicate samples of the contact crust were analyzed in each experiment. Two experiments included duplicate analyses of the control crust, analysis of a control crust spike (crust spiked with 250 ng anthraquinone prior to extraction), and a method blank. The third set included a single analysis of the control crust without any method blank or crust spike. In addition, a separate set of control crust spiking experiments (100 ng and 2500 ng spikes onto control crust) was performed prior to initiating the migration/contact experiments. Results from all these analyses are summarized in Table 3.2.

Table 3.1 Single Laboratory Method Performance of NCASI Method AQ-S108.01 in Samples

Compound	Concentration (ppm) ^a	MS/MSD Percent Recovery			MS/MSD or Native Duplicate Precision				
		Mean	SD	n	Acceptance Window ^b	Mean RPD (%)	SD (%)	Acceptance Limit ^c	
Anthraquinone	1	96.1	2.61	8	88.2 - 103.9	2.64	1.79	7	8.00
	4					0.02 - 4.99 ^e			
	16	95.3 - 98.7 ^d		2					
	40					3.60 - 6.73 ^e			2
2-Chloroanthraquinone	1	95.2	4.75	36	81.0 - 109.5				
	5	97.7	3.68	42	86.7 - 108.8				

^a (nominal) equivalent sample concentrations assuming 5.0 g sample size

^b calculated as [mean \pm (3 x SD)]

^c calculated as [mean + (3 x SD)]

^d one spike on each of two separate samples; range is given and no SD or acceptance window calculated

^e one pair of native duplicates for each of two separate samples; no range is given and no SD or acceptance window calculated

Table 3.2 Summary of Results from Analysis of All Samples Associated with Migration/Contact Study

Sample	Anthraquinone			2-Chloroanthraquinone		d ₁₀ -Benzophenone	
	native result (ng) ^a	spike level (ng)	% recovery	spike level (ng)	% recovery	spike level (ng)	% recovery
Batch 1 ^b :							
control crust spike	NA ^c	100	120	100	123	250	99
control crust spike	NA	2500	89	2500	96	25000	94
Batch 2:							
control crust	36.2			250	100	250	102
contact crust	208			250	102	250	102
contact crust DUP	243			250	107	250	102
Batch 2 Re-extract ^d :							
control crust	12.5			NS ^e	0.16	250	92
contact crust	9.85			NS	0.21	250	90
contact crust DUP	8.92			NS	0.44	250	90
Batch 3:							
method blank	9.94			250	52	250	96
control crust ^f	20.1			250	64	250	94
control crust DUP	34.4			250	111	250	98
control crust MS	(34.4) ^g	250	92 ^g	250	97	250	99
contact crust	269			250	112	250	101
contact crust DUP	189			250	105	250	100
Batch 4:							
method blank	18.8			250	90	250	95
control crust	34.0			250	106	250	97
control crust DUP	38.7			250	102	250	97
control crust MS	(36.4) ^h	250	98 ^h	250	106	250	100
contact crust	116			250	100	250	100
contact crust DUP	152			250	93	250	101

^a absolute mass (ng) found in 3"x3" piece of pizza crust

^b extracts analyzed by both full-scan GC/MS and GC/MS SIM; no unspiked crust analyzed

^c NA is not analyzed

^d after collecting ethanol from initial extraction, Batch 2 samples were re-extracted with fresh ethanol; no additional spike of 2-chloroanthraquinone prior to this repeat extraction, but d₁₀-benzophenone was spiked prior to silica gel cleanup

^e NS is not spiked

^f unknown fraction of extract lost during GPC cleanup; recovery of anthraquinone and 2-chloroanthraquinone known to be biased low

^g spike recovery calculated using result from DUP only (see table note d)

^h spike recovery calculated using mean of two control dough results

As indicated in the Table 3.2 notes, a portion of the Batch 3 control crust extract was lost during GPC cleanup, and this is reflected in the low recovery obtained for 2-chloroanthraquinone. As a consequence, the associated anthraquinone result is known to be biased low, so only the single result from the duplicate analysis is considered valid.

The recovery of 2-chloroanthraquinone was also low in the Batch 3 method blank, which was generated by spiking 300 mL of ethanol, refluxing it in an empty Soxhlet extractor, and working up the resulting extract as if it was a sample (including multiple passes through the GPC cleanup). This low recovery was attributed to loss to glassware during initial concentration (rotary evaporation); i.e., use of rotary evaporation in the absence of any oil keeper resulted in deposition of chemical over the entire inside surface area of the 500 mL round bottom flask, and subsequent rinsing with DCM to transfer the extract into the KD concentrator tube was not done in a manner providing full recovery of this deposited chemical. To check this, 1.5 g of oil was added to the Batch 4 method blank. This analysis gave 90% recovery of the 2-chloroanthraquinone spike and 18.77 ng of anthraquinone. This outcome supports the need for the oil as a keeper in the rotary evaporation step (all crust samples had this oil), and indicates that laboratory contamination contributed nominally 20 ng background to all anthraquinone measurements.

As summarized in Table 3.2, the Batch 2 crusts were extracted twice; i.e., following collection of the initial ethanol extract all crust samples were extracted a second time with a fresh 300 mL aliquot of ethanol. This was done to check extraction efficiency. Although at first glance the results appear to suggest that the initial extraction left some anthraquinone in the crusts, this conclusion is not supported by the overall results:

- Given that the control and contact crusts had initial anthraquinone concentrations differing by a nominal order of magnitude, the fact that the results from the re-extractions are essentially the same suggests that the source of anthraquinone in these re-extractions was not the original samples.
- Method blank results for anthraquinone are almost twice the levels of anthraquinone in the extracts from the crust re-extractions, indicating that the anthraquinone found on re-extraction was almost certainly laboratory contamination (results suggest that the Soxhlet glassware itself might have been the source of on the order of half of this background anthraquinone).

Overall, results from these re-extractions support Soxhlet extraction with ethanol as providing quantitative recovery of anthraquinone from this crust. Consistent with this, matrix spike recoveries ranged from 89 to 120%, with an overall mean of $100 \pm 14.0\%$ ($n=4$). In addition, the mean recovery of the pre-extraction surrogate (2-chloroanthraquinone) was $103 \pm 8.4\%$ ($n=15$), while the mean recovery of the cleanup surrogate was $97 \pm 4.0\%$ ($n=20$). These recoveries are in agreement with Table 3.1 results from analyses of pulps and uncoated papers. Finally, the mean anthraquinone mass found in the control crust replicates was 35.8 ± 2.16 ng ($n=4$). Overall, these results indicate that the analysis is capable of providing precise and unbiased measurements of anthraquinone in this pizza crust.

3.3 Anthraquinone Migration

Pooling the results from all contact crust replicates gives 196.1 ± 56.78 ng (mean \pm standard deviation, $n=6$) anthraquinone. Dividing this by the original mass of anthraquinone in the contact specimen (5.43 μ g) gives a percent migration of anthraquinone from the specimen into the pizza crust of $3.6 \pm 1.05\%$ (mean \pm standard deviation, $n=6$).

REFERENCES

- National Council for Air and Stream Improvement, Inc. (NCASI). 2008. *NCASI Method AQ-S108.01: Anthraquinone in uncoated papers and market pulps by Soxhlet extraction and GC/MS*. Technical Bulletin No. 953. Research Triangle Park, NC: National Council for Air and Stream Improvement, Inc.
- United States Environmental Protection Agency (USEPA). 1994. Method 3640A: Gel-permeation cleanup. SW-846. <http://www.epa.gov/epaoswer/hazwaste/test/pdfs/3640a.pdf>. Washington, DC: United States Environmental Protection Agency.

ncasi

NATIONAL COUNCIL FOR AIR AND STREAM IMPROVEMENT

RECEIVED
08 AUG 15 PM 1:27

**DETERMINATION OF THE ANTHRAQUINONE
PAPER/PAPERBOARD SIMULATED
SWEAT PARTITION COEFFICIENT FOR
THE ESTIMATION OF POTENTIAL
DERMAL CONTACT EXPOSURE**

**SPECIAL REPORT NO. 08-03
JULY 2008**

**by
Lawrence LaFleur
NCASI West Coast Regional Center
Corvallis, Oregon**

Acknowledgments

The report was prepared by Larry LaFleur with assistance from Dr. Jeff Louch, both from the NCASI West Coast Regional Center. The exposures and analyses of the simulated sweat were performed by Larry LaFleur. Dr. Barry Malmberg of the West Coast Regional Center and Dr. Paul Van Deusen of the NCASI Statistics and Model Development Group in Lowell, MA developed and implemented the solution to Fick's second law. Dean Hoy of the West Coast Regional Center performed the analysis of the paper sample. The American Forest & Paper Association's Chemical & Product Stewardship Group provided input and approval of the experimental design. The document was prepared for publication by Karen Phelps and Anna Aviza.

For more information about this research, contact:

Lawrence LaFleur
NCASI Fellow
NCASI West Coast Regional Center
P.O. Box 458
Corvallis, OR 97333
(541) 752-8801
llafleur@ncasi.org

Robert Fisher, Ph.D.
Vice President, Biological and Chemical Assessment
NCASI
P.O. Box 13318
Research Triangle Park, NC 27709-3318
(919) 941-6409
rfisher@ncasi.org

Jeffrey Louch, Ph.D.
Principal Scientist
NCASI West Coast Regional Center
P.O. Box 458
Corvallis, OR 97333
(541) 752-8801
jlouch@ncasi.org

For information about NCASI publications, contact:

Publications Coordinator
NCASI
P.O. Box 13318
Research Triangle Park, NC 27709-3318
(919) 941-6400
publications@ncasi.org

National Council for Air and Stream Improvement, Inc. (NCASI). 2008. *Determination of the anthraquinone paper/paperboard simulated sweat partition coefficient for the estimation of potential dermal contact exposure*. Special Report No. 08-03. Research Triangle Park, N.C.: National Council for Air and Stream Improvement, Inc.



servicing the environmental research needs of the forest products industry since 1943

PRESIDENT'S NOTE

In September of 2007, California's Office of Environmental Health Hazard Assessment (OEHHA) added anthraquinone to the Proposition 65 list of chemicals known to the state to cause cancer. As a result of this action, companies have to assess the need to label products for sale or use in California that contain anthraquinone and take appropriate action by September 28, 2008. To assist members in making this determination, the industry's product sector as represented in the American Forest & Paper Association's Chemical and Product Stewardship Group requested that NCASI conduct studies (1) to develop an analysis method for anthraquinone in product, (2) to determine the potential for migration of anthraquinone from pizza delivery boxes into pizza crust to help assess the potential for exposure via indirect ingestion, and (3) to develop a partition coefficient describing the equilibrium partitioning of anthraquinone between a board sample and simulated sweat to help assess the potential for exposure via dermal contact. This report provides the results of the work NCASI executed in response to the request for information on anthraquinone partitioning between simulated sweat and an uncoated paper product.

In this laboratory study, the partitioning of anthraquinone was investigated using a protocol that utilized simulated sweat in contact with an unbleached kraft paper sample of known anthraquinone content for time intervals of 1, 2, 4, 6, 8, and 24 hours at a temperature of 25°C. The experiments were conducted accordingly and the data were modeled using Fick's Second Law to derive an estimate of the equilibrium partition coefficient of 0.000928 ± 0.000140 (95% C.I.).

As per the product sector's request, NCASI has also completed development of an analytical method for measuring anthraquinone in uncoated product(s), and has measured the fraction of anthraquinone that migrated into pizza crust from linerboard under relevant conditions. The results of these efforts are summarized in related NCASI publications.

A handwritten signature in black ink, appearing to read "Ron Yeske".

Ronald A. Yeske

July 2008

**DETERMINATION OF THE ANTHRAQUINONE PAPER/PAPERBOARD
SIMULATED SWEAT PARTITION COEFFICIENT FOR THE ESTIMATION
OF POTENTIAL DERMAL CONTACT EXPOSURE**

SPECIAL REPORT NO. 08-03
JULY 2008

ABSTRACT

In response to new requirements to assess wood pulp derived materials marketed in California that may contain anthraquinone, experiments were conducted to aid the determination of the potential of anthraquinone (if any) contained in such materials to result in dermal contact exposure. The model used to assess this potential exposure requires an anthraquinone sweat/paper (or paperboard) partition coefficient. The American Forest & Paper Association Chemical & Product Stewardship Group (C&PSG) requested that NCASI conduct studies to determine this partition coefficient. This report summarizes the results of that study.

After discussion, C&PSG approved a study design that utilized simulated sweat in contact with an unbleached kraft paper sample of known anthraquinone content for time intervals of 1, 2, 4, 6, 8, and 24 hours at a temperature of 25°C. The experiments were conducted by NCASI and the data were modeled using Fick's Second Law. An estimate of the equilibrium partition coefficient was derived as 0.000928 ± 0.000140 (95% C.I.).

KEYWORDS

anthraquinone, equilibrium partition coefficient, dermal contact substance, exposure assessment, paper, paperboard, Proposition 65

RELATED NCASI PUBLICATIONS

Technical Bulletin No. 953 (July 2008). *NCASI Method AQ-S108.01: Anthraquinone in uncoated papers and market pulps by Soxhlet extraction and GC/MS.*

Special Report No. 08-02 (July 2008). *The potential for migration of anthraquinone from unbleached linerboard.*

Technical Bulletin No. 831 (August 2001). *Technical support document for assessing the safety of a food contact substance.*

Technical Bulletin No. 824 (May 2001). *Evaluation of food simulating liquids for paperboard food packaging.*

CONTENTS

1.0	INTRODUCTION	1
2.0	EXPERIMENTAL.....	1
2.1	Exposure Apparatus and Conditions.....	1
2.2	Analysis of Exposure Specimens and Exposed Simulated Sweat	2
3.0	RESULTS.....	3
3.1	Anthraquinone in Study Specimen of Unbleached Kraft Paper	3
3.2	Anthraquinone in Simulated Sweat	3
3.3	Anthraquinone Partitioning.....	5
	REFERENCES.....	8
	APPENDICES	
A	Transient Diffusion in a Semi-Infinite Medium	A1

TABLES

Table 2.1 MS Ions Used in Both Full Scan and SIM GC/MS Analysis 2

Table 3.1 Single Laboratory Method Performance of NCASI Method AQ-S108.01 in Samples 4

Table 3.2 Summary of Results from Analysis of Exposed Simulated Sweat 5

Table 3.3 Summary of Partition Coefficients 6

FIGURES

Figure 3.1 Partition Coefficient vs. Exposure Time..... 7

DETERMINATION OF THE ANTHRAQUINONE PAPER/PAPERBOARD SIMULATED SWEAT PARTITION COEFFICIENT FOR THE ESTIMATION OF POTENTIAL DERMAL CONTACT EXPOSURE

1.0 INTRODUCTION

Effective September 28, 2007, California's Office of Environmental Health Hazard Assessment (OEHHA) added anthraquinone to the Proposition 65 list of chemicals known to the state to cause cancer. Because of this listing, member companies will have to assess the need to label products for sale or use in California that contain anthraquinone and take appropriate action by September 2008.

Exposure assessment models have been developed for three potential modes of exposure: inhalation, ingestion, and dermal contact. The model for dermal contact involves consideration of the sequential transfer of the substance from paper/paperboard into sweat, from sweat into sebum, and finally from sebum into skin. One critical parameter in this model is the partition coefficient between sweat and paper/paperboard. However, a measured value for the anthraquinone partition coefficient does not exist.

To address this data gap, the American Forest & Paper Association Chemical & Product Stewardship Group (AF&PA C&PSG) requested that NCASI conduct studies to determine the anthraquinone partition coefficient for simulated sweat and paper/paperboard. After discussion, C&PSG approved a study design that utilized simulated sweat in contact with an unbleached kraft paper sample of known anthraquinone content for time intervals of 1, 2, 4, 6, 8, and 24 hours at a temperature of 25°C. This report summarizes the results of that study.

2.0 EXPERIMENTAL

2.1 Exposure Apparatus and Conditions

2.1.1 *Description of Exposure Apparatus*

The partition coefficient exposure apparatus was conceptually similar to the design used by FDA (Snyder and Breder 1985) and A.D. Little, Inc. (ADL 1983). For each exposure a total of 26 approximately 2 in. diameter paper specimens were suspended on three stainless steel rods (L shaped, with long sides approximately 16 cm and short sides approximately 1.5 cm) and separated by 5 mm glass beads. Holes were cut in the paper 13/16" from the center and spaced 120° from each other to provide stability and to assure that the paper disks did not contact each other when wet. Each stack was placed into a 500 mL tall I-Chem Series 300 glass jar with a Teflon™ lid.

2.1.2 *Exposure Conditions*

Simulated sweat was prepared as described in EN 1811 (CEN 2008) with the exception of using aerated water. The simulated sweat was pre-equilibrated at the required study exposure temperature of 25° C. At the beginning of each exposure, 450 mL was measured and added to each exposure apparatus. The jars were placed in a reciprocating shaker, temperature controlled water bath for the time intervals required in the study design. Upon completion of the exposure, the simulated sweat was vacuum filtered through Whatman 934-AH glass fiber filters, and the filtrates were transferred into amber bottles and refrigerated until extracted.

2.2 Analysis of Exposure Specimens and Exposed Simulated Sweat

2.2.1 Unbleached Kraft Paper Study Specimen Characterization and Analysis

The exposure specimen used in these experiments was unbleached (brown) kraft paper with a basis weight of nominally 60 lb (60 lb/3000 ft²). Prior to performing the partitioning experiments, this paper was analyzed to determine anthraquinone using NCASI Method AQ-S108.01 (NCASI forthcoming), which was developed by NCASI in late 2007 and early 2008 specifically for analyses of pulps and uncoated papers. This method is currently in the queue for publication in NCASI's *Methods Manual*, and can be obtained by contacting NCASI.

Briefly, Method AQ-S108.01 calls for samples of pulp or uncoated papers to be cut into nominal 1 x 1 cm squares, 5 g of which is then extracted using 300 mL of ethanol in a Soxhlet extractor for a minimum of 12 hours. Following extraction, the ethanol is concentrated to approximately 1 mL of hexane, which is then subjected to silica gel cleanup. The analytical cut from this column cleanup is concentrated to 1 mL hexane and analyzed by full scan gas chromatography/mass spectrometry (GC/MS). Prior to extraction, samples are fortified with 5 µg 2-chloroanthraquinone as a recovery surrogate. In addition, immediately preceding silica gel cleanup, the hexane extracts are fortified with 25 µg d₁₀-benzophenone as a cleanup recovery surrogate. Assessing recoveries of these two surrogates allows for differentiating problems originating with the extraction and initial concentration steps vs. subsequent cleanup and final concentration.

Immediately prior to GC/MS analysis, final extracts were fortified with 25 µg of d₁₀-anthracene as an injection internal standard, and 1 µL of the final extract was injected onto the GC/MS. Chromatographic separations were made on a Phenomenex ZB-5 GC column (30 m, 0.25 mm id, 0.25 µm, Phenomenex #7HG-G002-11) with the injection port at 270°C and the MS interface at 290°C. The GC temperature program was 50°C (1 min hold); to 160°C at 20°C/min; to 250°C at 8°C/min; and to 300°C at 25°C/min with a final 4 min hold (helium carrier, 7 psi constant head pressure, 35 cm/sec at 50°C). All internal standard quantifications were made using the average relative response factor (RRf) developed from a seven point calibration spanning the range from 0.2 µg/g to 20 µg/g (ppm, assuming an exact 5 g sample), and all analyses were performed by full scan GC/MS. Table 2.1 lists the relevant mass fragments (ions) and ion abundance criteria.

Table 2.1 MS Ions Used in Both Full Scan and SIM GC/MS Analysis

Compound	Quantification Ion (m/z)	Qualitative Ions (m/z)	Ion Abundance Criteria ^a
Anthraquinone	208	180, 152	±20%
2-Chloroanthraquinone	242	241, 151	±20%
d ₁₀ -Benzophenone	110	82, 192	±20%
d ₁₀ -Anthracene	188	94, 80	±20%

^a criteria relative to mean abundance ratios established from analysis of standards under the same MS tune (e.g., if mean from analysis of standards is 50% abundance, acceptance window is 30 to 70%)

2.2.2 *Simulated Sweat Analysis*

The filtered exposed simulated sweat samples were brought to room temperature and the volume recovered from the exposure apparatus was measured and recorded. This was always less than was added at the beginning of the exposure due to absorption of the sweat simulant by the study specimen and sweat simulant that tended to stick to the beads and stainless steel rod. The simulant was transferred to a separatory funnel and fortified with 12.2 μg 2-chloroanthraquinone as a recovery surrogate.

The simulant was extracted with 1 x 150 mL and 3 x 100 mL portions of DCM. In the first batch of samples a fifth partition was performed, concentrated, and analyzed separately. No detectable anthraquinone was found in any of these fifth partition extracts, providing evidence supporting quantitative extraction efficiency. It is quite possible that the fourth extraction may have been unnecessary, and future work should explore this possibility. The combined first four extracts were concentrated in a Kuderna Danish apparatus and the solvent was converted to 1 mL of hexane. Immediately prior to GC/MS analysis, final extracts were fortified with 24.8 μg of d_{10} -anthracene as an injection internal standard, and 1 μL of the final extract was injected onto the GC/MS. The GC/MS conditions were exactly as described in NCASI Method AQ-S108.01 (NCASI forthcoming).

3.0 RESULTS

3.1 Anthraquinone in Study Specimen of Unbleached Kraft Paper

Table 3.1 is taken directly from Method AQ-S108.01 (where it is Table 3), and summarizes surrogate recovery and matrix spike recovery results from analysis of multiple pulp and paper samples, including results obtained from analysis of the contact specimen. Duplicate analyses of the partition specimen gave 36.7 and 39.3 $\mu\text{g/g}$, respectively (giving a relative percent difference of 6.7%). A single matrix spike experiment performed at 195 $\mu\text{g/g}$ gave 95.2% recovery, while the mean ($n=3$) recoveries of the recovery (2-chloroanthraquinone) and cleanup (d_{10} -benzophenone) surrogates were $94.8 \pm 3.61\%$ and $97.4 \pm 0.66\%$, respectively. Matrix spike and surrogate recovery results from multiple analyses of this sample support the mean sample result of 38.0 $\mu\text{g/g}$ (on an as-received basis) as being both precise and unbiased.

Based on in-house measurements at the time of analysis, the contact specimen had a density of 0.068 g per square inch, corresponding to a calculated basis weight of 64.5 lb (lb per 3000 ft^2).

3.2 Anthraquinone in Simulated Sweat

Table 3.2 summarizes results of analyses of the exposed simulated sweat. Because the approximate 2" diameter study specimens had to be cut out by scissors, the exact weight of sample exposed in each jar was slightly different, so these weights are included in the table. In addition, the volume of simulated sweat recovered from each jar varied slightly, so these volumes are also recorded in the table. The use of these data is discussed in Section 3.3.

Analysis of the sweat simulants was accomplished in two batches. Method blanks (aliquots of simulated sweat) were analyzed with each batch and no detectable anthraquinone was found. Matrix spikes were also conducted in each batch using a dedicated, four hour exposed sample. The spike levels were 91 $\mu\text{g/L}$ and 114 $\mu\text{g/L}$ and recoveries were 97.9 % and 96.0 %, respectively. Taken together, the lack of any detectable anthraquinone in a fifth extraction, high surrogate recoveries, and high matrix spike recoveries indicate that measurements were quantitative.

Table 3.1 Single Laboratory Method Performance of NCASI Method AQ-S108.01 in Samples

Compound	Concentration (ppm) ^a	MS/MSD Percent Recovery			MS/MSD or Native Duplicate Precision				
		Mean	SD	n	Acceptance Window ^b	Mean RPD (%)	SD (%)	n	Acceptance Limit ^c
Anthraquinone	1	96.1	2.61	8	88.2 - 103.9	2.64	1.79	7	8.00
	4					0.02 - 4.99 ^e		2	
	16	95.3 - 98.7 ^d		2					
	40					3.60 - 6.73 ^e		2	
2-Chloroanthraquinone	1	95.2	4.75	36	81.0 - 109.7				
d ₁₀ -Benzophenone	5	97.7	3.68	42	86.7 - 108.8				

^a (nominal) equivalent sample concentrations assuming 5.0 g sample size

^b calculated as [mean ± (3 x SD)]

^c calculated as [mean + (3 x SD)]

^d one spike on each of two separate samples; range is given, no SD or acceptance window calculated

^e one pair of native duplicates for each of two separate samples; no range is given, no SD or acceptance window calculated

Table 3.2 Summary of Results from Analysis of Exposed Simulated Sweat

Exposure Time (hours)	Paper Weight (g)	Solvent Recovered (mL)	Simulated Sweat Concentration ($\mu\text{g/L}$)	Relative Percent Difference of Duplicates (%)	Surrogate Recovery (%)
1	5.9807	435	17.68		92.2
1	5.8577	435	17.31	2.1	100
2	6.003	435	23.22		98.4
2	5.8209	435	20.11	14.3	91
4	5.9838	435	25.98		98.2
4	6.0003	430	20.14	25.3	95.4
6	6.0382	425	27.76		95.9
6	6.0243	435	26.90	3.2	97.9
8	5.8675	430	28.37		99.3
8	5.571	430	23.86	17.3	103.8
24	6.0452	435	30.11		99.5
24	6.0108	435	30.57	1.5	98.6

3.3 Anthraquinone Partitioning

A sweat-paper partition coefficient is obtained by dividing the concentration of anthraquinone in the simulated sweat by the concentration of anthraquinone remaining in the paper at termination of the exposure/contact time. However, on completion of an experiment the paper was saturated with simulated sweat, making it impossible to measure the fraction of the original anthraquinone remaining in the paper; i.e., any measurement in the post-partitioning paper would be biased high due to the anthraquinone in the simulated sweat retained by the paper. Thus, the amount of anthraquinone remaining in the paper was obtained by subtracting the mass of anthraquinone in the total (initial) volume of simulated sweat from the initial total mass of anthraquinone in the paper discs. This calculation assumes that the concentration of anthraquinone in the volume of simulated sweat recovered and extracted is an accurate measure of the concentration of anthraquinone in the simulated sweat retained by the paper. Although the magnitudes of the corrections were small (on the order of 0.1 to 0.11%), all partition coefficients were calculated as described herein. Arithmetically, the calculation of each partition coefficient used Equations 1 and 2 (in sequence):

$$AQ_{\text{simulant}} = C_{\text{aliquot}} \times .450 \text{ L} \quad (\text{Eq. 1})$$

where: AQ_{simulant} = total (μg) anthraquinone in the simulated sweat

C_{aliquot} = concentration ($\mu\text{g/L}$) measured in the aliquot of stimulant recovered from the exposure apparatus

The final concentration in the paper was then calculated according to Equation 2.

$$CP_{\text{final}} = \frac{(SW \times CP_{\text{initial}}) - AQ_{\text{stimulant}}}{SW} \quad (\text{Eq. 2})$$

where: CP_{initial} = concentration of anthraquinone ($\mu\text{g}/\text{kg}$) measured in the paper before exposure
 SW = sample weight (g) exposed to stimulant
 CP_{final} = corrected concentration of anthraquinone at the time exposure was terminated

Although these corrections were small (on the order of 0.1 to 0.11%), the adjustment was deemed appropriate prior to calculation of the partition coefficient.

After making the correction to the concentration of anthraquinone in the paper after exposure, the partition coefficient can be calculated by dividing the concentration measured in the simulated sweat by the corrected final concentration in the paper. The resulting partition coefficients are summarized in Table 3.3.

Table 3.3 Summary of Partition Coefficients

Exposure Time (hours)	Partition Coefficient
1	4.82E-04
1	4.72E-04
2	6.39E-04
2	5.51E-04
4	7.19E-04
4	5.51E-04
6	7.70E-04
6	7.46E-04
8	7.90E-04
8	6.60E-04
24	8.41E-04
24	8.54E-04

The partition coefficient was plotted vs. the exposure time as shown in Figure 3.1. It is clear that the partition coefficient was still increasing at 24 hours and additional longer-term exposures would be necessary to more reliably define the equilibrium partition coefficient.

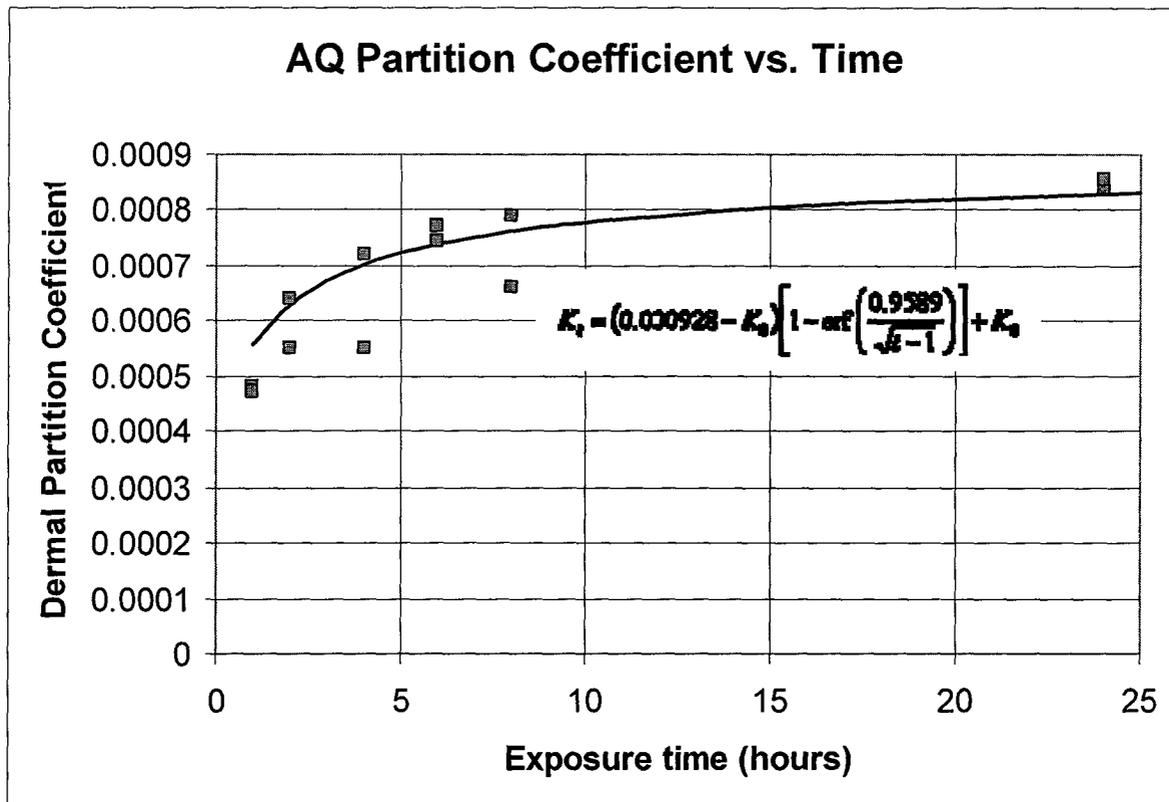


Figure 3.1 Partition Coefficient vs. Exposure Time

The migration of anthraquinone from paper into simulated sweat is not unlike the migration of a substance into food simulating liquids used for food packaging exposure assessments. Those types of studies typically model the migration behavior in terms of Fick's Second Law (ADL 1983; NCASI 2001). Thus (as discussed in NCASI 2001), partition coefficient data were plotted against the square root of the exposure time, resulting in a linear relationship with a non-zero slope as would be expected for migration conforming to Fick's Second Law. However, as observed by NCASI (2001), there was a significant non-zero intercept that the author interpreted as evidence of a potentially significant instantaneous migration followed by a slower migration that behaved according to Fick's Second Law.

Because the objective of the current work is to estimate the equilibrium partition coefficient, the data can be modeled using Equation 3, which was derived from Fick's Law (derivation shown in Appendix A). Figure 3.1 shows experimental data along with the Fick's Law model fit to those data. Using this approach, the equilibrium partition coefficient is estimated to be 0.000928 ± 0.000140 (95% C. I.).

$$K_t = (K_e - K_0) \left[1 - \operatorname{erf} \left(\frac{a}{\sqrt{t-1}} \right) \right] + K_0 \quad (\text{Eq. 3})$$

where: K_t = partition coefficient at time t
 K_e = equilibrium partition coefficient
 K_0 = initial partition coefficient
 a = parameter related to diffusional distance
 t = time

REFERENCES

- A.D. Little, Inc. (ADL). 1983. *ADL final summary report: A study of indirect food additive migration*. FDA Contract 223-77-236. Aurther D. Little, Inc.
- Comité Européen de Normalisation (CEN). 2008. *Reference test method for release of nickel from products intended to come into direct and prolonged contact with the skin*. European Standard EN 1811+A1. Brussels: Comité Européen de Normalisation (European Committee for Standardization).
- National Council for Air and Stream Improvement, Inc. (NCASI). 2001. *Evaluation of food simulating liquids for paperboard food packaging*. Technical Bulletin No. 824. Research Triangle Park, NC: National Council for Air and Stream Improvement, Inc.
- _____. 2008. *NCASI Method AQ-S108.01: Anthraquinone in uncoated papers and market pulps by Soxhlet extraction and GC/MS*. Technical Bulletin No. 953. Research Triangle Park, NC: National Council for Air and Stream Improvement, Inc.
- Snyder, R.C., and Breder, C.V. 1985. New FDA migration cell used to study migration of styrene from polystyrene into various solvents. *Journal of the Association of Official Analytical Chemists* 68(4):770-775.

APPENDIX A

TRANSIENT DIFFUSION IN A SEMI-INFINITE MEDIUM

The one dimensional transient diffusion equation (Equation A1) assumes that there is no bulk motion contribution and that no chemical reaction is taking place.

$$\frac{\partial K}{\partial t} = D_{AB} \frac{\partial^2 K}{\partial x^2} \quad (\text{Eq. A1})$$

Here K represents concentration, t is time, D_{AB} is the diffusion coefficient of anthraquinone (A) into the simulated sweat (B), x represents position.

The initial and boundary conditions for one directional mass transfer into a semi-infinite stationary medium with a fixed surface concentration:

$$K = K_0 \quad \text{at } t = 0 \quad \text{for all } x$$

$$K = K_e \quad \text{at } z = 0 \quad \text{for all } t$$

$$K = K_0 \quad \text{at } z \rightarrow \infty \quad \text{for all } t$$

The solution to the differential equation with the boundary conditions can be accomplished by Laplace transformation and separation of variables and is described in Crank (1975) and Carslaw and Jaeger (1959). The diffusion model is fit to time series data starting at $t = 1$ hour and extending to $t = 24$ hours so the starting time is taken as $(t-1)$. The initial partition coefficient at $(t-1) = 0$, the equilibrium partition coefficient, and the parameter a are estimated from experimental partition coefficient data.

$$K_t = (K_e - K_0) \left[1 - \operatorname{erf} \left(\frac{x}{2\sqrt{D_{AB}(t-1)}} \right) \right] + K_0 \quad (\text{Eq. A2})$$

Grouping $\left(\frac{x}{2\sqrt{D_{AB}}} \right)$ and labeling as a

$$K_t = (K_e - K_0) \left[1 - \operatorname{erf} \left(\frac{a}{\sqrt{(t-1)}} \right) \right] + K_0 \quad (\text{Eq. A3})$$

where: K_t = partition coefficient at time t
 K_e = equilibrium partition coefficient
 K_0 = initial partition coefficient
 a = parameter related to diffusional distance
 t = time

Solutions to Equation A1 with other boundary conditions are available and may be more appropriate to the experimental system than the ones used, as Equation A3 was a first attempt at modeling the partition coefficient data.

REFERENCES

Carslaw, H.S., and Jaeger, J.C. 1959. *Conduction of heat in solids*. Oxford: Oxford University Press.

Crank, J. 1975. *Mathematics of diffusion*. Oxford: Oxford University Press.