Quat Residue Group (QRG) comments to the Science Guidance Panel regarding the public health benefits and safety of antimicrobial quaternary ammonium compounds (QACs)

For the *Biomonitoring California Scientific Guidance Panel* meeting scheduled for March 4, 2020

The Quat Residue Group (QRG) is a science-based task force administered by Ignite Solutions, which is part of Household and Commercial Products Association (HCPA) trade association. Its members include companies that make sanitizing and disinfecting products and companies that manufacture antimicrobial quaternary ammonium compounds (QAC). The QRG was formed to conduct studies and analyses needed by US EPA for federal registration and related risk assessment activities for alkyl dimethyl benzyl ammonium chloride (ADBAC) and didecyl dimethyl ammonium chloride (DDAC) QACs.

Executive Summary

Quaternary ammonium compounds (QACs) have been widely used in a variety of household, institutional, and industrial products for decades. QACs consist of several sub-classes including but not limited to alkyl dimethyl benzyl ammonium chloride (ADBAC) QACs and didecyl dimethyl ammonium chloride (DDAC) QACs, which are registered active ingredients for use in antimicrobial products that are highly regulated by the US EPA and other regulatory agencies around the world. ADBAC and DDAC are critically important public health tools used to prevent or control the spread of pathogens associated with many serious public health diseases in environments where we live, work, learn, and play. ADBAC and DDAC have been extensively studied for safety and toxicity, environmental effects, and efficacy/performance. There is a robust database of guideline studies that have been developed and accepted by the Agencies that regulate ADBAC and DDAC and which substantiate their safe use. Key takeaways from the robust ADBAC and DDAC toxicity study database are:

- Antimicrobial QACs are not good candidates for biomonitoring for the simple reason that their mode of action is point-of-contact irritation, and there is no evidence from guideline studies to support that they cause systemic toxicity. ADBAC and DDAC are direct-acting agents, and they are not readily absorbed by the body. Biomonitoring is

intended to provide an integrated measure of internal dose that can be used to relate exposure to systemic effects. However, QAC toxicity does not relate to systemic effects.

- There are numerous high-quality guideline studies have consistently shown no specific systemic toxicity, and no evidence of mutagenicity, sensitization, endocrine disruption, or carcinogenicity.
    - These data include 10 guideline developmental and reproductive studies for ADBAC and DDAC and their results did not show evidence of developmental or reproductive effects.
- QACs have a strong potential to sorb to sediment, but they have low toxicity when bound. They will not accumulate in the environmental given that they are readily biodegradable.

Comment 1 – Antimicrobial QACs- Overview and Scope of Comments Provided

Quaternary ammonium compounds (QACs) have been widely used in a variety of household, institutional, and industrial products for decades. There are several different QAC sub-classes, and in some cases these sub-classes have very different functionality (e.g., herbicidal QACs vs non-herbicidal QACs). The comments provided by the QRG will focus on two main antimicrobial sub-classes - alkyl dimethyl benzyl ammonium chloride (ADBAC) QACs and didecyl dimethyl ammonium chloride (DDAC) QACs.

ADBAC and DDAC QACs are registered active ingredients with the US EPA and other Agencies around the world for use in antimicrobial products. These QACs are highly regulated under the US EPA Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA), the European Biocidal Products Regulation (BPR), Health Canada (HC), and the California Department of Pesticide Regulation (CA DPR). QAC antimicrobial products include household and institutional sanitizers and disinfectants, food processing sanitizers, algicides, cosmetics, and process water treatments.

Antimicrobial products, and QACs specifically, play a very important public health role in homes, schools, hospitals, and restaurants. Antimicrobial products are defined by the US EPA as: *substances or mixtures of substances used to destroy or suppress the growth of harmful microorganisms such as bacteria, viruses, or fungi on inanimate objects and surfaces.* Antimicrobials are not used in the human body like antibiotics.

Individual QACs with unique CAS numbers fall within the ADBAC sub-class (19 individual CAS numbers) and DDAC sub-class (5 individual CAS numbers) as part of the EPA FIFRA Antimicrobial registration framework. There is a large and robust mammalian and environmental toxicology

dataset that has been generated for these two sub-classes and the contents of this dataset is described further below.

The FIFRA registered ADBAC and DDAC substances differ in chain length in comparison to most other non-registered ADBAC and DDAC substances used for non-antimicrobial applications, such as softeners and cosmetic hair conditioners. There are numerous reviews of these non-antimicrobial QACs for environmental fate of fabric softeners and Cosmetic Ingredient Reviews for conditioners. Additionally, there is a big functional and mechanistic difference between the herbicidal classes and the other QAC sub-classes, which is a distinction that should be made when grouping classes and uses. The lack of commonality between the herbicidal QAC sub-class (di-quat and paraquat) and the other QAC sub-classes would be an important consideration for biomonitoring for example, because herbicidal QACs would not share common functional groups with the other sub-classes.

Comment 2 – Extensive ADBAC and DDAC Safety and Human Exposure Data Used to Support Active Ingredient Registration

ADBAC and DDAC QACs have been extensively studied for safety and toxicity, environmental effects, and efficacy/performance.

A significant number of guideline toxicology studies have been generated for ADBAC and DDAC QACs to support their registrations. Although this data is not currently published in the public literature, detailed summaries are publicly available in US EPA and EU registration documents, such as the ADBAC and DDAC US EPA Registration Review Work Plans and the EU BPR and REACH dossiers (e.g., ECHA, 2015a,b; 2019a,b; US EPA 2017a,b) (see comment 3). The Agencies who regulate ADBAC and DDAC have conducted a detailed peer review of the many toxicology studies submitted to support their registration.

The toxicology dataset that exists for both ADBAC and DDAC QACs is robust. The data are required to be of high quality, following GLP procedures, and conducted according to well-established US EPA, OECD, or EU test guidelines. These requirements must be met in order to use the studies for human health and ecological risk assessments.

The ADBAC and DDAC FIFRA database includes but is not limited to:

- 10 developmental / reproductive guideline studies showing no evidence of maternal or developmental toxicity
- Several oral repeat dose guidelines studies showing no systemic toxicity

- Inhalation repeat dose studies that demonstrated effects consistent with the QAC mode of action - direct acting irritation
- Genotoxicity and cancer studies are negative for these effects.
- Acute ecotoxicity and environmental fate data that support the EPA conclusion that ADBAC and DDAC QACs are not likely to concentrate in the aquatic environment

Since ADBAC and DDAC are antimicrobial active ingredients regulated governmental regulating agencies (US and international), the required safety reports are submitted directly to the agencies.

- **Detailed peer review** occurs directly by regulatory agency staff worldwide. Indeed, Agencies will reject any study that does not meet high data quality standards or follow proper methodology. The Agencies may not approve or might limit use of active ingredients with troublesome effects.
- Regulatory agencies typically provide public summaries of these studies (e.g., ECHA, 2015a,b; 2019a; US EPA 2017a,b)
    - Although the ADBAC and DDAC studies used to support registration are currently unpublished, manuscripts will be submitted for publication soon. The owners of the QAC safety studies are actively preparing manuscripts for publication to further make QAC safety information readily accessible to the public, health agencies, and academic researchers.

It is common practice among Agencies responsible for regulating antimicrobial products to conduct health protective risk assessments for each active ingredient for all labeled uses and application types. These conservative assessments are conducted for occupational and residential uses employ the highest permitted use rate for a particular application and require an appropriate margin of safety for each application. Cumulative risk is also part of the risk assessment process with the US EPA (see US EPA 2017a,b).

- These safety data are also supported by human subjects' exposure studies of task-specific application of antimicrobials, required by US EPA for risk assessment. These exposure data requests have been (or are being filled by) worker exposure studies conducted by the ACC Antimicrobial Exposure Assessment Joint Venture (AEATF II)[1] or consumer use studies conducted by Ignite Solutions' Antimicrobial Exposure Joint Venture (AEJV). In the worker studies, subjects' exposures to the active ingredient are monitored with multiple dosimeters while conducting specific applications of antimicrobial solutions. These studies are subject to US EPA Human Subjects Review

[1] See https://www.americanchemistry.com/Antimicrobial-Exposure-Assessment-Task-Force-II-AEATF-II-/

Board oversight. For each product use type, Unit Exposures (metrics that relay application amount to dermal, oral or inhalation exposure intake) are developed (e.g., see the current US EPA 2018[2] surrogate table of Unit Exposures).

- QACs were used as the surrogate active ingredient for many of these human subjects' exposure studies.

. [1]https://www.epa.gov/pesticide-science-and-assessing-pesticide-risks/exposure-surrogate-reference-table-pesticide-risk

Comment 3 – ADBAC/DDAC Mode of Action: Direct-acting Irritants Only

Mechanism – ADBAC and DDAC QACs are direct acting irritants and disrupt membranes of point-of-contact cells only (Gilbert and Moore, 2005; Wessels and Ingmer, 2013), e.g., which includes outer membranes and mitochondria of surface cells (Inácio et al., 2013).

- The mode of action (MOA) information database is very strong. *In vitro* studies provide evidence on the molecular initiating events (membrane disruption), while many *in vivo* animal studies are available to characterize the key events (irritation and inflammation) toward adverse outcomes at higher doses. Importantly, human *in vitro* and *in vivo* data also are available.
- For US EPA registration review, the QRG recently conducted a review of inhalation, oral and dermal toxicity studies of DDAC and ADBAC. The purpose of the review was to determine whether the EPA considered a direct acting irritation MOA in the selection of the critical effects and points of departures for use in human health risk assessments. The QRG review included a careful look at all histopathology data for effects in distant tissues available for ADBAC and DDAC.
  - The review concluded there is no evidence of cellular changes in tissues distant from the point of contact. These included high-dose, short term studies as well as subacute, subchronic and long-term toxicity studies in mice, rats, rabbits or dogs -- and from all routes of exposure, inhalation, oral and dermal. Secondary effects, such as changes in body weight were noted at high doses but are

[2]https://www.epa.gov/pesticide-science-and-assessing-pesticide-risks/exposure-surrogate-reference-table-pesticide-risk

downstream and consistent with gut irritation, inflammation and reduced food consumption.
    - The lack of findings at distant tissues is consistent with the ADME (Absorption, Distribution, Metabolism, Excretion) data, where ADBAC and DDAC have low oral absorption and very low dermal absorption, and are efficiently eliminated (US EPA 2017a,b).
    - Following dermal treatment, the primary route of exposure, no systemic toxicity was observed at any dose in long-term studies; the only effect reported was skin irritation (see US EPA 2017a,b).
- No *in situ* or systemic metabolism is relevant to ADBAC and DDAC toxicity.

Comment 4 –No Evidence of Developmental and Reproductive Effects in Repeat Dose Guideline Studies

There are 10 high quality GLP studies of antimicrobial QACs ADBAC and DDAC that follow well-established Developmental and Reproductive (DART) guidelines (OPPTS 870.3700/3800; OECD 414/416) (most are summarized in US EPA 2006a, 2006b, 2006c, 2006d, 2017a, 2017b; DPR 1996; ECHA 2015a, 2015b)

- All these studies are negative for reproductive or developmental toxicity (DART).
    - ADBAC and DDAC each have 2 rabbit and 1 rat prenatal developmental studies: developmental NOAEL consistently above highest dose tested, 3 to 20x > maternal NOAEL (Bushy Run 1989, 1991a, 1992a, 1992b; CIT 2005, 2008a)
    - ADBAC and DDAC each have 2 two-generation reproductive studies: effects do NOT occur in the absence of parental toxicity (Bushy Run 1990, 1991b; CIT 2008b, 2008c).
- These studies have been supplied to multiple regulatory authorities as part of active ingredient registration; <u>each authority provides stringent peer review of the study</u> conduct and data quality.
- The QAC manufactures who own these data have prepared manuscripts to publish ADBAC and DDAC DART studies, submission is expected soon.
- There are 3 published DART studies of QACs, 2 in mice and 1 in rats, that are negative (Fujitani et al. 2011; Palmer et al., 1983 and Momma et al., 1987).

- One set of related researchers reported DART effects in mice treated with a corrosive concentrate of mixed QACs ADBAC+DDAC in gel food (Melin et al. 2014, 2016; Hrubec et al. 2017), and whose methodology and conduct has been criticized for use of inappropriate doses, animal husbandry stresses, inadequate characterization of maternal toxicity, and poor exposure control (Hostettler, 2014, 2018).
    - The QRG views the Hrubec lab's work as anomalous to the existing database and should be viewed with caution. The study design also limits the ability to draw an association between the test material and observed adverse effects. Additionally, the study would not meet design criteria to allow for its use in human health risk assessment.
- Cal/EPA DPR, US EPA, and ECHA (EU) consistently conclude no DART concern for antimicrobial QACs (Cal/EPA -- DPR 1996; US EPA 2006a, 2006b, 2006c, 2006d, 2017a, 2017b; ECHA 2015a, 2015b).

Comment 5 - No Evidence of Endocrine Effects

In vitro work has hypothesized the potential for altered steroidogenesis by QACs due to their mitochondrial toxicity potential (Levine et al., 2007), and some have intimated that this could be a mechanism for endocrine disruption (Datta et al., 2017). However, the strong and extensive *in vivo* evidence does not support this hypothesis.

- In the dozens of studies in mice, rats, rabbits and dogs, there has been no evidence of changes to endocrine-responsive tissues following short-term or long-term dosing of antimicrobial QACs, by oral, dermal or inhalation routes.
- No reproductive effects were seen in properly conducted guideline studies.
- QACs are point-of-contact cytotoxic agents that act by membrane disruption and mitochondrial toxicity <u>to surface cells only</u> -- no cell necrosis, hyperplasia, or other cell-based change is observed other than at the point of contact (see above).
    - Direct irritation to the GI tract from very high oral doses of QACs would be expected to reduce serum cholesterol since the gut produces a significant fraction of total serum cholesterol in the body, or from altered nutritional intake.

Comment 6 - ADBAC DDAC Have Low Sensitization Potential

Antimicrobial QACs do not follow the Adverse Outcome Pathway (AOP) for dermal sensitization established and verified by multiple global agencies (OECD, 2012).

- Molecular Initiating Event (MEI, a required event in the AOP) – covalent binding to proteins – antimicrobial QACs do not do this (Gilbert and Moore, 2005; Wessels and Ingmer, 2013).
- ADBAC, DDAC and other antimicrobial QACs are not dermal sensitizers in animals (EU 2019a,b,c,d,e,f,g,h).
- Long-term, widespread safe use of QAC preservatives in cosmetics, eye drops, nasal sprays, and other medicines has not had a concern over sensitization, but some rare cases have been reported (e.g., Bernstein, 1994; Dao et al., 2012).
- No QAC antibody formation was found in exposed human subjects.

Comment 7 - Environmental profile

Full datasets of environmental fate and ecological toxicity studies for antimicrobial QACs are available, or recently requested by US EPA to fill in data gaps (US EPA 2017a,b). Ecological risk assessments are conducted for each registration review cycle (e.g., US EPA 2006f; 2017a,b).

ADBAC and DDAC are readily biodegradable

- Rapid degradation in recently conducted OECD 307 aerobic soil degradation studies (DT_{50} ~10 -15 days)
- Good degradation in OECD 308 aerobic water-sediment degradation studies (whole system DegT50s <30d)

ADBAC and DDAC are strongly adsorptive and rapidly partition to soil/sediment, making them essentially immobile and unlikely to leach or accumulate in surface or ground water (US EPA, 2017a,2017b)

- Adsorption to suspended matter, solids and soils significantly reduces toxicity of the QACs
    - Aquatic toxicity significantly reduced when suspended matter introduced into test system (US EPA, 2017a,2017b)
    - NOECs and/or EC_{50}s in terrestrial and sediment toxicity studies (earthworms, plants, microorganisms and benthic organisms) are mostly above 100 mg/kg bw indicating low toxicity (ECHA 2015a,2015b)

- High level of removal in wastewater treatment plants (both ADBAC and DDAC) (ECHA 2015a, 2015b)

Due to rapid degradation in the environment, ADBACs and DDACs are unlikely to bioconcentrate in the aquatic environment, despite being very soluble and having a strong affinity to sorb to sediment. This conclusion is supported by recent data from sediment core samples indicated that QAC levels in sediment have been decreasing appreciably since the 1980s (Patti, 2020).

Comment 8 – Public Health Importance and Unique Attributes of Antimicrobial/Disinfecting QACs

Antimicrobial QACs are a critically important public health tool to prevent or control the spread of many serious public health diseases in environments where we live, work, learn, and play. QACs are demonstrated to be efficacious against bacteria and viruses that cause colds, influenza, and food-borne illnesses[3]. Various rules and regulations prescribe or require that registered sanitizers or disinfectants be used after cleaning in hospitals, restaurants, and in food production (e.g. as part of the US Food Code[4] and US FDA FSMA[5,6] regulations or in Healthcare Infection Prevention requirements[7]). QACs are workhorse disinfectants – fit-for-purpose, safe and effective when used as directed (US EPA, 2006a,e,f; 2017a,b). QACs are efficacious at low concentrations, thus most sanitizing and disinfecting products have a very low percentage in the formula.

There are various considerations that must be made when formulating and selecting an antimicrobial product for different applications. ADBAC & DDAC QACs have unique attributes that make them very favorable as an active ingredient for use in antimicrobial products. Not all antimicrobial active ingredients are fit for each purpose or use. In selecting the appropriate active, the formulation, targeted pathogens, and projected use profile must all be taken into consideration. QACs have many benefits over other registered active ingredients such as: safety and handling, stability, formulation flexibility, high dilution potential, product performance or micro-efficacy, cost, material compatibility, and cleaning ability. Importantly, ADBAC and DDAC are not volatile and have very low dermal absorption

[3] https://www.epa.gov/pesticide-registration/selected-epa-registered-disinfectants. And see https://www.epa.gov/sites/production/files/2018-04/documents/list_g_disinfectant_list_3_15_18.pdf

[4] 2017 US FDA Food Code: https://www.fda.gov/food/retail-food-protection/fda-food-code

[5] US FDA Food Safety Modernization Act: https://www.fda.gov/food/guidance-regulation-food-and-dietary-supplements/food-safety-modernization-act-fsma

[6] Role of FSMA and importance of sanitization in food processing plants: https://foodsafetytech.com/feature_article/sanitizing-food-manufacturing-equipment-a-big-responsibility/

[7] US CDC Guidelines for Health Infection Prevention: https://www.cdc.gov/infectioncontrol/pdf/guidelines/environmental-guidelines-P.pdf

Comment 9 – Biomonitoring for QACs will not advance public health

Biomonitoring is unlikely to advance public health protection for QACs given the direct acting irritation mode of action for QACs as well as the high-quality human exposure data and conservative risk assessment methods required by regulatory agencies to support ADBAC and DDAC registrations.

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