May 29, 2019

Mr. Julian Leichty
Office of Environmental Health Hazard Assessment
Proposition 65 Implementation Program
P.O. Box 4010, MS-12B
Sacramento, California 95812-4010

Via electronic submission

Re: Request for Relevant Information on Acetaminophen

Dear Mr. Leichty,

This information is submitted on behalf of the Consumer Healthcare Products Association ("CHPA") in response to the March 15, 2019 Office of Environmental Health Hazard Assessment's ("OEHHA") notice (OAL notice number Z2019-0305-03; notice register number 2019, 11-Z) requesting information that may be relevant to the carcinogenicity of acetaminophen. CHPA, founded in 1881, is a member-based association representing the leading manufacturers and distributors of non-prescription (or over-the-counter; OTC) medicines and dietary supplements.

CHPA appreciates the opportunity to provide information relevant to the assessment of the potential carcinogenicity of acetaminophen. It is our understanding that the information received during this data call-in period will be reviewed and considered by OEHHA as it prepares the cancer hazard identification materials (HIM) on acetaminophen.

Past Submission

On September 20, 2011, CHPA submitted comments to OEHHA related to the prioritization of acetaminophen. That submission referenced data available through the first six months of 2011 relevant to the consideration of the carcinogenicity of acetaminophen, including human epidemiologic data, animal data, genotoxicity and an authoritative body review of acetaminophen. We assume OEHHA will consider this information as the HIM are prepared and are attaching it to this submission. This letter contains updated information.

Reviews and Evaluations by Regulatory Organizations

In November 2010, the Food and Drug Administration (FDA) reviewed and approved a New Drug Application for OFIRMEV™, a prescription only, intravenous formulation of acetaminophen. This included an extensive review of available data including animal carcinogenicity studies that indicated no (based on mice, male rats) or equivocal (based on female rats) evidence of carcinogenic activity. This finding was supported by the FDA's Executive Carcinogenicity Assessment Committee. Conclusions regarding genotoxicity data supported previously reported findings and were mixed and are consistent with the IARC 2011 review. The current approved labeling for OFIRMEV™ can be found here[1].

[1] https://www.accessdata.fda.gov/drugsatfda_docs/label/2018/022450s011lbl.pdf. Accessed 5/28/19.

Epidemiologic Studies

We consider the review of epidemiologic data on acetaminophen usage and the occurrence of cancer published by Weiss (2016)[2] to be a useful review of much of the epidemiologic data, and we request that this paper be included in the HIM. Weiss' work included studies published through the end of 2015. In order to update Weiss' work, a literature search was conducted for epidemiologic studies (e.g., observational database, retrospective, prospective, case-control, cohort, meta-analysis, systematic review) published from 1 Jan 2014 onward. Twenty-three publications were identified and will be discussed below with the sequence of forms of cancer matching the sequence in Weiss 2016.

Weiss' review of 63 published studies assessed occasional through daily use of acetaminophen at various doses and for various durations (e.g., weeks, months). Overall, the findings from Weiss 2016 and the current literature do not support a causal relationship between acetaminophen use and cancer risk. It is important that the HIM thoroughly describe the strengths and limitations of each epidemiologic study.

Interpretation of Study Results

Because not every member of the Carcinogen Identification Committee is an expert in epidemiology and because there is an unusually large number of epidemiologic studies of acetaminophen, OEHHA should consider including certain matters of terminology and methodology in the HIM. The terminology and methodology noted below are offered with that consideration in mind, recognizing that the Carcinogen Identification Committee has a number of experts familiar with these considerations.

Observed Associations

In the context of the epidemiologic studies reviewed, the terms odds-ratio (OR), hazard ratio (HR) and relative risk (RR) are all measures of estimated risk (of cancer occurrence) in those exposed versus those not exposed to acetaminophen. ORs are used in case-control studies, while cohort studies can estimate RRs or HRs, depending on the choice of statistical model. In some situations, cohort studies estimate ORs, again depending on the choice of the statistical model. RRs are the ratio of measures of absolute risks in those exposed versus unexposed. RRs are often used in describing the results of meta-analyses or as a general term including studies that use any of these measures. For purposes of this document, the three measures can be viewed as having essentially the same interpretation. [3]

Certain considerations are taken into account in the determination of whether an observed association is causal. The strength of the association is one consideration (alongside consistency, specificity,

[2] Weiss NS. Use of acetaminophen in relation to the occurrence of cancer: a review of epidemiologic studies. Cancer Causes Control. 2016;27(12):1411-1418.

[3] When equal to 1.0, all three measures (OR, HR and RR) mean no difference in risk between exposure groups. A value greater than 1.0 indicates higher risk in the exposed versus the unexposed and a value less than 1.0 means lower risk in the exposed versus the unexposed. Each ratio is an estimate typically accompanied by a confidence interval (CI) that puts a bound on the values that are statistically compatible with the true value, but this focuses on statistical variability and not bias. (A discussion of bias is presented below). If the confidence interval excludes 1.0 then it is considered statistically significant.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

temporality, biological gradient, plausibility, coherence, experiment, and analogy), and the greater the strength, the larger the association as measured by OR, HR or RR. A weak association is typically an OR, HR or RR ≤ 2. In discussing strength of association, Hill (1965)[4] referred to the large mortality rate of scrotal cancer in chimney sweeps, which was a strong association, at 200 times the rate for other workers, and to a lung cancer mortality rate in smokers that was nine to ten times the rate in nonsmokers. A weak association was ≤ 2, as described by Hill, pointing out that in such cases it would be easy to think of other factors that could be the true underlying cause. Hill classified a weak association as one that plausibly could be explained by other factors, if they were not taken into account.

Another element of Hill's considerations for causal association is evidence of a consistent dose-response association. Hill referred to the fact that the RR of lung cancer mortality was 20 to 30 in heavy smokers compared to nonsmokers.[5] Therefore, consistency in the analyses by duration or frequency of use is important.

Bias and Confounding

Epidemiologic studies have strengths and limitations, including channeling bias, recall bias, and other biases described below. These limitations should be carefully considered and described in the HIM.

Channeling Bias

Channeling bias is defined as the use of one drug to a greater extent compared to another in certain patient populations, in a way that affects the relative risk. It is well known that acetaminophen represents an important pain relief alternative to NSAIDs for consumers with other existing conditions such as asthma, CV, GI, renal disease and other conditions. Channeling could occur, for example, when patients with evidence of renal insufficiency are recommended to take acetaminophen, if an analgesic is needed, for pain-relief or fever based on limited therapies that are considered safe in this population. As a consequence, there is an increased number of patients in this population taking acetaminophen versus other drugs, and these patients have a greater risk for progression to renal failure because of their underlying disease, not due to exposure to acetaminophen. In Weinstein 2017[6], evidence of channeling bias in the prescribing of acetaminophen versus ibuprofen in a UK electronic health records database was reported. Those with a history of renal disease at the time of prescription were more likely to receive a prescription for acetaminophen (7.4%) than a prescription for ibuprofen (2.8%). This study assessed how well other studies in the literature controlled for channeling bias. Typically, studies account for channeling or confounding by controlling for a selected list of key covariates. Weinstein (2017)[7] shows that accounting for a few key covariates is not sufficient to completely eliminate bias but that applying methods that incorporate very large numbers of covariates (hundreds) will better remove residual bias. This example showing the disproportionate use of

[4] Hill AB. The environment and disease: Association or causation? Proc R Soc Med. 1965;58:295-300.
[5] Ibid.
[6] Weinstein RB, Ryan P, Berlin JA, et al. Channeling in the use of nonprescription paracetamol and ibuprofen in an electronic medical records database: evidence and implications. Drug Saf. 2017; 40:127901292. doi 10.1007/s40264-017-0581-7.
[7] Ibid.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

acetaminophen in patients with renal disease highlights the impact channeling bias can have on studies of the associations of acetaminophen with cancer.

Bias Related to Treatment for Early Symptoms of Disease

Another form of bias relates to treatment of early signs and symptoms of disease prior to diagnosis (protopathic bias). Many cancers can be painful, and some are associated with fever. Signs and symptoms may be noted long before diagnosis. Several studies mention, and some perform sensitivity analyses to try to adjust for, this form of bias. However, it is not clear whether and to what extent the adjustment has been adequate.

Bias Related to Capture of Over-the-Counter (OTC) and Prescription Use of Acetaminophen

Because acetaminophen is available OTC in many countries, use is not completely captured by prescriptions or dispensing in administrative or electronic medical records (EMR) databases. Therefore, studies that rely solely on such databases likely are not able to capture all use. This type of misclassification of exposure is known to bias results towards showing no association. Weiss (2016)[8] also noted this type of bias.

Recall Bias

Some studies capture exposure through the use of surveys, which rely on patients to accurately remember and report their use. Case-control studies, which often depend on cases (with the outcome of interest) and controls (without the outcome) to recall acetaminophen use, may be subject to recall bias. This bias is caused by patients over-reporting use after having been diagnosed with the condition and controls potentially under-reporting (or not remembering) use. This would lead to an artificially inflated effect estimate. Since cohort studies collect exposure data prior to any diagnosis, they are not subject to this type of bias.

In addition, surveys can ask about medication use over the prior day, week, month, 6 months or years. Many surveys ask about the frequency of use or the amount taken. In a study of medication use recall (Heard 2016)[9], subjects recorded medication use in a diary for 30 days and then were asked to remember use in the last day and the last 30 days. Overall accuracy was 90% for use in the preceding day but was only 76% in the preceding 30 days. Because those same subjects had completed diaries, their recall may have been enhanced, compared with subjects from many other studies being asked to recall past exposures without having completed diaries. Exposure misclassification should be considered carefully in the interpretation of all study results.

[8] Weiss NS. Use of acetaminophen in relation to the occurrence of cancer: a review of epidemiologic studies. Cancer Causes Control. 2016;27(12):1411-1418.
[9] Heard K, Anderson VE, Dart RC, Green JL. Accuracy of the structured Medication History Assessment Tool (MedHAT) compared with recorded real-time medication use. Pharmacotherapy. 2016;36(5):496-504.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Observational Database Studies

It has been shown that observational methods have less predictive accuracy in discriminating between null effects and small positive effects (RR<2), as compared with discriminating large effects. (Ryan and Schuemie 2013)[10]

It has also been shown that different observational study designs, when applied to various observational databases, have different performance in their ability to discriminate positive and null effects. In particular, case-control designs have been shown to have lower performance than other designs, such as cohort and self-controlled methods. (Madigan et al 2013; Ryan et al 2013; Ryan et al 2012; Schuemie et al 2013)[11,12,13,14]

Further empirical research has shown that all observational database analyses can be susceptible to systematic error, which requires quantification and calibration to ensure that statistical measures, such as p-value and confidence intervals, are interpreted properly. (Schuemie et al 2014; Schuemie et al 2018)[15,16]

Several publications by members of a collaborative called Observational Health Data Sciences and Informatics (OHDSI), based on these findings, have recommended that observational studies should be accompanied with negative controls as a means of testing for systematic error. (Arnold et al 2016; Lipsitch et al 2010; Tchetgen Tchetgen 2014)[17,18,19]

Update to 2016 Review of Epidemiologic Studies on Use of Acetaminophen in Relation to the Occurrence of Cancer

Since the Weiss (2016)[20] review, outcomes assessed for potential association with acetaminophen were pancreatic cancer (1 paper), lung cancer (2 papers), breast cancer (3 papers), endometrial cancer (2 papers), ovarian cancer (4 papers), cervical cancer (2 papers), prostate cancer (1 paper), renal cell

[10] Ryan PB, Schuemie MJ. Evaluating performance of risk identification methods through a large-scale simulation of observational data. Drug Saf. 2013;36 Suppl 1:S171-80. doi: 10.1007/s40264-013-0110-2.
[11] Madigan D, Schuemie MJ, Ryan PB. Empirical performance of the case-control method: lessons for developing a risk identification and analysis system. Drug Saf. 2013;36 Suppl 1:S73-82. doi: 10.1007/s40264-013-0105-z.
[12] Ryan PB, Stang PE, Overhage JM, et al. A comparison of the empirical performance of methods for a risk identification system. Drug Saf. 2013;36 Suppl 1:S143-58. doi: 10.1007/s40264-013-0108-9.
[13] Ryan PB, Madigan D, Stang PE, et al. Empirical assessment of methods for risk identification in healthcare data: results from the experiments of the Observational Medical Outcomes Partnership. Stat Med. 2012;31(30):4401-15. doi: 10.1002/sim.5620. Epub 2012 Sep 27.
[14] Schuemie MJ, Gini R, Coloma PM, et al. Replication of the OMOP experiment in Europe: evaluating methods for risk identification in electronic health record databases. Drug Saf. 2013;36 Suppl 1:S159-69. doi: 10.1007/s40264-013-0109-8.
[15] Schuemie MJ, Ryan PB, DuMouchel W, et al. Interpreting observational studies: why empirical calibration is needed to correct p-values. Stat Med. 2014;33(2):209-18. doi: 10.1002/sim.5925.
[16] Schuemie MJ, Hripcsak G, Ryan PB, et al. Empirical confidence interval calibration for population-level effect estimation studies in observational healthcare data. Proc Natl Acad Sci U S A. 2018;115(11):2571-2577. doi: 10.1073/pnas.1708282114.
[17] Arnold BF, Ercumen A, Benjamin-Chung J, Colford JM Jr. Brief Report: Negative controls to detect selection bias and measurement bias in epidemiologic studies. Epidemiology. 2016;27(5):637-41. doi: 10.1097/EDE.0000000000000504.
[18] Lipsitch M, Tchetgen Tchetgen E, Cohen T. Negative controls: a tool for detecting confounding and bias in observational studies. Epidemiology. 2010;21(3):383-8. doi: 10.1097/EDE.0b013e3181d61eeb.
[19] Tchetgen Tchetgen E. The control outcome calibration approach for causal inference with unobserved confounding. Am J Epidemiol. 2014;179(5):633-40. doi: 10.1093/aje/kwt303.
[20] Weiss NS. Use of acetaminophen in relation to the occurrence of cancer: a review of epidemiologic studies. Cancer Causes Control. 2016;27(12):1411-1418.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

cancer (3 papers), skin cancer (1 paper), brain cancers, specifically glioma and meningioma (1 paper), lymphohematopoietic neoplasms, specifically pediatric leukemia (1 paper), and liver cancer (2 papers).

Pancreatic Cancer

Since the Weiss (2016)[21] review, the results from a single additional population-based case-control study by Kho (2016)[22] found no increased risk of pancreatic cancer in acetaminophen users.

Kho PF, Fawcett J, Fritschi L, Risch H, Webb PM, Whiteman DC, Neale RE. Nonsteroidal anti-inflammatory drugs, statins, and pancreatic cancer risk: a population-based case-control study. Cancer Causes & Control. 2016;27(12):1457-1464.

- In 2016, Kho evaluated the risk of pancreatic cancer among NSAID and statin users in the Queensland Pancreatic Cancer Study, a population-based, case-control study. Acetaminophen was included as a comparator. No significant increase in risk was found among acetaminophen users. The highest frequency of acetaminophen use evaluated was 4+ times/week and the odds-ratio (OR) was 0.92 (95% CI 0.62–1.36). The OR for occasional acetaminophen use, defined as less than 1/month, was 0.94 (95% CI 0.70–1.26).

Lung Cancer

One case-control study (Erickson 2018)[23] and one abstract (Bittoni 2017)[24] of a cohort study were published since the Weiss (2016)[25] review. The case-control study did not fully examine the risk for acetaminophen in the data available because the focus was on aspirin use and so should be interpreted cautiously. The cohort study reported no increased risk.

Erickson P, Gardner LD, Loffredo CA, George DM, Bowman ED, Deepak J, Mitchell K, Meaney CL, Langenberg P, Bernat DH, Amr S. Racial and ethnic differences in the relationship between aspirin use and non–small cell lung cancer risk and survival. Cancer Epidemiology and Prevention Biomarkers. 2018 ;27(12):1518-26.

- In a case-control study of non-small cell lung cancer, aspirin use and the effect of inflammatory risk factors by race were evaluated. While results for acetaminophen and other NSAIDs were reported, they were evaluated in a limited manner. Although the models pertaining to acetaminophen were adjusted for age, sex and smoking pack years, they were not adjusted for other available covariates including family history of lung cancer, BMI and use of NSAIDs (including aspirin), which represents a substantial limitation of the results for acetaminophen. A full assessment of acetaminophen should include a fully adjusted statistical model. The OR

[21] Ibid.
[22] Kho PF, Fawcett J, Fritschi L, et al. Nonsteroidal anti-inflammatory drugs, statins, and pancreatic cancer risk: a population-based case-control study. Cancer Causes Control. 2016;27(12):1457-1464.
[23] Erickson P, Gardner LD, Loffredo CA, et al. Racial and ethnic differences in the relationship between aspirin use and non–small cell lung cancer risk and survival. Cancer Epidemiol Biomarkers Prev. 2018 ;27(12):1518-26.
[24] Bittoni M, Carbone D, Harris R. Chronic inflammation, NSAIDs and the risk of lung cancer death. J Thoracic Oncol. 2017;12(1):S257.
[25] Weiss NS. Use of acetaminophen in relation to the occurrence of cancer: a review of epidemiologic studies. Cancer Causes Control. 2016;27(12):1411-1418.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

for ever-regular use of acetaminophen (at least one pill per week for two months in the past five years) was 1.51 (95% CI 1.13–2.03).

- The OR for African Americans (AA) was 2.13 (95% CI 1.23–3.67). However, among AAs, the trend did not increase for increasing dose, but may reflect an increase compared to no exposure (i.e., for < one tablet/week OR 2.40, 95% CI 0.80–7.20 and for ≥ one tablet/week OR 2.12, 95% CI 1.15 - 3.93, P (trend)=0.008).
- The OR for European Americans (EA) was 1.25 (95% CI 0.88–1.78). The trend was for higher ORs at higher dose and duration (e.g., for < one tablet/week OR 1.08, 95% CI 0.57–2.03 and for ≥ one tablet/week OR 1.35, 95% CI 0.90–2.03, P (trend)=0.15).
- It is important to note that the models for aspirin use were adjusted for the covariates age, sex, and smoking pack years and when additional variables (e.g., family history of lung cancer, BMI, and use of NSAIDs) were added, the ORs did change indicating the presence of confounding. This change in ORs and the fact that the acetaminophen use models were not fully adjusted suggests the possibility of residual confounding for acetaminophen, limiting the value of these findings.
- Overall, in the total population, the associations for acetaminophen were inconsistent and there was no dose-response relationship.

Bittoni M, Carbone D, Harris R. Chronic inflammation, NSAIDs and the risk of lung cancer death. Journal of Thoracic Oncology. 2017;12(1):S257.

- This poster abstract evaluated the cross-sectional National Health and Nutrition Examination Survey (NHANES) from 1988-1994 and matched respondents to the National Death Index through 2006. A subset of analgesic users was followed for up to 18 years for subsequent lung cancer death. The results for acetaminophen were not statistically significant. No hazard rates or confidence intervals were provided.

<u>Breast Cancer</u>

Since Weiss (2016)[26], there have been three additional studies evaluating acetaminophen and breast cancer. The results from the three studies show no association. [27,28,29]

de Pedro M, Baeza S, Escudero M, Dierssen-Sotos T, Gómez-Acebo I, Pollán M, Llorca J. Effect of COX-2 inhibitors and other non-steroidal inflammatory drugs on breast cancer risk: A meta-analysis. Breast Cancer Research and Treatment. 2015;149(2):525-536.

- This meta-analysis assessed the relationship between COX-2 inhibitors and other NSAIDs and breast cancer risk overall and by hormone receptor status. For acetaminophen, the eight case-control studies identified had a combined OR of 0.85 (95% CI 0.76-0.95) and three cohort studies identified had a combined RR of 0.92 (95% CI 0.85–1.00). Overall, for estrogen receptor positive breast cancer, the combined RR was 0.92 (95% CI 0.85–1.00). The consistent

[26] Weiss NS. Use of acetaminophen in relation to the occurrence of cancer: a review of epidemiologic studies. Cancer Causes Control. 2016;27(12):1411-1418.
[27] de Pedro M, Baeza S, Escudero M, et al. Effect of COX-2 inhibitors and other non-steroidal inflammatory drugs on breast cancer risk: A meta-analysis. Breast Cancer Res Treat. 2015;149(2):525-536.
[28] Clarke CA, Canchola AJ, Moy LM, et al. Regular and low-dose aspirin, other non-steroidal anti-inflammatory medications and prospective risk of HER2-defined breast cancer: The California Teachers Study. Breast Cancer Res. 2017;19(1):52.
[29] Bertrand KA, Bethea TN, Gerlovin H, et al. Aspirin use and estrogen receptor-negative breast cancer risk in African American women. Cancer Res. 2018;78(13). doi: http://dx.doi.org/10.1158/1538-7445.AM2018-5247.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

results for case-control and cohort studies and also for breast cancer overall and by receptor status strongly support no association.

Clarke CA, Canchola AJ, Moy LM, Neuhausen SL, Chung NT, Lacey JV Jr, Bernstein L. Regular and low-dose aspirin, other non-steroidal anti-inflammatory medications and prospective risk of HER2-defined breast cancer: The California Teachers Study. Breast Cancer Research. 2017;19(1):52.

- This prospective cohort study among participants in the California Teachers Study assessed the risk of breast cancer overall and by subtype according to aspirin and other NSAID use. Breast cancer subtypes were defined jointly by hormone receptor (estrogen and progesterone receptor) and human epidermal growth factor receptor 2 (HER2) expression. Clarke and associates also assessed the association between breast cancer and acetaminophen use of three or more tablets per week compared to no use in the prior 3 years. The results showed that acetaminophen was not associated with breast cancer overall (HR 1.00, 95% CI 0.87-1.15) or with the HR positive/HER2-negative subtype (HR 0.98, 95% CI 0.83-1.16).

Bertrand KA, Bethea TN, Gerlovin H, et al. Aspirin use and estrogen receptor-negative breast cancer risk in African American women. Cancer Research. 2018;78(13). doi: http://dx.doi.org/10.1158/1538-7445.AM2018-5247.

- Estrogen receptor-negative breast cancer was studied in the Black Women's Health Study (Bertrand 2018, conference abstract), a cohort study which started in 1995. Risk in acetaminophen use was evaluated and found not to have any significant association with estrogen receptor-negative or positive breast cancer (HRs not provided).

Endometrial Cancer

Two papers[30,31] have been published since Weiss (2016)[32] and found that regular use of acetaminophen is not associated with occurrence of endometrial cancer.

Ding YY, Yao P, Verma S, Han ZK, Hong T, Zhu YQ, Li Hx. Use of acetaminophen and risk of endometrial cancer: evidence from observational studies. Oncotarget. 2017;8(21):34643-34651.

- This systematic review and meta-analysis assessed the relationship between acetaminophen use and risk of endometrial cancer. Evaluation of three case-control studies and four cohort studies for ever- versus never-use of acetaminophen showed no statistically increased risk of endometrial cancer (RR 1.11, 95% CI 0.88-1.40 and RR 1.00, 95% CI 0.90-1.12, for the case-control and cohort studies respectively). Duration and frequency of use were also evaluated, and the study found no significant trends.

[30] Ding YY, Yao P, Verma S, et al. Use of acetaminophen and risk of endometrial cancer: evidence from observational studies. Oncotarget. 2017;8(21):34643-34651.
[31] Webb PM, Na R, Weiderpass E, et al. Use of aspirin, other nonsteroidal anti-inflammatory drugs and acetaminophen and risk of endometrial cancer: the Epidemiology of Endometrial Cancer Consortium. Ann Oncol. 2019;30(2):310-316.
[32] Weiss NS. Use of acetaminophen in relation to the occurrence of cancer: a review of epidemiologic studies. Cancer Causes Control. 2016;27(12):1411-1418.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Webb PM, Na R, Weiderpass E, Adami HO, Anderson KE, Bertrand KA, Botteri E, Brasky TM, Brinton LA, Chen C, Doherty JA, Lu L, McCann SE, Moysich KB, Olson S, Petruzella S, Palmer JR, Prizment AE, Schairer C, Setiawan VW, Spurdle AB, Trabert B, Wentzensen N, Wilkens L, Yang HP, Yu H, Risch HA, Jordan SJ. Use of aspirin, other nonsteroidal anti-inflammatory drugs and acetaminophen and risk of endometrial cancer: the Epidemiology of Endometrial Cancer Consortium. Annals of Oncology. 2019;30(2):310-316.

- Combined individual-level data from seven cohort studies and five case-control studies were evaluated using random-effects meta-analysis. The focus of the study was on aspirin and the association between BMI and endometrial cancer risk. The association with acetaminophen was also assessed, but data were only available for the cohort studies. No association was found for regular acetaminophen use (defined as use at least once a week but differed slightly across studies depending on how the question was worded) and endometrial cancer among normal weight (OR 1.10, 95% CI 0.91-1.33) or obese women (OR 1.04, 95% CI 0.86-1.24). The use of individual-level data in this analysis makes it a slightly stronger study than if it was based on the published data for each of the individual studies included. It allows for standardized variable definitions for each included study. For example, age could be defined similarly across studies.
- There is no overlap in the studies included in each of the pooled studies (Ding 2017; Webb 2019).

Ovarian Cancer

Two cohort studies (Barnard 2018 and Trabert 2019)[33,34] and two case-control studies (Peres 2016 and Hannibal 2018)[35,36] have been published since the Weiss 2016 review. The cohort studies evaluated duration, frequency and quantity of acetaminophen use and found no increased risk of ovarian cancer.

Barnard ME, Poole EM, Curhan GC, Eliassen AH, Rosner BA, Terry KL, Tworoger SS. Association of analgesic use with risk of ovarian cancer in the Nurses' Health Studies. JAMA Oncology. 2018;4(12):1675-82.

- The Nurses' Health Study (NHS) and NHS II prospectively followed 93,664 and 111,834 women, respectively, for up to 14 years and evaluated regular use, frequency and duration of use of acetaminophen, aspirin and non-aspirin NSAIDs during follow-up. The first two to four years after exposure was excluded to limit possible reverse causality. The authors note "Because pain and abdominal discomfort are commonly reported in the year leading up to ovarian cancer diagnosis, we incorporated a latency period of two to four years to limit the potential for reverse causation. For example, aspirin use in 1980 was used when evaluating ovarian cancer incidence from 1982 to 1984, aspirin use in 1982 was used when evaluating incidence from 1984 to 1986, and so on."
- The OR for current and past use of acetaminophen was 1.02 (95% CI 0.86-1.21) and 1.05 (95% CI 0.88-1.25), respectively. Duration and frequency of use showed no significant trends.

[33] Barnard ME, Poole EM, Curhan GC, et al. Association of analgesic use with risk of ovarian cancer in the Nurses' Health Studies. JAMA Oncol. 2018;4(12):1675-82.
[34] Trabert B, Poole EM, White E, et al. Analgesic use and ovarian cancer risk: An analysis in the Ovarian Cancer Cohort Consortium. J Natl Cancer Inst. 2019;111(2):137-145. doi: 10.1093/jnci/djy100.
[35] Peres LC, Camacho F, Abbott SE, et al. Analgesic medication use and risk of epithelial ovarian cancer in African American women. Br J Cancer. 2016;114(7):819-25.
[36] Hannibal CG, Dehlendorff C, Kjaer SK. Use of paracetamol, low-dose aspirin, or non-aspirin non-steroidal anti-inflammatory drugs and risk of ovarian borderline tumors in Denmark. Gynecol Oncol. 2018 ;151(3):513-8.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Trabert B, Poole EM, White E, Visvanathan K, Adami HO, Anderson GL, Brasky TM, Brinton LA, Fortner RT, Gaudet M, Hartge P. Analgesic use and ovarian cancer risk: An analysis in the Ovarian Cancer Cohort Consortium. Journal of the National Cancer Institute. 2019;111(2):137-145. doi: 10.1093/jnci/djy100.

- This study combined individual-level data from 13 prospective cohort studies from Europe and North America in the Ovarian Cancer Cohort Consortium to create a study population of 758,829 women. Frequent use of acetaminophen (defined as approximately four to five days/week for six or more months) was not associated with an increased risk of ovarian cancer (HR 1.05, 95% CI 0.88-1.24) and there was no increasing risk for increasing frequency or duration. However, daily use was of borderline significance (HR 1.28, 95% CI 1.00-1.65, p=0.05, n=71 ovarian cancer cases). Daily use was defined as daily use or nearly daily use (six to seven days per week or 28 days per month) for six months.
- The potential for reverse causation (i.e., treating early symptoms of the cancer) was evaluated by examining the associations of frequency of use with ovarian cancer cases occurring less than five years, five up to ten years, and ten years or more after the start of follow-up.
- Despite many strengths, the authors stated "Importantly, the increased risk with daily acetaminophen use observed in this study was based on a limited number of exposed cases and should be interpreted with caution."

Hannibal CG, Dehlendorff C, Kjaer SK. Use of paracetamol, low-dose aspirin, or non-aspirin non-steroidal anti-inflammatory drugs and risk of ovarian borderline tumors in Denmark. Gynecologic Oncology. 2018 ;151(3):513-8.

- A nested case-control study using Danish data evaluated ever use of acetaminophen for serous and mucinous borderline tumors and found no significant risk (OR 1.03, 95% CI 0.86–1.23 and OR 0.87, 95% CI 0.71–1.06, respectively). Use was defined as at least two prescriptions dispensed at least one year before the index date on separate dates. The study also evaluated recency, dose and time since last prescription redeemed and found no increased risk.

Peres LC, Camacho F, Abbott SE, Alberg AJ, Bandera EV, Barnholtz-Sloan J, Bondy M, Cote ML, Crankshaw S, Funkhouser E, Moorman PG, Peters ES, Schwartz AG, Terry P, Wang F, Schildkraut JM. Analgesic medication use and risk of epithelial ovarian cancer in African American women. British Journal of Cancer. 2016;114(7):819-25.

- This study evaluated risk of ovarian cancer among African American women and similarly found no increased risk for ever regular use (OR 0.89, 95% CI 0.49-1.62), defined as at least once a week or at least five days out of the month, at any point in their lifetime. The authors examined risk by duration and frequency of use and found no significant elevation for ovarian cancer.

Cervical Cancer

Weiss included no studies on cervical cancer in his review. One paper[37] and one abstract[38] were published after the Weiss review; the paper and abstract did not show increased risk of cervical cancer.

[37] Friel G, Liu CS, Kolomeyevskaya NV, et al. Aspirin and acetaminophen use and the risk of cervical cancer. J Low Genit Tract Dis. 2015;19(3):189-193. doi: http://dx.doi.org/10.1097/LGT.0000000000000104.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

All authors on the abstract were also authors on the paper, so it is not clear to what extent these studies included some of the same data.

Friel G, Liu CS, Kolomeyevskaya NV, Hampras SS, Kruszka B, Schmitt K, Cannioto RA, Lele SB, Odunsi KO, Moysich KB. Aspirin and acetaminophen use and the risk of cervical cancer. Journal of Lower Genital Tract Disease. 2015;19(3):189-193. doi: http://dx.doi.org/10.1097/LGT.0000000000000104.

- This hospital-based, case-control study conducted in the US evaluated aspirin and acetaminophen use separately and the risk of cervical cancer. Acetaminophen exposure (frequency and duration of use) was based on self-reported use of medications before the current illness. Regular use of acetaminophen, defined as at least once a week for at least six months, was also obtained. The OR for regular use was not statistically significant (OR 1.13, 95% CI 0.73–1.75). The trend tests were not statistically significant.

Liu C, Friel G, Kolomeyevskaya NV, Lele SB, Odunsi KO, Moysich K. Aspirin and acetaminophen decrease the risk of cervical cancer in long-term users. Gynecologic Oncology. 2014;133:42-43. doi: http://dx.doi.org/10.1016/j.ygyno.2014.03.123.

- This abstract largely included the same authors as the Friel paper. It was not clear to what extent these studies included some of the same or entirely different respondents. Frequent, long-term acetaminophen use, defined as ≥ seven tablets a week for ≥ five years, was not associated with an increased risk of cervical cancer (OR 0.19, 95% CI 0.05–0.82).

Prostate Cancer

One new study for acetaminophen and prostate cancer[39] was published since the Weiss review and reported no increased risk.

Wright JL, Chéry L, Holt S, Lin DW, Luedeke M, Rinckleb AE, Maier C, Stanford JL. Aspirin and NSAID use in association with molecular subtypes of prostate cancer defined by TMPRSS2:ERG fusion status. Prostate Cancer Prostatic Disease. 2016;19(1):53-6.

- This case-control study in Washington State assessed the association between molecular subtypes of prostate cancer defined by *TMPRSS2:ERG* fusion status and NSAID use. They also assessed the association with acetaminophen and found no association for fusion negative tumors (OR 1.12, 95% CI 0.58–2.18) or fusion positive tumors (OR 0.93, 09% CI 0.43–1.98) with current use, defined as at least once per week for three or more months at the time of the interview.

Renal Cell Carcinoma (RCC)

Weiss (2016)[40] reviewed the epidemiology studies in renal cell cancer and reported equivocal results. Acetaminophen is the primary metabolite of phenacetin which has an increased incidence of

[38] Liu C, Friel G, Kolomeyevskaya NV, et al. Aspirin and acetaminophen decrease the risk of cervical cancer in long-term users. Gynecol Oncol. 2014;133:42-43. doi: http://dx.doi.org/10.1016/j.ygyno.2014.03.123.
[39] Wright JL, Chéry L, Holt S, et al. Aspirin and NSAID use in association with molecular subtypes of prostate cancer defined by TMPRSS2:ERG fusion status. Prostate Cancer Prostatic Dis. 2016;19(1):53-6.
[40] Weiss NS. Use of acetaminophen in relation to the occurrence of cancer: a review of epidemiologic studies. Cancer Causes Control. 2016;27(12):1411-1418.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

transitional cell bladder and renal pelvis tumors. Weiss pointed out that although a similar relationship with acetaminophen would reasonably be expected, it is not what has been observed.

Since the Weiss review, two cohort studies, one case-control study and two meta analyses were presented in three publications (Choueiri 2014; Karami 2016; Preston 2016).[41,42,43] The case-control study (Karami 2016) did not show an increased risk for OTC and prescription use of acetaminophen. Of the two cohort studies presented, one study that had several important limitations found an increased risk (Karami 2016) and one study found no association (Preston 2016). In one of two meta-analyses (Choueiri 2014), once duplicate studies were dropped, no increased risk was seen (Karami 2016). The second meta-analysis, conducted in 2016 (Karami 2016), included newer studies and reported an increased risk; however, it did not systematically review the quality of the studies which may have been subject to channeling bias and which would have a substantial impact on the meta-analysis results. Overall, the weight of evidence does not support a relationship to renal cancer as discussed more fully below.

Choueiri TK, Je Y, Cho E. Analgesic use and the risk of kidney cancer: A meta-analysis of epidemiologic studies. International Journal of Cancer. 2014;134(2):384-96.

- This meta-analysis of analgesic use and risk of kidney cancer found no increased risk for acetaminophen in the three cohort studies (RR 1.19, 95% CI 0.93-1.52) but found a significant odds ratio for the 11 case-control studies of 1.30 (95% CI 1.13-1.49). However, as Karami noted (Karami 2016), Choueiri et. al. included a combined analysis of individual case-control studies (McCredie 1995)[44] in addition to the individual studies (McCredie 1993, Chow 1994, Mellemgaard 1994)[45,46,47] in their meta-analysis. Once the combined analysis was dropped, so the individual studies were not included twice, Karami found that the resulting OR for the case-control studies was no longer significantly increased: 1.22 (95% CI 0.99-1.49) (Karami 2016).

Karami S, Daughtery SE, Schwartz K, Davis FG, Ruterbusch JJ, Wacholder S, Graubard BI, Berndt SI, Hofmann JN, Purdue MP, Moore LE, Colt JS. Analgesic use and risk of renal cell carcinoma: A case-control, cohort and meta-analytic assessment. International Journal of Cancer. 2016;139(3):584-92.

In 2016, Karami evaluated analgesic use and renal cell cancer in three different study designs: 1) a case-control study which showed inconsistent results, 2) a cohort study which had limitations related to study design and data collection and 3) a meta-analysis in which the quality of studies or control for possible confounding factors was not assessed.

---

[41] Choueiri TK, Je Y, Cho E. Analgesic use and the risk of kidney cancer: A meta-analysis of epidemiologic studies. Int J Cancer. 2014;134(2):384-96.
[42] Karami S, Daughtery SE, Schwartz K, et al. Analgesic use and risk of renal cell carcinoma: A case-control, cohort and meta-analytic assessment. Int J Cancer. 2016;139(3):584-92.
[43] Preston MA, Zhang X, Graff RE, et al. Analgesic use and risk of renal cell cancer: Results from two prospective cohort studies. J Clin Oncol. 2016;34(2_suppl):588-588. (abstract)
[44] McCredie M, Pommer W, McLaughlin JK, et al. International renal-cell cancer study. II. Analgesics. Int J Cancer. 1995;60:345–349.
[45] McCredie M, Stewart JH, Day NE. Different roles for phenacetin and paracetamol in cancer of the kidney and renal pelvis. Int J Cancer. 1993; 53:245–249.
[46] Chow WH, McLaughlin JK, Linet MS, et al. Use of analgesics and risk of renal cell cancer. Int J Cancer. 1994; 59:467–470.
[47] Mellemgaard A, Niwa S, Mehl E, et al. Risk factors for renal cell carcinoma in Denmark: Role of medication and medical history. Int J Epidemiol. 1994; 23:923–930.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

- In the case-control study, the results were inconsistent. For OTC use-only, there was a significant increase in risk (OR 1.35 (95% CI 1.01-1.83)) and the trend for years of use was also significant (P=0.01). Use of prescription acetaminophen was not associated with renal cell cancer (OR 0.96 (95% CI 0.74-1.24)) and there was no trend with duration of use (P=0.67). For OTC or prescription use overall, there was no increased risk (OR 1.09, 95% CI 0.87-1.37). The test for trend was also not significant (P = 0.07). The authors reported that in their case-control study, the results may have been biased, since data on indication for analgesic use were not collected. Furthermore, the inconsistent findings for OTC versus prescription use, as well as the lack of a consistent dose-response trend, are important limitations to consider in the interpretation of the results. An in-home self-reported questionnaire was used to collect participants' use of acetaminophen and NSAIDs and required participants to know which drug(s) included acetaminophen. Combination products may have been included in this study.
- In the cohort study, a secondary use of data from a cancer screening trial (i.e., the US Prostate, Lung, Cervical, and Ovarian Cancer Screening Trial), the risk was significantly increased for regular use (at least once per week): HR 1.68 (95% CI 1.19-2.39). The p-value for trend in duration of use was 0.06, but the highest duration of ten or more years was not significantly increased (HR 1.08, 95% CI 0.55-2.10). For use less than ten years, the HR was 2.09 (95% CI 1.39-3.14). In this study, data collection for analgesics use began in 2006-2007 while the follow-up for cancer ended in December 2009. Although they restricted their analyses to cases with *more* than two years of follow-up after completing the analgesic data questionnaire, the relatively short follow-up leaves open the possibility of channeling bias if participants were being followed for renal disease. Additionally, according to the authors, it was possible that the cases may have been treating early symptoms of undiagnosed renal cell carcinoma. Other limitations that should temper the interpretation and that were mentioned by the authors were a reliance on self-report of acetaminophen use and a lack of dose data.
- For the meta-analysis, the combined estimate of four cohort studies was significant (RR 1.34, 95% CI 1.13–1.59) and the combined estimate of the nine case-control studies was of borderline significance (OR 1.20, 95% CI 1.01-1.42). The authors did not assess the quality of the studies included in the combined estimates. In particular, it would be important, in those studies, to control for possible confounding factors, i.e. comorbid renal disease and related conditions such as hypertension.

Preston MA, Zhang X, Graff RE, Sanchez A, Chang SL, Meir J, Stampfer T, Choueiri K, Wilson KM, Cho E. Analgesic use and risk of renal cell cancer: Results from two prospective cohort studies. Journal of Clinical Oncology. 2016;34(2_suppl):588-588. (abstract)

- This 2016 conference abstract reported a combined evaluation of analgesics and renal cell carcinoma from the Nurses' Health Study and the Health Professionals Follow-up Survey and found no significant association for acetaminophen use (RR 1.07, 95% CI: 0.80-1.44).

Skin Cancer

In Weiss (2016),[48] no association for acetaminophen and melanoma or other skin cancers were observed. No studies of acetaminophen and melanoma were published since Weiss 2016. One study

[48] Weiss NS. Use of acetaminophen in relation to the occurrence of cancer: a review of epidemiologic studies. Cancer Causes Control. 2016;27(12):1411-1418.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

of basal and squamous cell cancers was published since Weiss 2016 and found no association with acetaminophen use.[49]

Pandeya N, Olsen CM, Thompson BS, Dusingize JC, Neale RE, Green AC, Whiteman DC; QSkin Study. Aspirin and nonsteroidal anti-inflammatory drug use and keratinocyte cancers: a large population-based cohort study of skin cancer in Australia. British Journal of Dermatology. Mar 28, 2019. doi: 10.1111/bjd.17938.

A cohort study of 34,360 people in Queensland, Australia evaluated the association of aspirin or NSAIDs and the keratinocyte cancers, basal cell cancer (BCC) and squamous cell cancer (SCC) in a median of three years of follow-up. Acetaminophen was also evaluated as a 'control analgesic' to test a theory of inflammation and skin cancer and also to help evaluate residual confounding. The evaluation was carried out separately for high risk individuals who had prior skin biopsies or treatment for more than five actinic lesion skin excisions compared to low risk individuals.

In fully adjusted models, no association was found in participants at high risk for BCC for either frequent use (at least once a week) or infrequent use (less than once a week) compared to no use (HR 1.01, 95% CI 0.87 - 1.16 for infrequent, and HR 0.99, 95% CI 0.83 - 1.18 for frequent use). Furthermore, no association was found in participants at low risk for BCC for either frequent use or infrequent use compared to no use (HR 0.89, 95% CI 0.68 - 1.16 for infrequent and 0.83, 95% CI 0.59 - 1.17 for frequent use).

Similarly, for SCC in high risk individuals no association was found for either frequent use or infrequent use compared to no use (HR 0.92, 95% CI 0.75 – 1.13 for infrequent and HR 0.85, 95% CI 0.67 - 1.09 frequent use). For SCC in low risk individuals the results were similar (HR 0.96, 95% CI 0.58 – 1.58 for infrequent and HR 1.34, 95% CI 0.75 – 2.42 for frequent use).

<u>Brain Cancer – Glioma and meningioma</u>

One study of brain cancer was published since the Weiss review.[50] As reported below, this new study did not find any increased association between acetaminophen and gliomas and meningiomas.

Egan KM, Nabors LB, Thompson ZJ, Rozmeski CM, Anic GA, Olson JJ, LaRocca RV, Chowdhary SA, Forsyth PA, Thompson RC. Analgesic use and the risk of primary adult brain tumor. European Journal of Epidemiology. 2016;31(9):917-25.

- In a case-control study of glioma and meningioma, acetaminophen use for body pain or other indications (such as dental pain, menstrual symptoms and as a sleep aid) excluding headache was unrelated to risk of either brain cancer. The odds-ratios for body pain for glioma and meningioma, respectively were OR 0.94 (95% CI 0.64-1.39) and OR 1.27 (95% CI 0.72-2.26). For headache, the OR was 1.49 (95% CI 1.09, 2.04) for glioma and 2.35 (95% CI 1.53, 3.61) for meningioma.
- The authors concluded, "In this large case-control study, we observed evidence of confounding by indication for associations with analgesic use. For both brain tumors (glioma and meningioma), strong positive associations were observed in association with all classes of analgesics when taken for headache. In contrast, in glioma only, inverse associations were

[49] Pandeya N, Olsen CM, Thompson BS, et al. Aspirin and nonsteroidal anti-inflammatory drug use and keratinocyte cancers: a large population-based cohort study of skin cancer in Australia. Brit J Dermatol. Mar 28, 2019. doi: 10.1111/bjd.17938.

[50] Egan KM, Nabors LB, Thompson ZJ, et al. Analgesic use and the risk of primary adult brain tumor. Eur J Epidemiol. 2016;31(9):917-25.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

noted for aspirin taken for all other indications excluding headache. As headache is a major presenting symptom in brain tumor patients, noted positive associations with analgesic use for headache are likely to reflect reverse causation or 'protopathic bias'."

Lymphohematopoietic neoplasms

Since the Weiss review there have been no publications on lymphomas, plasma cell carcinoma or multiple myeloma. There was one publication on pediatric leukemia that evaluated maternal and early life exposures and found no significant increase (discussed further below).[51]

Weiss reviewed the lymphohematopoietic neoplasms and noted there have been only a small number of published studies examining a potential relationship between use of acetaminophen and plasma cell disorders, leukemia, and lymphoma. Weiss stated:

> "A larger number of studies have reported results for lymphoma, and several of these have observed increases in risk associated with [acetaminophen] use. However, just as often the results for lymphoma have not been positive. At present, while an increased risk of one or more lymphohematopoietic neoplasms among users of [acetaminophen] is a plausible hypothesis, it should not be regarded as anything more than this."[52]

Weiss discussed the possibility of pain as a source of protopathic bias in several studies for lymphomas, plasma cell carcinoma and leukemia. Many of the studies in the review conducted sensitivity analyses to test this hypothesis and observed a reduction in the RR, but not always an elimination of the risk, suggesting other factors underlying the association. This reflects the possibility of residual channeling bias due to emerging signs of underlying disease.

Signs and symptoms of cancer may arise years prior to diagnosis and may lead to the choice of acetaminophen for fever and pain relief throughout that time. For multiple myeloma (MM), bacterial and viral infections as well as autoimmune diseases have been recognized as potential early signs of risk (Brown 2008; Lindqvist 2011; Lindqvist 2017).[53,54,55] In a retrospective cohort study of US male veterans, relative risk of pneumonia was 1.82 (95% CI, 1.53-2.16) two to four years prior, 1.50 (95% CI, 1.25-1.80) five to nine years prior and 1.42 (95% CI, 1.23-1.63) ten or more years prior to diagnosis of MM compared to those without a MM diagnosis. (Brown 2008)[56] In a nested case-control study of men and women diagnosed with MM over a 40-year period in data from Sweden, a personal history of sinusitis, pneumonia, meningitis, septicemia, herpes zoster, infectious mononucleosis, and myocarditis was associated with a significantly increased risk of MM (OR 1.2; 95% CI, 1.1-1.3)

---

[51] Couto AC, Ferreira JD, Pombo-de-Oliveira MS, Koifman S. Pregnancy, maternal exposure to analgesic medicines, and leukemia in Brazilian children below 2 years of age. Eur J Cancer Prev
. 2015;24(3):245-252.

[52] Weiss NS. Use of acetaminophen in relation to the occurrence of cancer: a review of epidemiologic studies. Cancer Causes Control. 2016;27(12):1411-1418.

[53] Brown LM, Gridley G, Check D, Landgren O. Risk of multiple myeloma and monoclonal gammopathy of undetermined significance among white and black male United States veterans with prior autoimmune, infectious, inflammatory, allergic disorders. Blood. 2008;111:3388–94.

[54] Lindqvist EK, Goldin LR, Landgren O, et al. Personal and family history of immune-related conditions increase the risk of plasma cell disorders: a population-based study. Blood. 2011;118(24):6284-6291.

[55] Lindqvist EK, Landgren O, Lund SH, et al. History of autoimmune disease is associated with impaired survival in multiple myeloma and monoclonal gammopathy of undetermined significance: a population-based study. Ann Hematol. 2017;96(2):261-269.

[56] Brown LM, Gridley G, Check D, Landgren O. Risk of multiple myeloma and monoclonal gammopathy of undetermined significance among white and black male United States veterans with prior autoimmune, infectious, inflammatory, allergic disorders. Blood. 2008;111:3388–94.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

(Lindqvist 2011)[57]. In addition, the OR for pneumonia with more than a five-year latency period was 1.2 (95% CI, 1.04-1.4). The HIM should consider these findings in reviewing the epidemiology literature.

Leukemia

Couto AC, Ferreira JD, Pombo-de-Oliveira MS, Koifman S. Pregnancy, maternal exposure to analgesic medicines, and leukemia in Brazilian children below 2 years of age. European Journal of Cancer Prevention. 2015;24(3):245-252.

- This 2015 case-control study of pediatric leukemia evaluated the risks associated with maternal and early life exposure to analgesics. Cases included children ages birth to 2 years who were diagnosed with acute myeloid leukemia (AML) and acute lymphocytic leukemia (ALL). Controls were in the same age range and hospitalized at the same hospitals as cases and undergoing treatment for life-threatening but nonmalignant disease. Data on environmental exposures and drugs used were collected through surveys administered through face-to-face interviews with the mothers. In analyses of exposures during the three months prior to pregnancy, each trimester, and breastfeeding in the three months after birth, the adjusted ORs ranged from 0.39 to 0.75 for ALL and 0.11 to 0.61 for AML and were not significantly increased.

Liver Cancer

Weiss 2016 had no studies on liver cancer to review. Two studies were published since 2016 by nearly the same authors. The overall results suggest no association. In the first study, which focused on statin use, an increased risk of liver cancer was noted in an unadjusted OR for acetaminophen.[58] In the second study, which found a borderline association, the authors noted the possibility of channeling bias.[59]

McGlynn KA, Hagberg K, Chen J, Graubard BI, London WT, Jick S, Sahasrabuddhe VV. Statin use and risk for primary liver cancer in the clinical practice research datalink. Journal of the National Cancer Institute. 2015;107(4):Article No.: djv009.

- The association between use of statins and liver cancer was evaluated in a nested case-control study using a UK electronic medical records database. The study only evaluated acetaminophen as a covariate in the statin-liver cancer analysis. They reported a crude OR of 1.52 (95% CI 1.31–1.75). However, the acetaminophen assessment was limited to univariate analysis and was not adjusted for confounding factors (e.g., alcohol abuse, BMI, underlying diagnosis leading to use of analgesics). In the context of a significant yet unadjusted OR, the result should be interpreted as hypothesis-generating, rather than hypothesis-testing.

[57] Lindqvist EK, Goldin LR, Landgren O, et al. Personal and family history of immune-related conditions increase the risk of plasma cell disorders: a population-based study. Blood. 2011;118(24):6284-6291
[58] McGlynn KA, Hagberg K, Chen J, et al. Statin use and risk for primary liver cancer in the clinical practice research datalink. J Natl Cancer Inst. 2015;107(4):Article No.: djv009.
[59] Yang B, Petrick JL, Chen J, et al. Associations of NSAID and paracetamol use with risk of primary liver cancer in the Clinical Practice Research Datalink. Cancer Epidemiol. 2016;43:105-11.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Yang B, Petrick JL, Chen J, Hagberg KW, Sahasrabuddhe VV, Graubard BI, Jick S, McGlynn KA. Associations of NSAID and paracetamol use with risk of primary liver cancer in the Clinical Practice Research Datalink. Cancer Epidemiology. 2016;43:105-11.

- The association between use of NSAIDs and liver cancer was evaluated in a nested case-control study using a UK electronic medical records database. They found a slightly increased odds-ratio of borderline significance with two or more acetaminophen prescriptions (OR 1.18, 95% CI 1.00–1.39).
- Current use (use ending within one year before the diagnosis date) and long-term use of acetaminophen (time between first and last prescription > five years) were associated with increased risk of liver cancer (OR 1.30, 95% CI 1.08–1.56 and OR 1.26, 95% CI 1.03–1.54, respectively).
- There were, however, some significantly elevated odds ratios among those without liver disease. For example (in their Supplementary Table 2), for current use, the odds ratio was 1.36 (95% CI 1.12-1.64) and for current use in the 10-19 prescription category, the odds ratio was 1.67 (95% CI 1.19 – 1.52). The odds ratios were not consistent, though, across categories of exposure. For example, for current use of 20-39 prescriptions, the odds ratio was 1.20 (95% CI 0.86-1.67).
- Past use (defined as use ending two years prior to the diagnosis) was not significantly associated with liver cancer risk (OR 1.03, 95% CI 0.84–1.27). Results were similar, but slightly attenuated when the index date was moved one additional year before the diagnosis date for a two year latency period. This pattern of association by duration of use and the attenuated OR for a longer latency period is consistent with protopathic bias and should be taken into account when considering the borderline significance of the OR for two or more prescriptions. Channeling as source of bias was also mentioned by the authors "…patients at highest liver cancer risk (e.g., those with cirrhosis and portal hypertension with thrombocytopenia) may be advised to avoid NSAID use due to risk of gastrointestinal bleeding and renal failure [Singh 2014],[60] and these patients may be more likely to receive paracetamol rather than NSAIDs…."
- Evidence of residual confounding was also observed. The unadjusted OR was 1.45 (95% CI 1.27–1.68) and the adjusted OR was 1.18, suggesting that residual confounding may exist. This is also a limitation that should be considered in the interpretation of the study findings.

In summary, a significant number of epidemiologic studies across a multitude of cancer types including pancreatic, lung, breast, endometrial, ovarian, cervical, prostate, renal cell, skin, brain and lymphohematopoietic and leukemia have shown no association with acetaminophen use. In those cancer types where some studies showed a potential association, the studies had major limitations that suggest caution in their interpretation. Other considerations to determine a causal effect, including consistency, temporality, specificity, biological gradient, and experiment have not been fully demonstrated. In conclusion, the weight of the evidence does not support a causal relationship to acetaminophen use.

Animal Carcinogenicity Studies

[60] Singh S, Singh PP, Roberts LR, Sanchez W. Chemopreventive strategies in hepatocellular carcinoma. Nat Rev Gastroenterol Hepatol. 201411(1):45-54. doi: 10.1038/nrgastro.2013.143.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

We are aware of no animal carcinogenicity studies published since our submission to OEHHA in 2011. All of the existing long-term animal carcinogenicity studies of acetaminophen are identified by OEHHA in its July 2011 document entitled, "Chemical for CIC Consultation: Acetaminophen."[61]

IARC (1999)

We encourage OEHHA to describe in the HIM IARC's comprehensive evaluation of the animal carcinogenicity studies of acetaminophen, as described in IARC monograph, Volume 73 (IARC, 1999). With respect to the animal studies, IARC concluded: "There is *inadequate evidence* in experimental animals for the carcinogenicity of paracetamol [i.e., acetaminophen]."[62] IARC's evaluation is relevant because IARC evaluated the complete set of long-term carcinogenicity studies of acetaminophen, as well as several short-term cancer promotion studies.

NTP (1993)

The National Toxicology Program's (NTP) cancer bioassay (TR-394) is the most recent long-term animal carcinogenicity study of acetaminophen.[63] In its cancer bioassay of acetaminophen, rats and mice were administered 0, 600, 3000 or 6000 ppm of acetaminophen in the diet, and the NTP reached the following conclusions:

- male mice: "*no evidence of carcinogenic activity*"
- female mice: "*no evidence of carcinogenic activity*"
- male rats: "*no evidence of carcinogenic activity*"
- female rats: "*equivocal evidence of carcinogenic activity*."

The NTP's determination of "*equivocal evidence of carcinogenic activity*" in female rats is based on a statistically significant increase in the incidence of mononuclear cell leukemia (MCL) at the high dose only. It is highly doubtful that the "*equivocal evidence of carcinogenicity activity*" in female rats in the NTP bioassay is treatment-related. NTP noted: (1) the increase could be due to chance based on the highly variable incidence of MCL observed spontaneously among Fischer rats, and (2) in contrast to the increased incidence of MCL in high dose female rats, the incidence of MCL decreased with dose in male rats given acetaminophen. In fact, the IARC Working Group noted "the high and variable incidence of mononuclear cell leukemia between and within studies with Fischer rats and considered that this was not a treatment-related effect."[64] Further, no increase in MCL was observed in male or female rats in three other carcinogenicity studies of acetaminophen.

Flaks and Flaks (1983); Flaks et al. (1985)

Two studies by the same group of investigators reported increased incidences of tumors in mice and rats administered acetaminophen (Flaks and Flaks, 1983; Flaks et al., 1985).[65] However, both of these

[61] OEHHA (2011) https://oehha.ca.gov/media/downloads/crnr/101211acetaminophencic.pdf
[62] IARC Monograph (1999) Volume 73, p. 438. https://monographs.iarc.fr/wp-content/uploads/2018/06/mono73-20.pdf
[63] NTP (1993) Toxicology and Carcinogenesis Studies of Acetaminophen (CAS No. 103-90-2) in F344/N Rats and B6C3F1 Mice (Feed Studies). TR-394. https://ntp.niehs.nih.gov/ntp/htdocs/lt_rpts/tr394.pdf
[64] Id., p. 415.
[65] Flaks A, Flaks B (1983) Induction of liver cell tumors in IF mice by paracetamol. Carcinogenesis 4:363-368. Flaks, B, Flaks A, Shaw AP (1985) Induction by paracetamol of bladder and liver tumors in the rat. Effect on hepatocyte fine structure. Acta Pathol Microbiol Immunol Scand 93:367-377.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

studies exhibit serious limitations. For example, in the Flaks and Flaks (1983) study, the high dose level (10,000 ppm in the diet or 500 mg/kg bw/day), which was the only dose that produced an increase in any tumors in mice, greatly exceeded the Maximum Tolerated Dose (MTD). The death rate at the high dose level was 55% (33/60) and 12% (7/60) among the male and female mice, respectively (Flaks and Flaks, 1983); most of these deaths occurred during the first two days of exposure. Exposure to acetaminophen in the diet was terminated for the surviving high-dose mice at 18 months of exposure when the body weights of the males and females were approximately 38% and 31%, respectively, lower than those of the controls. In long-term toxicity studies, dose levels that produce greater than a 10% reduction in body weight compared to controls are considered to exceed the MTD.

Other limitations of the Flaks and Flaks (1983) study include: inadequate description of the methodology, lack of statistical analysis of the tumor data, no randomized assignment of animals, no observation of clinical symptoms, no measurement of food consumption, no testing of diets to validate the concentration and stability of the test material, frequency of body weight measurements not stated, and only two dose levels of acetaminophen.

For their study of acetaminophen in rats, Flaks et al. (1985) used Leeds rats, an inbred strain of rats. These investigators observed statistically significant increases in benign, but not malignant, bladder tumors in males at the high dose (10,000 ppm or 600 mg/kg bw/day) only and in females at the low dose (5000 ppm) only and in liver "neoplastic nodules" in both sexes at the high dose only. However, the IARC Working Group "noted that in the study in rats in which tumours were induced (Flaks et al., 1985) no tumours were found in either male or female controls, which is a highly unusual finding and raises questions about the interpretation of the findings."[66] Other limitations of the Flaks et al. (1985) study include: limited description of methods, no description of the statistical methods, no randomized assignment of animals, no observation of clinical symptoms, no testing of diets to validate the concentration and stability of the test material, and infrequent (monthly) measurements of body weights.

Weisburger et al. (1973)

This older study by Weisburger et al. (1973)[67] was included under the section on long-term dietary studies in mice in the prioritization document prepared by OEHHA in 2011 entitled, "Chemical for CIC Consultation: Acetaminophen."[68] This study was described as an "11-month study in female NIH mice." We suggest a more complete description of the study, now that we are past the prioritization stage, would be as a tumor promotion study of acetaminophen in rats, mice, and hamsters. The focus of the study was to evaluate the effect of acetaminophen (11,000 ppm in the diet) and acetanilide on the incidence of tumors initiated by two known carcinogens: N-2-fluorenylacetamide (FAA) and N-hydroxy-N-2-fluorenylacetamide (N-OHFAA).

In the 2011 prioritization document, the result of this study was reported as "increase in bladder epithelial tumor" in female mice, which is not accurate[69]. Among the female mice, there were no

[66] IARC Monograph (1999) p. 415.

[67] Weisburger JH, Weisburger EK, Madison RM et al. (1973) Effect of acetanilide and p-hydroxyacetanilide on the carcinogenicity of N-2-fluorenylacetamide and N-hydroxy-N-2-fluorenylacetamide in mice, hamsters, and female rats. J Natl Cancer Inst 51:235-240.

[68] OEHHA (2011) https://oehha.ca.gov/media/downloads/crnr/101211acetaminophencic.pdf

[69] We recognize that OEHHA's prioritization documents are intended to give a high-level picture of information and that it is not possible for OEHHA to devote substantial time to reading each study identified in a prioritization

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

urinary bladder tumors observed in the group given acetaminophen alone or in the negative control group. In male mice, two urinary bladder tumors were reported among the 26 mice exposed to acetaminophen alone, and no urinary bladder tumors were reported among the 27 negative control males. The authors did not conduct a statistical analysis of the tumor data, but this small increase in the males is not expected to be statistically significant. When acetaminophen was given in combination with FAA or N-OHFAA, the incidence of bladder tumors was lower than observed when FAA or N-OHFAA were given alone among the male mice. Among the female mice, neither FAA nor N-OHFAA alone produced a significant increase in bladder tumors, and the combination of acetaminophen and FAA or N-OHFAA did not increase the incidence of bladder tumors.

The full results of this study should be included in the HIM. For example, acetaminophen inhibited mammary tumors in female rats produced by two known carcinogens. It also inhibited liver tumors in male and female mice produced by two known carcinogens. And, in hamsters, acetaminophen inhibited the incidence of gastric tumors induced by a known carcinogen. There was no convincing evidence that acetaminophen alone produced tumors in rats, mice or hamsters, but the duration of exposure was less than 1 year for all species.

Other Long-term Animal Carcinogenicity Studies

As noted in OEHHA's 2011 prioritization document, no acetaminophen treatment-related tumor findings were observed in the following long-term animal carcinogenicity studies of acetaminophen:

- Hiraga and Fujii (1985) study in male rats (0, 4500, 9000 ppm in the diet) and female rats (0, 6500, 13,000 ppm in the diet)[70]
- Johansson (1981) study in male rats (0, 5350 ppm in the diet)[71]
- Amo and Matsuyama (1985) study in male and female mice (0, 3000, 6000 ppm in the diet)[72], and
- Hagiwara & Ward (1986) study in male mice (0, 5000, 10,000 ppm in the diet).[73]

Tumor Promotion Studies

In the 2011 prioritization document, OEHHA identified three tumor promotion studies of acetaminophen in which mice or rats were exposed to various nitrosamine tumor initiators: Tsuda et al. (1984), Kurata et al. (1986), and Hagiwara and Ward (1986). As noted in the 2011 prioritization document, no acetaminophen treatment-related tumor findings were observed in the Kurata et al. (1986) and Hagiwara and Ward (1986) studies.

The 2011 prioritization document summarized the results of the Tsuda et al. (1984) study as "Increase in renal cell adenoma." This is an incomplete description of the results of this study. The study authors concluded that acetaminophen inhibited liver tumors and enhanced renal cell adenomas (but not carcinomas) in male Fischer 344 rats given a known carcinogen (N-nitrosoethyl-N-

---

summary. We are trying to be helpful in pointing out information for OEHHA. Like OEHHA, our interest is in helpful and accurate hazard identification materials.

[70] Hiraga K, Fujii T (1985) Carcinogenicity testing of acetaminophen in F344 rats. Jpn J Cancer Res 76:79-85.

[71] Johansson SL (1981) Carcinogenicity of analgesics: long-term treatment of Sprague-Dawley rats with phenacetin, phenazone, caffeine, and paracetamol (acetamidophen). Int J Cancer 27:521-529.

[72] Amo H, Matsuyama M (1985) Subchronic and chronic effects of feeding of large amounts of acetaminophen in B6C3F1 mice. Jpn I Hyg 40:567-574.

[73] Hagiwara A, Ward JM (1986) The chronic hepatotoxic, tumor-promoting, and carcinogenic effects of acetaminophen in male B6C3F1 mice. Fund Appl Toxicol 7:376-386.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

hydroxyetylamine) in drinking water followed by administration of 13,000 ppm of acetaminophen in the diet for 29 weeks. In comparison, no renal cell adenomas or carcinomas were observed in male rats given acetaminophen in the absence of N-nitrosoethyl-N-hydroxyethylamine.

Two additional tumor promotion studies of acetaminophen were found: Shibata et al. (1995) and Williams and Iatropoulos (1997).[74] These two studies do not appear in OEHHA's 2011 prioritization document. In a model of urinary bladder carcinogenesis, Shibata et al. (1995) evaluated the tumor promotion potential of acetaminophen in male Fischer 344 rats treated 0.1% N-nitrosodi(2-hydroxypropyl)amine in the drinking water and 3% uracil in the diet for the first four weeks. The rats were subsequently administered 0 or 0.8% acetaminophen in the diet for 35 weeks. Acetaminophen did not significantly increase the incidences of tumors of the renal tubules, renal pelvis, ureter or urinary bladder when compared to the tumor-initiated control group.

In a model of intestinal carcinogenesis, Williams and Iatropoulos (1997) evaluated the tumor promotion potential of acetaminophen in male Fischer 344 rats administered diets containing 0, 250 or 5000 ppm of acetaminophen for 44 weeks. Beginning two weeks after the start of the acetaminophen diets, the rats were given weekly subcutaneous injections of 50 mg/kg of 3,2′-dimethyl-4-aminobiphenyl (DMAB) for 20 weeks. The survival rate of rats receiving DMAB alone was less than 50% mainly due to intestinal obstruction from tumor growth. No intestinal tumors were observed in the negative control group or among the rats receiving acetaminophen alone. Acetaminophen inhibited the intestinal tumors initiated by DMAB.

Tumor Initiation Studies in Rats with Compromised Liver Function

In addition to the tumor promotion studies described above, we are aware of two tumor initiation studies of acetaminophen in rats with partial hepatectomies or with choline-induced liver damage: Hasegawa et al., (1988) and Maruyama et al. (1990).[75] Neither of these studies was identified in OEHHA's 2011 prioritization document. The first of these studies (Hasagawa et al.,1988) was described in the IARC monograph (1990):

> "Groups of male Fischer 344 rats [numbers unspecified], six weeks of age, were subjected to a two-thirds partial hepatectomy and 24 h later received either intragastric intubations of paracetamol (purity, >99%) at 0 or 1000 mg/kg bw in 0.2% tragacanth gum twice a week for five weeks, or a single intragastric instillation of paracetamol at 500 mg/kg bw. Two weeks after the end of paracetamol treatment, the animals were administered phenobarbital (pharmacopoeial grade) at 0 or 1 mg/ml drinking-water for 12 weeks. The experiment was terminated at the end of phenobarbital treatment (weeks 13 and 18). Livers, kidneys, thyroid glands and any gross lesions were examined histologically. The tumour-initiating activity of paracetamol was evaluated by the formation of placental-type glutathione S-transferase-positive foci in liver cells; treatment with paracetamol did not result in the induction of such foci (Hasegawa et al., 1988). [The Working Group noted that the rate of absorption of

[74] Shibata MA, Sano M, Hagiwara et al. (1995) Modification by analgesics of lesion development in the urinary tract and various other organs of rats pretreated with dihydroxy-di-N-propylnitrosamine and uracil. Jpn J Cancer Res 86(2):160-167.
Williams GM, Iatropoulos MJ (1997) Inhibition by acetaminophen of intestinal cancer in rats induced by an aromatic amine similar to food mutagens. Eur J Cancer Prev 6(4):357-362.
[75] Hasegawa R, Furukawa F, Toyoda K et al. (1988) Study for tumor-initiating effect of acetaminophen in two stage liver carcinogenesis in male F344 rats. Carcinogenesis 9:755-759.
Maruyama H, Takashima Y, Murata Y et al. (1990) Lack of hepatocarcinogenic potential of acetaminophen in rats with liver damage associated with a choline-devoid diet. Carcinogenesis 11(6):895-901.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

> paracetamol from the tragacanth suspension was not measured, and the limited reporting of the experiment.]"[76]

Hasegawa et al. (1988) concluded: "These results indicate that [acetaminophen] possesses no tumor-initiating activity in the rat liver."

In the second study, Maruyama et al. (1990) evaluated the potential carcinogenic activity of acetaminophen in rats with pre-existing liver damage induced by a choline-devoid diet. This study was described in the IARC Monograph (1999):

> "In a short-term model of fatty liver and liver cirrhosis, groups of 8-22 male Fischer 344 rats, six weeks of age, were fed either a choline-deficient or a choline-supplemented diet for four weeks, followed by a single oral gavage of acetaminophen at a [sic] doses of 0, 0.5, 1 or 1.5 mg/kg bw in 0.2% tragacanth gum solution. Four hours after the paracetamol treatment, the rats were subjected to a two-thirds hepatectomy, followed by a two-week recovery period, and then two weeks of feeding of 0.02% of 2-acetylaminofluorene coupled with a single gavage of carbon tetrachloride at the mid-point. The rats were killed nine weeks after the beginning of the study after an 18-h fast. A positive control group received a single intraperitoneal injection of N-nitrosodiethylamine in the place of paracetamol. Foci of hepatocellular alteration were assayed by immunohistochemical staining for γ-glutamyltranspeptidase or the placental form of glutathione-S-transferase. Paracetamol did not significantly alter the number or size of liver foci when compared with the relevant control values (Maruyama et al., 1990)."[77]
>
> "In a long-term model of fatty liver and liver cirrhosis, groups of 8-13 male Fischer 344 rats, six weeks of age, were fed either a choline-deficient diet for 27 weeks to produce liver cirrhosis or a choline-supplemented diet as a control group. From 27 weeks, the rats were fed a diet containing 0,0.05, 0.45, or 0.9% paracetamol, basal diet or choline-supplemented diet, until week 52, at which time the surviving animals were killed. Foci of hepatocellular alteration were assayed by immunohistochemical staining for γ-glutamyl-transpeptidase or the placental form of glutathione-S-transferase. Paracetamol did not alter the number or size of liver foci when compared with the relevant control groups (Maruyama et al., 1990)."[78]

Maruyama et al. (1990) concluded: "Thus, these results indicate that [acetaminophen] does not possess significant carcinogenic activity in damaged rat liver."

Genotoxicity Studies

The available data demonstrate that acetaminophen does not cause gene mutations in bacteria or mammalian cells. While there are published data showing that acetaminophen causes chromosomal damage in mammalian cells *in vitro* and *in vivo*, these results are only observed under conditions that are toxic to the test systems.

[76] IARC Monograph (1990) Volume 50, p. 313.
[77] IARC Monograph (1999) Volume 73, p. 416.
[78] Id.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

In 1996, Bergman et al. published a comprehensive review of the genotoxicity and carcinogenicity of acetaminophen.[79] In their review of the genotoxicity data, the authors note that the clastogenic effects of acetaminophen occur by three possible threshold mechanisms: (1) inhibition of ribonucleotide reductase; (2) increase in cytosolic and intranuclear Ca2+ levels; and (3) DNA damage caused by N-acetyl-p-benzoquinone imine (NAPQI) after glutathione depletion. They further note that "[rat] studies, which employed doses ranging from the dose resulting in human therapeutic peak plasma levels to highly toxic doses, give convincing evidence that genotoxic effects of paracetamol appear only at dosages inducing pronounced liver and bone marrow toxicity." Bergman also notes that the only published carefully designed human randomized, placebo-controlled trial evaluating the genotoxicity of acetaminophen was negative (Kirkland 1992)[80].

The assessment of the available data up to 1996 by Bergman, et al., supports the conclusion that acetaminophen's clastogenic effects are only observed at doses or concentrations that are toxic to the test systems. In the sections below we have provided more detailed summaries and interpretations of studies that have been published after 1996.

New Genotoxicity Studies Since Bergman et al., 1996 and Interpretation of Results

A literature review identified thirteen additional studies published after Bergman et al., 1996. These studies are summarized below along with a study report for an unpublished study conducted by Janssen Research & Development, LLC. These publications and the unpublished study further support the conclusion that acetaminophen is not a carcinogenic hazard.

Published Studies

Suzuki Y, Goto K, Nakayama Y, Saratani M, Takata T, Okamoto T, Okazaki. Evaluation of a single-dose PIGRET assay for acetaminophen in rats compared with the RBC Pig-a assay. S. Mutat Res. 2016;811:16-20.

A genotoxicity study on acetaminophen was performed using the red blood cell Pig-a and PIGRET assays. While the Pig-a assay does not currently have an Organisation for Economic Cooperation and Development (OECD) test guideline, preparations are now underway for a new OECD guideline for the Pig-a assay (Chikura et al. 2019)[81]. In addition, the assay is recommended in the International Conference on Harmonization (ICH) guideline M7(R1) (Chikura et al. 2019).[82] The dose levels by Suzuki et al. (2016) were set at 0, 500, 1000, and 2000 mg/kg and acetaminophen was administered once by oral gavage to male Sprague Dawley rats. The RBC Pig-a and PIGRET assays were performed using peripheral blood collected at pre-dosing and 1, 2 and 4 weeks after dosing. In both the assays, there were no changes in the Pig-a gene mutant frequency by acetaminophen treatment at any time point. The results demonstrate that acetaminophen is not mutagenic at any of the doses tested in the Pig-a and PIGRET assays.

---

[79] Bergman K, Müller L, Teigen SW. Series: current issues in mutagenesis and carcinogenesis, No. 65. The genotoxicity and carcinogenicity of paracetamol: a regulatory (re)view. Mutat Res. 1996;349(2):263-88.
[80] Kirkland DJ, Dresp JH, Marshall RR, et al. Normal chromosomal aberration frequencies in peripheral lymphocytes of healthy human volunteers exposed to a maximum daily dose of paracetamol in a double blind trial. Mutat Res. 1992;279(3):181-94.
[81] Chikura S, Kimoto T, Itoh S, et al. Standard protocol for the total red blood cell Pig-a assay used in the interlaboratory trial organized by the Mammalian Mutagenicity Study Group of the Japanese Environmental Mutagen Society. Genes Environ. 2019;41:5. https://doi.org/10/1186/s41021-019-0121-z.
[82] Ibid

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

McGill MR, Sharpe MR, Williams CD, Taha M, Curry SC, Jaeschke H. J. The mechanism underlying acetaminophen-induced hepatotoxicity in humans and mice involves mitochondrial damage and nuclear DNA fragmentation. Clin Invest. 2012;122(4):1574-83.

McGill et al. prospectively examined patients admitted to the University of Kansas Hospital or to the Banner Good Samaritan Medical Center following acetaminophen overdose. All of the participants fulfilled at least two of the following inclusion criteria: (1) patient-reported history of acetaminophen overdose, (2) high-serum acetaminophen levels, and (3) abnormal liver function tests (LTs) (based on ALT, aspartate aminotransferase [AST], Prothrombin Time, bilirubin). Levels of biomarkers of mitochondrial damage (glutamate dehydrogenase [GDH] and mitochondrial DNA [mtDNA]) and nuclear DNA fragments were measured in plasma. Overdose patients with no or minimal hepatic injury who had normal LTs) (normal LT group) and healthy volunteers served as controls. Peak GDH activity and mtDNA concentration were increased in plasma from patients with abnormal LT but not in patients with minimal hepatic injury (normal LTs). Similarly, peak nuclear DNA fragmentation in the abnormal LT cohort was increased over that of controls but not in those patients with normal LT. Parallel studies in mice were also performed. Male C57BL/6 mice were fasted overnight before receiving an i.p. injection of 300 mg/kg acetaminophen in warm 0.9% saline. A comparison of plasma DNA fragments and nuclear DNA damage in mouse liver after acetaminophen exposure revealed that both parameters correlated with ALT release. This result is consistent with the hypothesis that nuclear DNA damage only occurs at concentrations where there is liver cell injury.

Klopcic I, Poberznik M, Mavri J, Dolenc MS. A quantum chemical study of the reactivity of acetaminophen (paracetamol) toxic metabolite N-acetyl-p-benzoquinone imine with deoxyguanosine and glutathione. Chemico-Biological Interactions. 2015;242: 407-414.

The purpose of this study was to evaluate the chemical reactivity of an acetaminophen metabolite NAPQI using quantum chemical methods. Specifically, the authors evaluated the activation energy necessary for NAPQI to react with glutathione (GSH) or DNA. The authors reported that the interaction between NAPQI and DNA is not a simple reaction but involves a complex reaction cascade with a rate-limiting step. The authors reported an activation barrier of 26.7 kcal/mol for the rate-limiting step, demonstrating that the interaction between NAPQI and DNA is slow (on a time scale of hours). In contrast, the authors reported that the reaction between NAPQI and GSH is very fast, with an activation barrier of 12.9 kcal/mol. The authors concluded that scavengers such as GSH react with NAPQI at a much faster rate than DNA. These results suggest that interaction of NAPQI with DNA is only possible under specific conditions where there is a long-term depletion of GHS and other thiol proteins such as individuals with alcoholism, HIV, COPD, diabetes, cystic fibrosis, and rheumatoids. Additionally, the authors noted that only 5% of acetaminophen is metabolized to NAPQI by CYP2E1. Taken together, the results of this study suggest that there is a very low probability that NAPQI will interact with DNA and lead to genotoxic changes and is consistent with the threshold mechanisms described by Bergman et al, 1996.

Hantson P, de Saint-Georges L, Mahieu P, Léonard ED, Crutzen-Fayt MC, Léonard A. Evaluation of the ability of paracetamol to produce chromosome aberrations in man. Mutation Research. 1996; 368, 293-300.

The ability of paracetamol to induce structural chromosome aberrations in human peripheral blood lymphocytes was evaluated in five volunteers who had been administered a single oral dose of

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

paracetamol (3 g), in five patients who had received 2 g of propacetamol (pro-drug of paracetamol) by intravenous infusion every 6 h for at least 7 days, and in five self-poisoned patients who had ingested >10 g paracetamol. The authors found no significant difference in the percentage of abnormal cells before and after application of paracetamol in volunteers and in patients after intravenous infusion of a total of 28 g paracetamol (as 56 g of propacetamol). The yield of abnormal cells was not modified in self-poisoned persons who had ingested greater than 10 g of paracetamol. The negative results in this study are consistent with previous analyses in humans (Kirkland, 1992)[83] which report that genotoxicity resulting from acetaminophen exposure does not occur at non-hepatotoxic doses.

Brennan RJ, Schiestl RH. Aniline and its metabolites generate free radicals in yeast. Mutagenesis. 1997;12: 215-220.

Brennan et al. (1997) examined whether 4-acetamidophenol (acetaminophen) causes recombinagenic activity in *S.cerevisiae.* The authors reported that acetaminophen was non-toxic and non-recombinagenic in yeast at concentrations up to 130 mM. While the OECD guideline for "Genetic Toxicology: *Saccharomyces cerevisiae*, Mitotic Recombination Assay"[84] was deleted/recalled in 2014, the results presented by Brennan et al. 1997 are consistent with previous analyses which demonstrate that acetaminophen is non-mutagenic.

Ibrulj S, Rahmanovic A, Haveric S, Haveric A, Pasic AD. Cytogenetic evaluation of paracetamol effects in human lymphocytes culture. Drug Chem Toxicol. 2007;30: 133-143.

Ibrulj et al. (2007) examined cytogenetic effects in human peripheral blood lymphocytes after exposure to 50 – 200 µg/mL (0.3 – 1.3 mM) paracetamol. While frequencies of binuclear cells with micronuclei and total number of micronuclei as well as nuclear division index were not statistically different in comparison with controls, the authors concluded that cytogenetic tests revealed clastogenic activity of high paracetamol concentration of 200 µg/mL (1.3 mM). Current ICH guidelines recommend a top dose of 1 mM for genotoxic hazard identification (ICH 2011)[85] and therefore this should be noted in the HIM and considered in evaluating the relevance of this study.

Kanki K, Nishikawa A, Masumura K, Umemura T, Imazawa T, et al. In vivo mutational analysis of liver DNA in gpt delta transgenic rats treated with the hepatocarcinogens N-nitrosopyrrolidine, 2-amino-3-methylimidazo[4,5-f]quinoline, and di(2-ethylhexyl)phthalate. Mol Carcinog. 2005;42: 9-17.

Kanki et al. (2005) investigated the in vivo mutagenicity and mutation spectra of known genotoxic rat hepatocarcinogens N-nitrosopyrrolidine (NPYR), and 2-amino-3-methylimidazo[4,5-f]quinoline (IQ), as well as the nongenotoxic hepatocarcinogen di(2-ethylhexyl) phthalate (DEHP) and "the noncarcinogen acetaminophen" in guanine phosphoribosyltransferase (gpt) delta transgenic rats, a recently developed animal model for genotoxicity analysis (Kanki et al. 2005: p. 9). Animals were fed

[83] Kirkland DJ, Dresp JH, Marshall RR, et al. Normal chromosomal aberration frequencies in peripheral lymphocytes of healthy human volunteers exposed to a maximum daily dose of paracetamol in a double blind trial. Mutat Res. 1992;279(3):181-94.
[84] OECD. Test No 481: Genetic Toxicology: Saccharomyces cerevisiae, Mitotic Recombination Assay, OECD Guidelines for the Testing of Chemicals, Section 4, OECD Publishing. Paris. https://doi.org/10.1787/9789264071421-en
[85] ICH S2 (R1) Genotoxocity testing and data interpretation for pharmaceuticals intended for human use. Published 01/12/2011. Reference number CHMP/ICH/126642/08.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

10,000 ppm of acetaminophen in their diet for 13 weeks and were then sacrificed. A section of the left lateral lobe of the liver was utilized for multiple in vivo mutation assays. An increase in the liver/bodyweight ratio ($p<0.01$) of animals treated with acetaminophen was observed relative to controls. However, the number and area of GST-P positive liver cell foci were not significantly different between acetaminophen-treated rats and controls. Few GST-P-positive liver cell foci were observed in acetaminophen-treated and untreated control rats (as well as DEHP-treated rats), compared to significantly increased number and area of GST-P-positive liver cell foci in IQ and NPYR treated groups. With respect to acetaminophen treated animals, there was no genotoxicity.

Markovic D, Katic J, Stojkovic R, Borovic S, Zarkovic N, Fucic A. Lipid peroxidation, detoxification capacity, and genome damage in mice after transplacental exposure to pharmaceutical drugs. Braz J Med Biol Res. 2013;46: 1014-1020.

Markovic et al. (2013) analyzed the transplacental genotoxicity of paracetamol in BALB/c mice. Pregnant mice were administered 60 mg/kg paracetamol via intraperitoneal injection for three days. The micronucleus frequency was reported for both dams and newborn pups. Paracetamol did not increase the micronucleus frequency in dams. However, paracetamol increased the micronucleus frequency in newborn pups. This study did not adhere to OECD 474 guidelines[86] since it did not test a minimum of three dose levels. Other limitations and confounding factors in the study include that it did not evaluate general toxicity or systemic exposure, lacked a suitable positive control for the pups, and only evaluated a single time point for micronuclei in the pups. In addition, intraperitoneal injection is an inappropriate route of administration to evaluate genotoxicity for an oral compound and the authors suggest that the dose used was hepatotoxic. Thus, this study has limited biological relevance. The above limitations and confounding factors should be noted in the HIM and considered in evaluating the relevance of this study.

Martinez A, Urios A, Blanco M. Mutagenicity of 80 chemicals in Escherichia coli tester strains IC203, deficient in OxyR, and its oxyR(+) parent WP2 uvrA/pKM101: detection of 31 oxidative mutagens. Mutat Res. 2000;467: 41-53.

Martinez et al. (2000) investigated the mutagenicity of 80 chemicals, including paracetamol, using a new bacterial reversion assay, the WP2 Mutotoxitest, in *Escherichia coli* strain IC203[87]. The authors reported that paracetamol was non-mutagenic in concentrations up to 1500 µg/plate. These results are consistent with previous studies which demonstrate that acetaminophen is non-mutagenic.

Matsushita K, Kijima A, Ishii Y, Takasu S, Jin M, Kuroda K, Kawaguchi H, Miyoshi N, Nohmi T, Ogawa K, Umemura T. Development of a Medium-term Animal Model Using gpt Delta Rats to Evaluate Chemical Carcinogenicity and Genotoxicity. J Toxicol Pathol. 2013;26: 19-27.

In this study, acetaminophen was used as a non-genotoxic control in the development of an animal model for detecting chemical carcinogens. Groups of 15 six-week-old male *gpt* delta rats were fed 6,000 ppm acetaminophen in their diets or control diets for 4 weeks, then had a partial hepatectomy where samples were used in the in vivo mutation assay. Eighteen hours after the partial hepatectomy, an i.p. injection of 10 mg/kg diethylnitrosamine was administered. The rats continued on the control or

[86] OECD. Test No. 474: Mammalian Erythrocyte Micronucleus Test, 1997. OECD Publishing, Paris. https://doi.org/10.1787/9789264071285-en.
[87] Blanco M, Urios A, Martínez A. New Escherichia coli WP2 tester strains highly sensitive to reversion by oxidative mutagens. Mutat Res. 1998;413(2):95-101.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

acetaminophen diets until ten weeks after the start of the experiment when liver tissues were fixed and immunohistochemistry was performed for the quantitative analysis of GST-P-positive foci. For the liver samples from the partial hepatectomy, the mutation frequency of the reporter genes were examined as an indication of tumor initiation. The mutation frequency of the acetaminophen group was not significantly different from control indicating that acetaminophen treatment did not result in mutagenicity in vivo. For the liver samples collected at ten weeks, immunohistochemistry showed that the number and area of GST-P positive foci were significantly decreased in acetaminophen treated rats in comparison to control. The GST-P positive foci following a proliferative stimuli (i.e., partial hepatectomy) measure tumor promoting activities which were not exhibited by rats receiving acetaminophen. While the ip route of administration in this study is not relevant to oral administration of acetaminophen, these results are consistent with previous studies which demonstrate that acetaminophen is non-mutagenic.

Simkó M, Kriehuber R, Lange S. Micronucleus formation in human amnion cells after exposure to 50 Hz MF applied horizontally and vertically. Mutation Research/Genetic Toxicology and Environmental Mutagenesis. 1998;418: 101-111.

In this study, acetaminophen was used alone and in conjunction with magnetic fields as an inhibitor of DNA-repair mechanisms to better understand the effects of magnetic fields. Human amniotic fluid cells were treated with acetaminophen (0.3, 0.6, 1.0, 1.5, 2.0, 2.5 mM) for 24, 48, and 72 hours and were assessed for the induction of micronuclei (MN). While the authors noted a dose- and time-dependent induction of MN after exposure to acetaminophen, only 1.3 mM exposure after 72 hours was noted to be significant compared to controls. Current ICH guidelines recommend a top dose of 1 mM for genotoxic hazard identification (ICH 2011)[88] and therefore this should be noted in the HIM and considered in evaluating the relevance of this study.

Oshida K, Iwanaga E, Miyamoto-Kuramitsu K, Miyamoto Y. An in vivo comet assay of multiple organs (liver, kidney and bone marrow) in mice treated with methyl methanesulfonate and acetaminophen accompanied by hematology and/or blood chemistry. J Toxicol Sci. 2008;33: 515-24.

The purpose of this study was to evaluate and validate the *in vivo* comet assay, which is a simple and reliable method for measuring DNA damage in cells with or without the capability of cell division, in multiple organs. In this study, male Crlj:CD1(ICR) mice (N=3/group) were intraperitoneally exposed to acetaminophen at doses of 12, 60, and 300 mg/kg acetaminophen. This method followed OECD test guideline 489 standards[89], with the exception of the route of exposure. A vehicle control group was included as a negative/absolute control. Mice were sacrificed at 4 and 24 hour post-injection for sample collection of liver, kidney and bone marrow tissues. Acetaminophen caused moderate statistically significant DNA damage in only the liver but not the kidney and bone marrow at 300 mg/kg acetaminophen at 4 hours and 24 hours post-injection. The corresponding significant alterations in liver injury markers in mice exposed to 300 mg/kg acetaminophen at 4 hours and 24 hours demonstrate that there was hepatotoxicity at this dose level. The results in this study suggest that DNA damage occurs following high doses of acetaminophen in the liver at hepatotoxic doses. These conclusions are consistent with previous studies that genotoxicity resulting from acetaminophen

[88] ICH S2 (R1) Genotoxocity testing and data interpretation for pharmaceuticals intended for human use. Published 01/12/2011. Reference number CHMP/ICH/126642/08.
[89] OECD. Test No. 489: In Vivo Mammalian Alkaline Comet Assay, OECD Guidelines for the Testing of Chemicals, Section 4, 2016. OECD Publishing, Paris, https://doi.org/10.1787/9789264264885-en.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

exposure only occurs at toxic doses. It is important to note the following limitations of this study for evaluating acetaminophen genotoxicity in the HIM: the exposure route was intraperitoneal injection (not suitable for evaluating the genotoxicity of an orally administered drug) and the doses that were used caused hepatoxicity, which confound the ability to evaluate genotoxicity potential.

Matsushima T, Hayashi M, Matsuoka A, Ishidate M Jr, Miura KF, Shimizu H, Suzuki Y, Morimoto K, Ogura H, Mure K, Koshi K, Sofuni T. Validation study of the *in vitro* micronucleus test in a Chinese hamster lung cell line (CHL/IU). Mutagenesis. 1999;14: 569-80.

The purpose of this study was to validate the Chinese hamster lung cell line CHL/IU for use in the *in vitro* MN assay. This study is currently cited in the OECD test guideline standard for *in vitro* MN assay (OECD 487)[90]. Matsushima et al. (1999) exposed CHL/IU monolayer cell culture plates to 66 chemicals, one of which is acetaminophen. For acetaminophen, concentrations ranged from 15 – 120 µg mL (0.01 mM – 0.79 mM) and cells were continuously exposed for 24 or 48 hours. While data for MN responses after acetaminophen exposure were indicated as positive by the authors there was no indication of statistical significance or number of replicates. It was also unclear if any cytotoxicity testing was performed to evaluate the maximum concentration for exposure before 50% cytotoxicity. Therefore, the biological relevance of the results by Matsushima et al. are in question. The limitations and confounding factors should be included in the HIM.

Rogers LK, Moorthy B, Smith CV (1997) Acetaminophen binds to mouse hepatic and renal DNA at human therapeutic doses. Chem Res Toxicol 10: 470-476.

Rogers et al. (1997) injected male ICH mice with radio-labeled acetaminophen via i.p. at doses of 10, 50, 100, and 400 mg/kg. The authors observed an increase in radio-labeled DNA isolated from hepatic and renal tissue for all doses and suggested that this was a specific binding between DNA and acetaminophen or an acetaminophen metabolite. However, the authors did not control for in vivo dissociation of the radio-label which limits the interpretation of the specificity for the increase in radio-labeled DNA. DNA adduct formation in mice was also tested using 32P-postlabeling and no differences were observed for acetaminophen treated mice compared to control mice. While the lack of in vivo controls and use of i.p. route of administration limit the interpretation for oral use of acetaminophen, the lack of DNA adduct formation is consistent with previous studies which demonstrate that acetaminophen is non-mutagenic.

Unpublished Study (manuscript in preparation)

1-Month repeated dose oral toxicity study of acetaminophen in the rat including Pig-a mutation, micronucleus, comet and liver biomarker assessments. Unpublished Janssen Research & Development, LLC Data.

This study was conducted to evaluate the viability of the Pig-a mutation as a genotoxicity endpoint as part of a consortium. Acetaminophen was selected as one of many test compounds. This study evaluated the potential of acetaminophen to induce mutations in the Pig-a gene leading to glycosylphosphatidylinositol (GPI) anchor deficiency in peripheral blood erythrocytes, structural and/or numerical chromosome aberrations in peripheral blood erythrocytes (micronucleus), DNA

[90] OECD. Test No. 487: Guideline for the testing of chemicals, In vitro mammalian cell micronucleus Test, 2016. http://www.oecd-ilibrary.org/environment/test-no-487-in-vitro-mammalian-cell-micronucleus-test_9789264264861-en.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

damage in peripheral blood leukocytes and liver cells (comet). In addition, the toxicity in liver and kidney of male rats was investigated. Mutagenesis was also measured following a 28 day recovery period post the acetaminophen treatment in the highest dose treatment and control groups. The study also included hematology and liver biomarkers from high dose acetaminophen treatment at exposure values far exceeding human relevant exposures. A manuscript describing this study is being prepared for publication and a brief summary of the methods, results and conclusions are provided below.

Methods

Acetaminophen was administered once daily by the oral gavage to groups of five or six male Sprague-Dawley rats as an aqueous suspension in demineralized water containing 0.5% w/v Methocel (hydroxylpropyl methylcellulose) at 0, 250, 500 and 1000 mg/kg /day for 28 days.

At 0 and 1000 mg/kg/day, an additional group was included receiving a recovery period of 28 days. Peripheral blood samples were collected and analyzed on Days -1, 14, 28 and 56 for mutations in the Pig-a mutation assay. Peripheral blood samples were also collected on Days 3 and 28 and analyzed for chromosome aberrations in the micronucleus test. In addition, peripheral blood samples and liver cells were collected on Day 28 and analyzed for DNA damage in the comet assay.

Results

Pig a Mutation Assay

In the Pig-a mutation assay, no biologically relevant increase in the frequency of mutant phenotype (CD59-negative) erythrocytes was observed in the acetaminophen dose groups on Days 14, 28, or 56 (recovery phase). These results demonstrate that acetaminophen did not induce mutations in the Pig-a assay.

Micronucleus Assay

In the micronucleus assay, no biologically relevant or statistically significant increase in the percentage of micronucleated reticulocytes (i.e., evidence of chromosomal damage) was observed in the dose groups up to 500 mg/kg/day on Day 3. The 1000 mg/kg/day dose evaluated on Day 3 could not be evaluated for micronucleus induction due to severe bone marrow toxicity. There was a reduction in percent reticulocytes which indicates impact on erythropoiesis/hematopoietic system.

The micronucleus endpoint evaluated on Day 28 demonstrated no micronucleus/chromosomal aberration changes at the 250 mg/kg/day dose level. Doses of 500 and 1000 mg/kg showed hematotoxicity and therefore should not be considered for further evaluation per the ICH S2[91] guidance and OECD 474[92] given that marked hematotoxicity can affect the ability to detect micronuclei. The ICH S2 also states that data such as this should be interpreted with caution as it is not uncommon to see small increases in micronucleated reticulocytes at dose levels which impact the erythropoiesis/hematopoietic system. (Tweats et. al. 2007, ICH 2011, OECD 481)[93, 94,95]. It should be

[91] ICH S2 (R1). Genotoxocity testing and data interpretation for pharmaceuticals intended for human use. Published 01/12/2011. Reference number CHMP/ICH/126642/08.
[92] OECD. Test No. 474: Mammalian Erythrocyte Micronucleus Test, 1997. OECD Publishing, Paris. https://doi.org/10.1787/9789264071285-en.
[93] Tweats DJ, Blakey D, Heflich RH, et al. Determination of genetic toxicity and potential carcinogenicity in vitro--challenges post the Seventh Amendment to the European Cosmetics Directive. Mutagenesis. 2007;627:78-91.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

noted that a small, but statistically significant increase in the percentage of micronucleated reticulocytes was presented in the report after 28 days of exposure to hematotoxic doses of 500 and 1000 mg/kg.

Comet Assay

In the comet assay, no biologically relevant or statistically significant increase in the level of DNA migration (as represented by % tail intensity) was observed in peripheral blood leukocytes in the acetaminophen dose groups on Day 28. In liver cells, no biologically relevant or statistically significant increase in the level of DNA migration was noted up to 500 mg/kg/day on Day 28 and there were no histologic findings at this dose. At 1000 mg/kg/day, there was a slight statistically significant increase due to two male rats that showed small increases in the DNA migration levels. However, histology results revealed single cell and focal necrosis in the liver of these rats. Therefore, the small increases in DNA migration levels observed were not considered biologically relevant because the effect of the hepatocellular necrosis was a confounding effect. This assay demonstrates that acetaminophen does not cause DNA damage in peripheral blood leukocytes at doses up to 1000 mg/kg/day or liver cells at doses up to 500 mg/kg/day.

Toxicity Evaluations

Acetaminophen was administered via oral gavage once daily to a separate toxicity groups of male Sprague-Dawley rats as an aqueous suspension in demineralized water containing 0.5% w/v Methocel (hydroxylpropyl methylcellulose) at 0 and 1000 mg/kg body weight/day (mg/kg/day) for 7 and 28 days.

No mortality occurred, and no clinical abnormalities were observed during the dosing period. Dosing at 1000 mg/kg/day led to a decrease in body weight and/or body weight gain during the entire dosing period and reduced food consumption during the first week of dosing. The haematology data indicated that the findings on Day 28 in the toxicity rats were comparable to those in the genotoxcity rats. Although red blood cell counts were comparably reduced from Day 7 onwards, the strongest reduction in haematocrit and hemoglobin was detected on Day 7, while the mean cell volume increased from Day 15 onwards. In addition, reticulocytes were also clearly reduced on Day 7, after which their levels rose again to a marginal elevation on Day 15.

There was a slight to moderate test article related effect on liver function at 1000 mg/kg/day, evidenced by the elevations in bilirubin, AST, ALT, GLDH, SDH and total bile acids, at one or more time points during this study. The marginal to slight increases in total protein, albumin and cholesterol at 1000 mg/kg/day at one or more time points also suggest a potential minor effect on liver function. At 1000 mg/kg/day for 1 week or 1 month resulted in a slight increase in absolute and/or relative (% body weight) liver and kidney weights, gastric distention (both time-points) and small/soft testes correlating to moderate atrophy (after 1 month). At histology, minimal to slight liver changes were observed in the centrilobular region of 5/5 rats after 1 week and 2/5 rats after 1 month. The affected rats presented one or more of the following findings: centrilobular chronic inflammation or fibrosis (with some bridging), apoptosis/single cell necrosis, brown pigmented macrophages, hepatocellular vacuolization, dense staining cytoplasm or hydropic degeneration. Centrilobular regeneration (dark/irregular/bi-nucleated hepatocytes) was noted in some rats after 1 week.

---

[94] ICH S2 (R1) Genotoxocity testing and data interpretation for pharmaceuticals intended for human use. Published 01/12/2011. Reference number CHMP/ICH/126642/08.

[95] OECD. Test No 481: Genetic Toxicology: Saccharomyces cerevisiae, Mitotic Recombination Assay, OECD Guidelines for the Testing of Chemicals, Section 4, OECD Publishing. Paris. https://doi.org/10.1787/9789264071421-en

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

In the genotoxicity portion of the study, dosing at 500 mg/kg/day resulted in bone marrow toxicity but did not result in any histological liver findings. However, there were effects on liver biomarkers, specifically increases in bilirubin, ALT, GLDH and total bile acids, were observed as marginal increases at 500 or 1000 mg/kg/day.

Conclusion

This study provides *in vivo* data demonstrating an absence of genotoxic potential for acetaminophen.

CLOSING

In closing, we would like to emphasize that acetaminophen plays a significant role for US consumers seeking symptomatic relief. FDA[96] has acknowledged the importance of acetaminophen in effectively relieving pain and fever. In prescription medicines, acetaminophen is administered with other active ingredients to treat moderate to severe pain. OTC acetaminophen effectively treats a wide variety of minor pain, including toothache, muscle aches, headache, backache, menstrual cramps and osteoarthritis. Notably, acetaminophen represents an important pain relief option for patients with certain medical conditions (e.g., asthma, heart disease, stomach ulcers, bleeding problems, or kidney disease). Accordingly, acetaminophen is identified as either a first-line or preferred OTC pain relief option by groups representing those patients (e.g., American Heart Association[97], National Kidney Foundation[98], American Geriatrics Society[99] and the American College of Gastroenterology[100]). Its importance as a first-line therapy in treating osteoarthritis pain cannot be overstated. Many professional organizations, including the American College of Rheumatology[101] and the Osteoarthritis Research Society International[102] recommend acetaminophen for treating osteoarthritis.

We hope the information provided herein will prove helpful to OEHHA in developing the HIM for acetaminophen. If OEHHA experiences any difficulty in obtaining these documents, CHPA would be pleased to assist.

---

[96] U.S. Food and Drug Administration. Acetaminophen Information. https://www.fda.gov/drugs/information-drug-class/acetaminophen-information. Accessed May 17, 2019.

[97] Antman EM, Bennett JS, Daugherty A, et al. (2007) Use of nonsteroidal antiinflammatory drugs: an update for clinicians: a scientific statement from the American Heart Association. Circulation.115:1634-1642.

[98] https://www.kidney.org/atoz/content/painmeds_analgesics Accessed 5/19, 2019.

[99] American Geriatrics Society. (2009) Pharmacological management of persistent pain in older persons. J Am Geriatr Soc. 57:1331-1346.

[100] Caufield SP, Schafer TW. Peptic ulcer disease. American College of Gastroenterology Web site. http://patients.gi.org/topics/peptic-ulcer-disease. Updated 2012. Accessed 5/19/19.

[101] American College of Rheumatology (2012) Recommendations for the Use of Nonpharmacologic and Pharmacologic Therapies in Osteoarthritis of the Hand, Hip, and Knee. https://www.rheumatology.org/Portals/0/Files/ACR%20Recommendations%20for%20the%20Use%20of%20Nonpharmacologic%20and%20Pharmacologic%20Therapies%20in%20OA%20of%20the%20Hand,%20Hip%20and%20Knee.pdf. Accessed May 17, 2019.

[102] McAlindon, T.E., Bannuru, R.R., Sullivan, M.C. et al. (2014) OARSI guidelines for the non-surgical management of knee osteoarthritis. Osteoarthritis Cartilage. 22: 363-388.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Sincerely,

Barbara A. Kochanowski, Ph.D.
Sr. Vice President, Regulatory & Scientific Affairs

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

CHPA SUBMISSION TO OEHHA

September 20, 2011

Members of the Carcinogen Identification Committee

Ms. Cynthia Oshita (*via email)*
Office of Environmental Health Hazard Assessment (OEHHA)
Proposition 65 Implementation
P.O. Box 4010
1001 I Street, 19th Floor
Sacramento, CA 95812-4010

Re: Prioritization of Acetaminophen

Dear Chairperson Mack and Members of the Carcinogen Identification Committee

The Consumer Healthcare Products Association (CHPA) requests the Proposition 65 Carcinogen Identification Committee (CIC) recommend a "low priority" for the review of acetaminophen. CHPA, founded in 1881, is a member-based association representing the leading manufacturers and distributors of nonprescription, over-the-counter (OTC) medicines and dietary supplements.

Summary

Acetaminophen should be assigned a "low priority" for several reasons:

- There is no convincing epidemiologic evidence of a link between acetaminophen exposure and cancer in humans.
- There is limited evidence of carcinogenicity of acetaminophen in animals.
- No new convincing evidence has been presented since OEHHA prioritized acetaminophen as "low" priority in 1997.
- Since the 1997 prioritization, FDA has reviewed and approved a New Drug Application for OFIRMEV™, a prescription only intravenous formulation of acetaminophen. This approval included a review of carcinogenicity data that indicated no (based on mice and male rats) or equivocal (based on female rats) evidence of carcinogenic activity.
- The public health benefits of acetaminophen in treating pain and fever safely and effectively are well recognized.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Introduction and Background

On October 13, the CIC will consider the prioritization of 39 chemicals identified by OEHHA in the Notice entitled, "Prioritization: Chemicals for Consultation by the Carcinogen Identification Committee" dated July 22, 2011 (OEHHA Notice). To identify these 39 chemicals, OEHHA conducted a preliminary toxicological evaluation for the chemicals discussed at 2007 and 2008 meetings of the CIC. The OEHHA Notice provided an exposure assessment, along with human data, animal data and other relevant data evaluating carcinogenicity potential. OEHHA compiled a summary of the relevant studies that were identified during the preliminary toxicological evaluation.

The following sections of this submission briefly summarize the available data relevant to an assessment of the potential carcinogenicity of acetaminophen, following the general categories in the summary table in the OEHHA Notice. In addition, a summary of a recent review of the potential carcinogenicity of acetaminophen conducted by the FDA prior to the approval of a prescription acetaminophen product is also provided. When all of the evidence is considered, it is apparent that acetaminophen should not be a priority for further review by the CIC. CHPA believes the available evidence supports a "low priority" recommendation for acetaminophen.

Human Data

In 1990 and again in 1999, the International Agency for Research on Cancer (IARC) concluded that there was inadequate evidence for the carcinogenicity of acetaminophen in humans, and that overall, acetaminophen could not be classified as to its carcinogenicity to humans (Group 3). In May 1997, OEHHA, as part of its Proposition 65 prioritization process, determined that there was a "low level of carcinogenicity concern over acetaminophen" (OEHHA, 1997). More recently, in November 2010, FDA approved a New Drug Application (NDA) for OFIRMEV™, a prescription acetaminophen intravenous formulation, with no carcinogenicity concerns identified.

The Summary Table in the OEHHA Notice prioritization document correctly indicates there are analytic epidemiology studies that included acetaminophen. Appendix A in the OEHHA assessment identifies a number of case-control studies and cohort studies published since May 1997 that included acetaminophen, often as one of many exposures evaluated in relation to cancer. Results for epidemiologic studies included in Appendix A were inconsistent for all types of cancers evaluated and continue to support no clear association between acetaminophen use and development of cancer. While some studies reported an increased risk with acetaminophen use, most studies reported no increased risk.
It should be noted that because of the observational nature of cohort and case-control epidemiologic studies, methodologic concerns exist, including recall bias and confounding by indication. The latter is of particular concern for acetaminophen, since acetaminophen is often recommended to those who are ill for the relief of pain of their developing and undiagnosed condition (*e.g.*, cancer) and may be more likely to be falsely associated with that condition.

Most epidemiologic studies in the July 2011 OEHHA Appendix A reported that acetaminophen use was *not* associated with an increased risk of cancer and have included studies of the following cancer types: ovarian, breast, endometrial, colon/colorectal, renal, bladder, urothelial, prostate, non-Hodgkin's lymphoma, leukemia, esophageal, and gliobalstoma multiforme. In a few studies, a protective effect of acetaminophen use, *i.e.*, a reduced risk of cancer, was reported. Most recently, a study examining the relationship between analgesic use and renal cell cancer risk in the Nurses' Health Study and the Health Professionals Follow-Up Study reported no association between acetaminophen use and renal cell cancer risk over 16-20 years (Cho, 2011).

In summary, a review of the studies in Appendix A and one more recently published study does not support a change in the current "low" priority level for acetaminophen.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Animal Data

OEHHA last reviewed the epidemiologic, animal carcinogenicity and mutagenicity data for acetaminophen in 1997. The OEHHA determination was based in part on the extensive IARC (1990) review of the available literature. The 1990 IARC concluded that there was limited evidence for carcinogenicity of paracetamol [acetaminophen] in experimental animals and inadequate evidence of carcinogenicity of paracetamol in humans. Since the 1997 review of acetaminophen by OEHHA, two additional scientific reviews have occurred. Bergman *et al* (1996) conducted an extensive review of the carcinogenicity and mutagenicity of acetaminophen, reaching similar conclusions to the IARC and OEHHA determinations. In 1999 IARC again reviewed acetaminophen, including the results of the National Toxicology Program Bioassays in rats and mice (NTP, 1993), as well as other laboratory-based and epidemiologic studies. This IARC assessment concluded that there was inadequate evidence in humans and in experimental animals for the carcinogenicity of paracetamol.

Genotoxicity

Since acetaminophen was first marketed in the United States in 1953, it has been extensively evaluated for potential genotoxicity using both *in vitro* and *in vivo* assays with a variety of endpoints. This research used many different testing models and included human and rodent primary cell cultures and cell lines. Regulatory and peer reviewed published data support lack of mutagenicity; however, the assays measuring chromosomal damage present mixed results. These inconsistent results indicate that acetaminophen is not directly DNA damaging; rather, it is a secondary effect to toxicity (Djordjevic *et al.* 1986, Brunborg *et al.* 1995, Oshida *et al.* 2008). More recently (Nov 2010), FDA's review of a new submission, OFIRMEV™ (acetaminophen) for IV injection resulted in no concerns regarding genotoxicity which further supports assignment of acetaminophen of as "low priority."

Recent Authoritative Body Review of Acetaminophen

Within the last year, the Food and Drug Administration (FDA) reviewed and approved (November 2010) a New Drug Application for OFIRMEV™, a prescription only, intravenous formulation of acetaminophen. This included an extensive review of available data including animal carcinogenicity studies that indicated no (based on mice, male rats) or equivocal (based on female rats) evidence of carcinogenic activity. This finding was endorsed by the FDA's Executive Carcinogenicity Assessment Committee. Conclusions regarding mutagenicity data supported previously reported findings and were mixed (Ames negative, threshold effect in clastogenicity testing and positive results in *in vitro* mouse lymphoma and *in vitro* chromosomal aberration assay using human lymphocytes). This recent and comprehensive review of scientific data by FDA supports a low priority for acetaminophen.

Exposure and Public Health Importance

OEHHA has correctly identified "widespread exposure" to describe acetaminophen. This is not surprising, given the well-recognized public health benefits of acetaminophen in treating fever and pain. Acetaminophen is the most commonly used drug ingredient in the U.S. In any given week, nearly 25% of adults report using an acetaminophen-containing product, including over-the-counter single-ingredient and combination products as well as prescription (Rx) narcotic/acetaminophen combination medicines (Kaufman 2002). FDA (2009) has acknowledged the importance of acetaminophen in effectively relieving pain and fever. Over-the-counter (OTC) acetaminophen effectively treats a wide variety of pain, including dental, muscle, headache, menstrual cramps and osteoarthritis. Its importance as first-line therapy in treating osteoarthritis pain cannot be overstated. Many professional organizations, including the American College of Rheumatology (2000), the Osteoarthritis Research Society International (Zhang 2008), and the Agency for Healthcare Research and Quality (2009) recommend acetaminophen as first-line therapy for treating osteoarthritis.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Conclusion

When all of the evidence is considered, it is apparent that acetaminophen should not be a priority for further review by the CIC. There is no convincing human data to support carcinogenicity. FDA's recent review of a prescription acetaminophen product did not identify any concerns regarding the carcinogenicity of acetaminophen. There is limited evidence of carcinogenicity of acetaminophen in animals. Human exposure to acetaminophen is widespread, and there is good reason for it. It provides a significant public health benefit. The well-recognized public health benefits should not be disregarded. CHPA believes the available evidence supports a "low priority" recommendation for acetaminophen.

We appreciate the opportunity to submit these comments. We look forward to participating in the October 12-13, 2011, CIC meeting.

Sincerely,

Barbara A. Kochanowski, Ph.D.
Vice President, Regulatory & Scientific Affairs

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

References

Agency for Healthcare Research and Quality. (2007) Choosing Non-Opioid Analgesics for Osteoarthritis. AHRQ Publication No. 06(07)-EC009-3.; available at: http://effectivehealthcare.ahrq.gov/repFiles/Osteoarthritis_Clinician_Guide.pdf. Accessed: September 11, 2011.

American College of Rheumatology Subcommittee on Osteoarthritis Guidelines. (2000) Recommendations for the medical management of osteoarthritis of the hip and knee: 2000 update. Arthritis Rheum. 43:1905-1915.

Arnold B.F., Ercumen A., Benjamin-Chung J., Coford J.M. Jr. (2016) Brief Report: Negative controls to detect selection bias and measurement bias in epidemiologic studies. Epidemiology. 27(5):637-41. doi: 10.1097/EDE.0000000000000504.

Barnard M.E., Poole E.M., Curhan G.C., Eliassen A.H., Rosner B.A., Terry K.L., Tworoger S.S. (2018) Association of analgesic use with risk of ovarian cancer in the Nurses' Health Studies. JAMA Oncol. 4(12):1675-82.

Bergman, K., Muller, L. and Teigen, S.W. (1996) Series: current issues in mutagenesis and carginogenesis, No. 65. The genotoxicity and carcinogenicity of paracemamol: a regulatory (re)review. Mutation Res. 349: 263-268.

Bertrand K.A., Bethea T.N., Gerlovin H., et al. (2018) Aspirin use and estrogen receptor-negative breast cancer risk in African American women. Cancer Res. 78(13). doi: http://dx.doi.org/10.1158/1538-7445.AM2018-5247.

Bittoni M., Carbone D., Harris R. (2017) Chronic inflammation, NSAIDs and the risk of lung cancer death. J Thoracic Oncol. 12(1):S257.

Blanco, M., Urios, A., and Martínez A. (1998) New Escherichia coli WP2 tester strains highly sensitive to reversion by oxidative mutagens. Mutat Res. 413(2):95-101.

Brennan R.J. and Schiestl R.H. (1997) Aniline and its metabolites generate free radicals in yeast. Mutagenesis. 12: 215-220.

Brown L.M., Gridley G., Check D., Landgren O. (2008) Risk of multiple myeloma and monoclonal gammopathy of undetermined significance among white and black male United States veterans with prior autoimmune, infectious, inflammatory, allergic disorders. Blood. 111:3388–94.

Brunborg, G., Holme, J.A., and Hongslo, J.K. (1995) Inhibitory effects of paracetamol on DNA repair in mammalian cells. Mutat Res. 342: 157-170.

Chikura S., Kimoto T., Itoh S., Sanada H., Muto S., Horibata K. (2019) Standard protocol for the total red blood cell Pig-a assay used in the interlaboratory trial organized by the Mammalian Mutagenicity Study Group of the Japanese Environmental Mutagen Society. Genes Environ. 41:5. https://doi.org/10/1186/s41021-019-0121-z.

Cho, E., Curhan, G., Hankinson, S.E., Kantoff, P., Atkins, M.B., Stampfer, M. and Choueiri, T.K. (2011) Prospective evaluation of analgesic use and risk of renal cell cancer. Arch. Intern. Med. 171(16); 1487-93.

Choueiri T.K., Je Y., Cho E. (2014) Analgesic use and the risk of kidney cancer: A meta-analysis of epidemiologic studies. Int J Cancer. 134(2):384-96.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Couto A.C., Ferreira J.D., Pombo-de-Oliveira M.S., Koifman S. (2015) Pregnancy, maternal exposure to analgesic medicines, and leukemia in Brazilian children below 2 years of age. Eur J Cancer Prev. 24(3):245-252.

Chow W.H., McLaughlin J.K., Linet M.S., Niwa S., Mandel J.S.. (1994) Use of analgesics and risk of renal cell cancer. Int J Cancer. 59:467–470.

Clarke C.A., Canchola A.J., Moy L.M., Neuhausen S.L., Chung N.T., Lacey J.V. Jr, Bernstein L. (2017) Regular and low-dose aspirin, other non-steroidal anti-inflammatory medications and prospective risk of HER2-defined breast cancer: The California Teachers Study. Breast Cancer Res. 19(1):52.

de Pedro M., Baeza S., Escudero M., Dierssen-Sotos T., Gómez-Acebo I., Pollán M., Llorca J. (2015) Effect of COX-2 inhibitors and other non-steroidal inflammatory drugs on breast cancer risk: A meta-analysis. Breast Cancer Res Treat. 149(2):525-536.

Ding Y.Y., Yao P., Verma S., Han Z.K., Hong T., Zhu Y.Q., Li H.X.. (2017) Use of acetaminophen and risk of endometrial cancer: evidence from observational studies. Oncotarget. 8(21):34643-34651.

Djordjevic, S.P., Nicholas, H. K., and Lavin, M.F., Martin, L.F. (1996) Effect of N-hydroxyparacetamol on cell cycle progression. Biochem Pharmacol. 35(20): 3511-3516.

Egan K.M., Nabors L.B., Thompson Z.J., Rozmeski C.M., Anic G.A., Olson J.J., LaRocca R.V., Chowdhary S.A., Forsyth P.A., Thompson R.C. (2016) Analgesic use and the risk of primary adult brain tumor. Eur J Epidemiol. 31(9):917-25.

Erickson P., Gardner L.D., Loffredo C.A., George D.M., Bowman E.D., Deepak J., Mitchell K., Meaney C.L., Langenberg P., Bernat D.H., Amr S. (2018) Racial and ethnic differences in the relationship between aspirin use and non–small cell lung cancer risk and survival. Cancer Epidemiol Biomarkers Prev. 27(12):1518-26.

Friel G., Liu C.S., Kolomeyevskaya N.V., Hampras S.S., Kruszka B., Schmitt K., Cannioto R.A., Lele S.B., Odunsi K.O., Moysich K.B. (2015) Aspirin and acetaminophen use and the risk of cervical cancer. J Low Genit Tract Dis. 19(3):189-193. doi: http://dx.doi.org/10.1097/LGT.0000000000000104.

Hannibal C.G., Dehlendorff C., Kjaer S.K. (2018) Use of paracetamol, low-dose aspirin, or non-aspirin non-steroidal anti-inflammatory drugs and risk of ovarian borderline tumors in Denmark. Gynecol Oncol. 151(3):513-8.

Heard K., Anderson V.E., Dart R.C., Green J.L. (2016) Accuracy of the structured Medication History Assessment Tool (MedHAT) compared with recorded real-time medication use. Pharmacotherapy. 36(5):496-504.

Hill A.B. (1965) The environment and disease: Association or causation? Proc R Soc Med. 58:295-300.

Ibrulj S., Rahmanovic A., Haveric S., Haveric A., Pasic A.D. (2007) Cytogenetic evaluation of paracetamol effects in human lymphocytes culture. Drug Chem Toxicol. 30: 133-143.

ICH S2 (R1). Genotoxocity testing and data interpretation for pharmaceuticals intended for human use. Published 01/12/2011. Reference number CHMP/ICH/126642/08.

International Agency for Research on Cancer (IARC, 1990) IARC Monographs on the Evaluation of Carcinogenic Risks to Humans: Pharmaceutical Drugs. Vol. 50, pp. 307 -332, Lyon, France.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

International Agency for Research on Cancer (IARC, 1999) IARC Monographs on the Evaluation of Carcinogenic Risks to Humans: Paracetamol. Vol. 73, pp. 401-449, Lyon, France.

Kanki K., Nishikawa A., Masumura K., Umemura T., Imazawa T., et al. (2005) In vivo mutational analysis of liver DNA in gpt delta transgenic rats treated with the hepatocarcinogens N-nitrosopyrrolidine, 2-amino-3-methylimidazo[4,5-f]quinoline, and di(2-ethylhexyl)phthalate. Mol Carcinog. 42: 9-17.
..
Karami S., Daughtery S.E., Schwartz K., Davis F.G., Ruterbusch J.J., Wacholder S., Graubard B.I., Berndt S.I., Hofmann J.N., Purdue M.P., Moore L.E., Colt J.S. (2016) Analgesic use and risk of renal cell carcinoma: A case-control, cohort and meta-analytic assessment. Int J Cancer. 139(3):584-92.

Kaufman DW, Kelly, J.P., Rosenberg, L., Anderson, T.E. and Mitchell, A.A. (2002) Recent patterns of medication use in the ambulatory adult population of the United States: the Slone survey. JAMA 287(3): 337-44.

Kho P.F., Fawcett J., Fritschi L., Risch H., Webb P.M., Whiteman D.C., Neale R.E. (2016) Nonsteroidal anti-inflammatory drugs, statins, and pancreatic cancer risk: a population-based case-control study. Cancer Causes Control. 27(12):1457-1464.

Kirkland D.J., Dresp J.H., Marshall R.R., Baumeister M., Gerloff C., Gocke E. (1992) Normal chromosomal aberration frequencies in peripheral lymphocytes of healthy human volunteers exposed to a maximum daily dose of paracetamol in a double blind trial. Mutat Res. 279(3):181-94.

Klopcic I., Poberznik M., Mavri J., Dolenc M.S. (2015) A quantum chemical study of the reactivity of acetaminophen (paracetamol) toxic metabolite N-acetyl-p-benzoquinone imine with deoxyguanosine and glutathione. Chemico-Biological Interactions. 242: 407-414.

Lindqvist E.K., Goldin L.R., Landgren O., Blimark C., Mellqvist U.H., Turesson I., Wahlin A., Björkholm M., Kristinsson S.Y. (2011) Personal and family history of immune-related conditions increase the risk of plasma cell disorders: a population-based study. Blood. 118(24):6284-6291.

Lindqvist E.K., Landgren O., Lund S.H., Turesson I., Hultcrantz M., Goldin L., Björkholm M., Kristinsson S.Y. (2017) History of autoimmune disease is associated with impaired survival in multiple myeloma and monoclonal gammopathy of undetermined significance: a population-based study. Ann Hematol. 96(2):261-269.

Lipsitch M., Tchetgen Tchetgen E., Cohen T. (2010) Negative controls: a tool for detecting confounding and bias in observational studies. Epidemiology. 21(3):383-8. doi: 10.1097/EDE.0b013e3181d61eeb.

Liu C., Friel G., Kolomeyevskaya N.V., Hampras S.S., Kruszka B., Schmitt K., Cannioto R.A., Lele S.B., Odunsi K.O., Moysich K.B. (2014) Aspirin and acetaminophen decrease the risk of cervical cancer in long-term users. Gynecol Oncol. 133:42-43. doi: http://dx.doi.org/10.1016/j.ygyno.2014.03.123.

Madigan D., Schuemie M.J., Ryan P.B. (2013) Empirical performance of the case-control method: lessons for developing a risk identification and analysis system. Drug Saf. 36 Suppl 1:S73-82. doi: 10.1007/s40264-013-0105-z.

Markovic D., Katic J., Stojkovic R., Borovic S., Zarkovic N., Fucic A. (2013) Lipid peroxidation, detoxification capacity, and genome damage in mice after transplacental exposure to pharmaceutical drugs. Braz J Med Biol Res. 46: 1014-1020.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Martinez A., Urios A., Blanco M. (2000) Mutagenicity of 80 chemicals in Escherichia coli tester strains IC203, deficient in OxyR, and its oxyR(+) parent WP2 uvrA/pKM101: detection of 31 oxidative mutagens. Mutat Res. 467: 41-53.

Matsushima T., Hayashi M., Matsuoka A., Ishidate M. Jr, Miura K.F., Shimizu H., Suzuki Y., Morimoto K., Ogura H., Mure K., Koshi K., Sofuni T. (1999) Validation study of the in vitro micronucleus test in a Chinese hamster lung cell line (CHL/IU). Mutagenesis. 14: 569-80.

McCredie M., Stewart J.H., Day N.E. (1993) Different roles for phenacetin and paracetamol in cancer of the kidney and renal pelvis. Int J Cancer. 53:245–249.

McCredie M., Pommer W., McLaughlin J.K., Stewart J.H., Lindblad P., Mandel J.S., Mellemgaard A., Schlehofer B., Niwa S. (1995) International renal-cell cancer study. II. Analgesics. Int J Cancer. 60:345–349

McGill M.R., Sharpe M.R., Williams C.D., Taha M., Curry S.C., Jaeschke H.J. (2012) The mechanism underlying acetaminophen-induced hepatotoxicity in humans and mice involves mitochondrial damage and nuclear DNA fragmentation. Clin Invest. 122(4):1574-83.

McGlynn K.A., Hagberg K., Chen J., Graubard B.I., London W.T., Jick .S, Sahasrabuddhe V.V. (2015) Statin use and risk for primary liver cancer in the clinical practice research datalink. J Natl Cancer Inst. 107(4):Article No.: djv009.

Mellemgaard A., Niwa S., Mehl E., Engholm G., McLaughlin J.K., Olsen J.H.. (1994) Risk factors for renal cell carcinoma in Denmark: Role of medication and medical history. Int J Epidemiol. 23:923–930.

National Toxicology Program (NTP, 1993) Toxicology and Carcinogenesis Studies of Acetaminophen (Cas No. 103-90-2) in F344 Rats and B6C3F1 Mice (Feed Studies). NTP Technical Report Series No. 394. National Institutes of Health, Public Health Service, U.S. Department of Health and Human Services.

OEHHA, Prioritized Candidate Chemicals Under Consideration for Carcogenicity Evaluation: Batch #1 Office of Environmental Health Hazard Assessment California Environmental Protection Agency (May 1997) Accessed from http://oehha.ca.gov/prop65/pdf/batch1.pdf on September 16, 2011.

OECD. Test No. 474: Mammalian Erythrocyte Micronucleus Test, 1997. OECD Publishing, Paris. https://doi.org/10.1787/9789264071285-en.

OECD. Test No 481: Genetic Toxicology: Saccharomyces cerevisiae, Mitotic Recombination Assay, OECD Guidelines for the Testing of Chemicals, Section 4, OECD Publishing. Paris. https://doi.org/10.1787/9789264071421-en.

OECD. Test No. 487: Guideline for the testing of chemicals, , In vitro mammalian cell micronucleus Test, 2016. http://www.oecd-ilibrary.org/environment/test-no-487-in-vitro-mammalian-cell-micronucleus-test_9789264264861-en.

OECD. Test No. 489: In Vivo Mammalian Alkaline Comet Assay, OECD Guidelines for the Testing of Chemicals, Section 4, 2016. OECD Publishing, Paris, https://doi.org/10.1787/9789264264885-en.

OEHHA Prioritization: Chemicals For Consultation By The Carcinogen Identification Committee [07/22/11] http://www.oehha.ca.gov/prop65/public_meetings/CIC101211/101211ChemPriorCIC2011.pdf and http://www.oehha.ca.gov/prop65/public_meetings/prior072211.html. Accessed September 11, 2011.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Oshida, K., Iwanaga, E., Miyamoto-Kuramitsu, K., and Miyamoto, Y. (2008) An in vivo comet assay of multiple organs (liver, kidney and bone marrow) in mice treated with methyl methanesulfonate and acetaminophen accompanied by hematology and/or blood chemistry. J Toxicol Sci. 33(5): 515-524.

Pandeya N., Olsen C.M., Thompson B.S., Dusingize J.C., Neale R.E., Green A.C., Whiteman D.C.; QSkin Study. (2019) Aspirin and nonsteroidal anti-inflammatory drug use and keratinocyte cancers: a large population-based cohort study of skin cancer in Australia. Brit J Dermatol. doi: 10.1111/bjd.17938.

Peres L.C., Camacho F., Abbott S.E., Alberg A.J., Bandera E.V., Barnholtz-Sloan J., Bondy M., Cote M.L., Crankshaw S., Funkhouser E., Moorman .P.G, Peters E.S., Schwartz A.G., Terry P., Wang F., Schildkraut J.M. (2016) Analgesic medication use and risk of epithelial ovarian cancer in African American women. Br J Cancer. 114(7):819-25.

Preston M.A., Zhang X., Graff R.E., Sanchez A., Chang S.L., Meir J., Stampfer T., Choueiri K., Wilson K.M., Cho E. (2016) Analgesic use and risk of renal cell cancer: Results from two prospective cohort studies. J Clin Oncol. 34(2_suppl):588-588. (abstract)

Rogers L.K., Moorthy B., Smith C.V. (1997) Acetaminophen binds to mouse hepatic and renal DNA at human therapeutic doses. Chem Res Toxicol 10: 470-476.

Ryan P.B., Madigan D., Stang P.E., Overhage J.M., Racoosin J.A., Hartzema A.G. (2012) Empirical assessment of methods for risk identification in healthcare data: results from the experiments of the Observational Medical Outcomes Partnership. Stat Med. 31(30):4401-15. doi: 10.1002/sim.5620.

Ryan P.B., Schuemie M.J. (2013) Evaluating performance of risk identification methods through a large-scale simulation of observational data. Drug Saf. 36 Suppl 1:S171-80. doi: 10.1007/s40264-013-0110-2.

Ryan P.B., Stang P.E., Overhage J.M., Suchard M.A., Hartzema A.G., DuMouchel W., Reich C.G., Schuemie M.J., Madigan D. (2013) A comparison of the empirical performance of methods for a risk identification system. Drug Saf. 36 Suppl 1:S143-58. doi: 10.1007/s40264-013-0108-9.

Schuemie M.J., Gini R., Coloma P.M., Straatman H., Herings R.M., Pedersen L., Innocenti F., Mazzaglia G., Picelli G., van der Lei J., Sturkenboom M.C. (2013) Replication of the OMOP experiment in Europe: evaluating methods for risk identification in electronic health record databases. Drug Saf. 36 Suppl 1:S159-69. doi: 10.1007/s40264-013-0109-8.

Schuemie M.J., Hripcsak G., Ryan P.B., Madigan D., Suchard M.A. (2018) Empirical confidence interval calibration for population-level effect estimation studies in observational healthcare data. Proc Natl Acad Sci U S A. 115(11):2571-2577. doi: 10.1073/pnas.1708282114.

Schuemie M.J., Ryan P.B., DuMouchel W., Suchard M.A., Madigan D. (2014) Interpreting observational studies: why empirical calibration is needed to correct p-values. Stat Med. 33(2):209-18. doi: 10.1002/sim.5925.

Simkó M., Kriehuber R., Lange S. (1998) Micronucleus formation in human amnion cells after exposure to 50 Hz MF applied horizontally and vertically. Mutation Research/Genetic Toxicology and Environmental Mutagenesis. 418: 101-111.

Singh S., Singh P.P., Roberts L.R., Sanchez W. (2014) Chemopreventive strategies in hepatocellular carcinoma. Nat Rev Gastroenterol Hepatol. 11(1):45-54. doi: 10.1038/nrgastro.2013.143.

Suzuki Y., Goto K., Nakayama Y., Saratani M., Takata T., Okamoto T., Okazaki S. Evaluation of a single-dose PIGRET assay for acetaminophen in rats compared with the RBC Pig-a assay. S. Mutat Res. 2016;811:16-20.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

Tchetgen Tchetgen E. (2014) The control outcome calibration approach for causal inference with unobserved confounding. Am J Epidemiol. 179(5):633-40. doi: 10.1093/aje/kwt303.

Trabert B., Poole E.M., White E., Visvanathan K., Adami H.O., Anderson G.L., Brasky T.M., Brinton L.A., Fortner R.T., Gaudet M., Hartge P., Hoffman-Bolton J., Jones M., Lacey J.V., Larsson S.C., Mackenzie G.G., Schouten L.J., Sandler D.P., O'Brien K., Patel A.V., Peters U., Prizment A., Robien K., Setiawan V.W., Swerdlow A., van den Brandt P.A., Weiderpass E., Wilkens L.R., Wolk A., Wentzensen N., Tworoger S.S.; Ovarian Cancer Cohort Consortium (OC3). (2019) Analgesic use and ovarian cancer risk: An analysis in the Ovarian Cancer Cohort Consortium. J Natl Cancer Inst. 111(2):137-145. doi: 10.1093/jnci/djy100.

Tweats D.J., Blakey D., Heflich R.H., Jacobs A., Jacobsen S.D., Morita T., Nohmig T., O'Donovan M.R., Sasaki Y.F., Sofuni T., Ticek R. (2007) Determination of genetic toxicity and potential carcinogenicity in vitro--challenges post the Seventh Amendment to the European Cosmetics Directive. Mutagenesis. 627:78-91.

U.S. Food and Drug Administration. (2009) June 29-30, 2009: Joint Meeting of the Drug Safety and Risk Management Advisory Committee with the Anesthetic and Life Support Drugs Advisory Committee and the Nonprescription Drugs Advisory Committee: Meeting Announcement. http://www.fda.gov/AdvisoryCommittees/Calendar/ucm143083.htm. Accessed September 11, 2011.

U.S. Food and Drug Administration. (2010) Drug approval package for Ofirmev (acetaminophen) injection, 10 mg/mL, Cadence Pharmaceuticals, Approval Date 11/02/2010. http://www.accessdata.fda.gov/drugsatfda_docs/nda/2010/022450_ofirmev_toc.cfm. Accessed September 11, 2011.

Veitonmäki T., Murtola T.J., Määttänen L., Taari K., Stenman U.H., Tammela T.L., Auvinen A. (2014) Prostate cancer risk and nonsteroidal antiinflammatory drug use in the Finnish prostate cancer screening trial. Br J Cancer. 111(7):1421-31.

Webb P.M., Na R., Weiderpass E., Adami H.O., Anderson K.E., Bertrand K.A., Botteri E., Brasky T.M., Brinton L.A., Chen C., Doherty J.A., Lu L., McCann S.E., Moysich K.B., Olson S., Petruzella S., Palmer J.R., Prizment A.E., Schairer C., Setiawan V.W., Spurdle A.B., Trabert B., Wentzensen N., Wilkens L., Yang H.P., Yu H., Risch H.A., Jordan S.J. (2019) Use of aspirin, other nonsteroidal anti-inflammatory drugs and acetaminophen and risk of endometrial cancer: the Epidemiology of Endometrial Cancer Consortium. Ann Oncol. 30(2):310-316.

Weinstein R.B., Ryan P., Berlin J.A., Matcho A., Schuemie M., Swerdel J., Patel K., Fife D. (2017) Channeling in the use of nonprescription paracetamol and ibuprofen in an electronic medical records database: evidence and implications. Drug Saf. 40:127901292. doi 10.1007/s40264-017-0581-7.
.
Weiss NS. (2016) Use of acetaminophen in relation to the occurrence of cancer: a review of epidemiologic studies. Cancer Causes Control. 27(12):1411-1418.

Wright J.L., Chéry L., Holt S., Lin D.W., Luedeke M., Rinckleb A.E., Maier C., Stanford J.L. (2016) Aspirin and NSAID use in association with molecular subtypes of prostate cancer defined by TMPRSS2:ERG fusion status. Prostate Cancer Prostatic Dis. 19(1):53-6.

Yang B., Petrick J.L., Chen J., Hagberg K.W., Sahasrabuddhe V.V., Graubard B.I., Jick S., McGlynn K.A. (2016) Associations of NSAID and paracetamol use with risk of primary liver cancer in the Clinical Practice Research Datalink. Cancer Epidemiol. 43:105-11.

Zhang W, Moskowitz RW, Nuki G, et al. (2008) OARSI recommendations for the management of hip

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa

and knee osteoarthritis, Part II: OARSI evidence-based, expert consensus guidelines. Osteoarthritis Cartilage. 16:137-162.

1625 Eye Street, NW, Suite 600 | Washington, DC 20006 | T 202.429.9260 | F 202.223.6835 | chpa.org | @chpa