

Studies in Experimental Animals



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Outline

Overview of studies in experimental animals

Study quality assessment

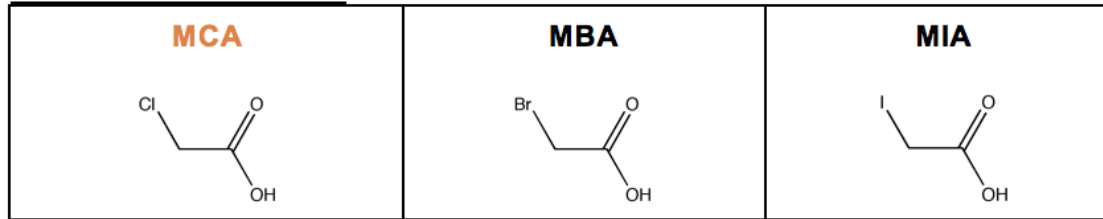
Cancer assessment

- Liver tumors
 - Tumors at other sites
 - Summary
-

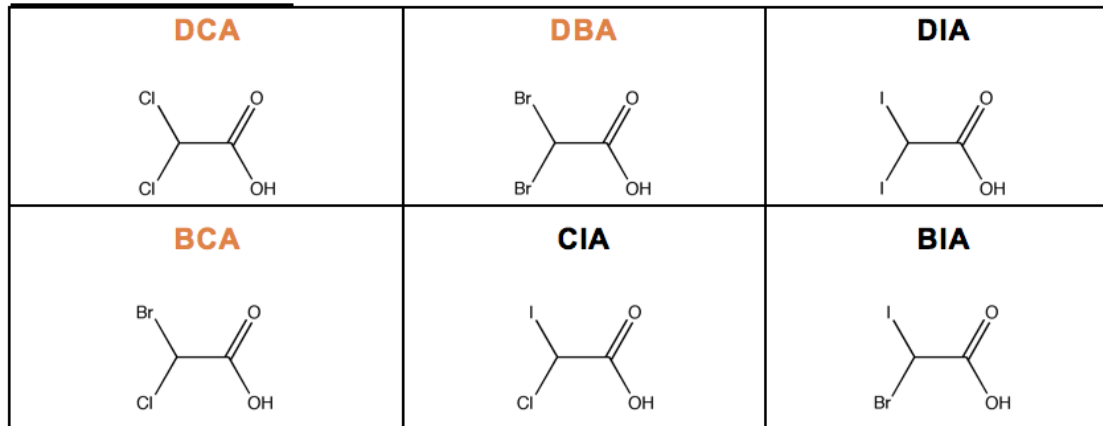


Six haloacetic acids had chronic cancer data available

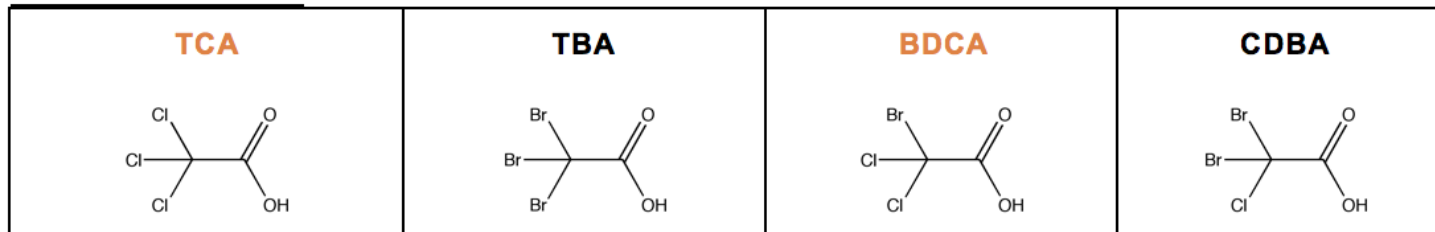
Mono-haloacetic acids



Di-haloacetic acids



Tri-haloacetic acids





- 19 publications met inclusion criteria.
 - Reported on the presence or absence of neoplastic and related lesions.
 - Had a concurrent or historical control group.
 - Chronic studies with a duration of 12 months or greater or were studies with transgenic animals or co-carcinogen studies, e.g., initiation-promotion (i-p) studies.



Study Quality and Utility Assessment

Signaling Questions

Study design/population

Exposure conditions

Outcome assessment and measurement

Confounding

Analysis and reporting

Overall study utility:

High (++++)

Moderate (++)

Low (+)

Inadequate (0)

Responses for questions:

- Minimal concern
- Some concern
- Major concern
- Critical concern
- No information



- Study quality assessment
 - Follows [RoC Protocol](#); details in [RoC Handbook](#).
 - Consensus results of two independent reviewers reported in Appendix C of monograph.
- Initiation-promotion studies and transgenic studies; provided supportive information.
 - Dichloroacetic acid and trichloroacetic acid are complete carcinogens (Herren-Freund 1987), as well as cancer promoters (Pereira *et al.* 1997).
 - Dichloroacetic acid used as a positive to test TG.AC and p53 (+/-) transgenic mouse models, found to be inadequate models (NTP 2007b).
- Chronic carcinogenicity studies most informative.



Chronic Carcinogenicity Studies

Haloacetic acids	Species	Exposure route	Reference	Utility*
*High = +++, Moderate = ++, Low +				
Monohaloacetic acids				
Monochloroacetic acid	Mouse ♂♀	Gavage	NTP 1992	+++
	Rat ♂♀	Gavage	NTP 1992	++
	Rat ♂	Drinking water	DeAngelo <i>et al.</i> 1997	+++
Dichloroacetic acids				
Dichloroacetic acid	Mouse ♂♀	Drinking water	Wood <i>et al.</i> 2015	++
	Mouse ♂	Drinking water	DeAngelo <i>et al.</i> 1991, 1999	+++
			Bull <i>et al.</i> 1990	+
			Daniel <i>et al.</i> 1992	++
	Mouse ♀	Drinking water	Pereira <i>et al.</i> 1996	++
	Rat ♂	Drinking water	Richmond <i>et al.</i> 1995	++
			DeAngelo <i>et al.</i> 1996	++
Dibromoacetic acid	Mouse ♂♀	Drinking water	NTP 2007	+++
	Rat ♂♀	Drinking water	NTP 2007	+++
Bromochloroacetic acid	Mouse ♂♀	Drinking water	NTP 2009	+++
	Rat ♂♀	Drinking water	NTP 2009	+++
Trichloroacetic acids				
Trichloroacetic acid	Mouse ♂	Drinking water	Herren-Freund <i>et al.</i> 1987	++
			Bull <i>et al.</i> 1990	+
			DeAngelo <i>et al.</i> 2008	+++
	Mouse ♀	Drinking water	Pereira <i>et al.</i> 1996	++
	Mouse ♂♀	Intraperitoneal Inj.	Von Tungeln <i>et al.</i> 2002	++
	Rat ♂	Drinking water	DeAngelo <i>et al.</i> 1997	+++
Bromodichloroacetic acid	Mouse ♂♀	Drinking water	NTP 2015	+++
	Rat ♂♀	Drinking water	NTP 2015	+++



Haloacetic acids caused liver neoplasms

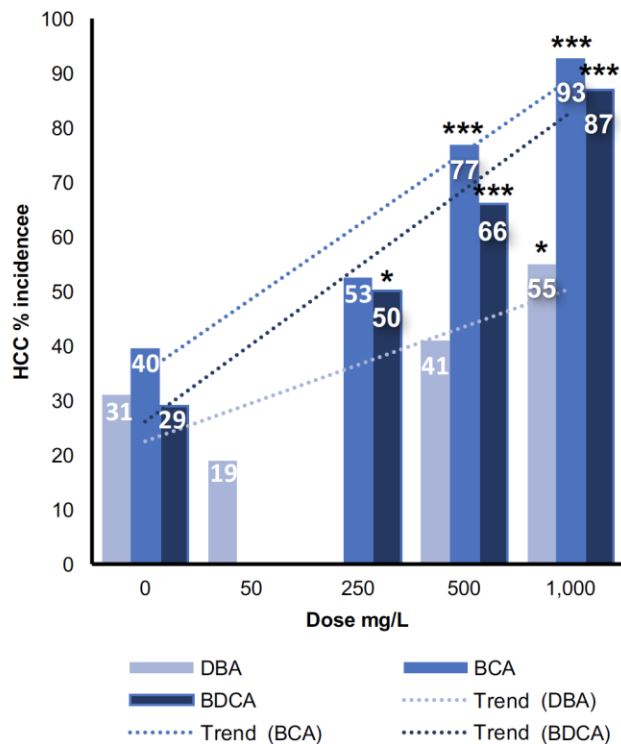
HALOACETIC ACID	RAT		MOUSE	
	Male	Female	Male	Female
Monochloroacetic acid	-	-	-	-
Dichloroacetic acid	HCC	NT	HCC	HCC
Trichloroacetic acid	-	NT	HCC	HCC
Dibromoacetic acid	-	-	HCC, HBI	HCC
Bromochloroacetic acid	-	-	HCC, HBI	HCC
Bromodichloroacetic acid	-	-	HCC, HBI	HCC, HBI

HCC = hepatocellular carcinoma; HBI = hepatoblastoma (carcinoma variant); NT = not tested; - = no liver tumors

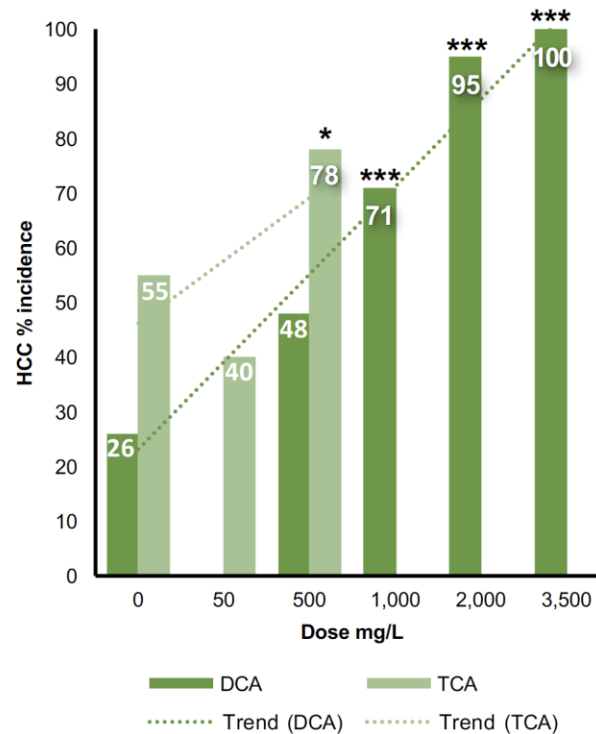


HAAs induce hepatocellular carcinoma in male mice

A. Bromine-containing HAAs in male mice



B. Chlorine-containing HAAs in male mice



Individual comparisons: * $P < 0.05$ *** $P < 0.001$.

Trend test P -value < 0.001 ; $P < 0.01$ for TCA

Sources: NTP 2007a, 2009, 2015; DeAngelo *et al.*, 1999, 2008



Other Cancer Sites

Drinking water exposure to brominated HAAs caused increased tumor incidence at sites in addition to liver

Haloacetic acid	Tissue site or neoplasia	Species, sex
Dibromoacetic acid	Malignant mesothelioma	Rat ♂
	Mononuclear cell leukemia	Rat ♀
	Lung	Mouse ♂
Bromochloroacetic acid	Malignant mesothelioma	Rat ♂
	Mammary gland	Rat ♀
	Large intestine	Rat ♂♀
Bromodichloroacetic acid	Malignant mesothelioma	Rat ♂
	Skin	Rat ♂
	Mammary gland	Rat ♀
	Harderian gland	Mouse ♂



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	Mammary gland	Rat ♀
	Large intestine	Rat ♂♀
Bromodichloroacetic acid	Malignant mesothelioma	Rat ♂
	Skin	Rat ♂
	Mammary gland	Rat ♀
	Harderian gland	Mouse ♂



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	Skin	Rat ♂
	Mammary gland	Rat ♀
	Harderian gland	Mouse ♂



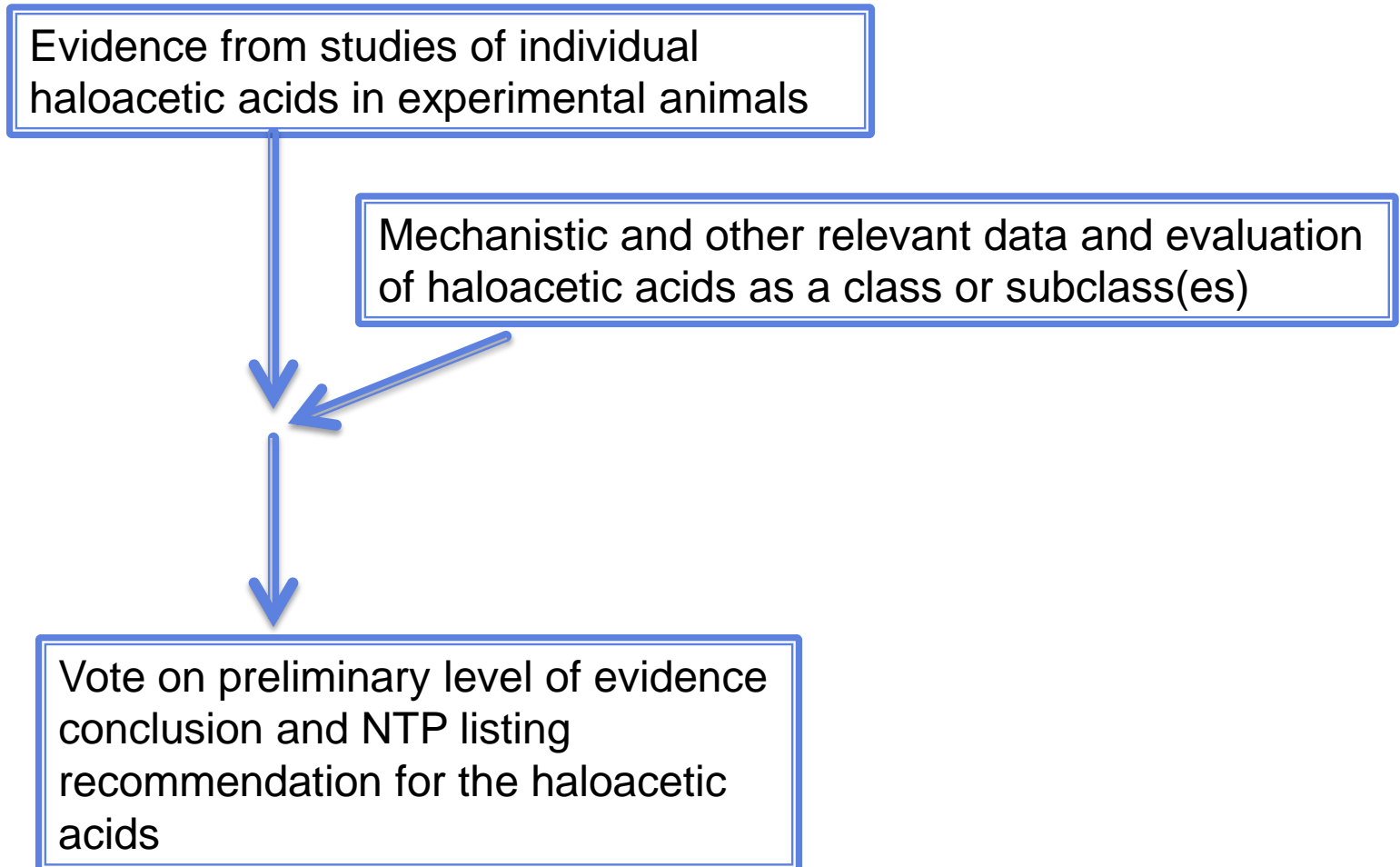
Drinking water exposure to haloacetic acids causes cancer in experimental animals*

Neoplasm or tissue	DCA				DBA				BCA				TCA				BDCA			
	Rats		Mice		Rats		Mice		Rats		Mice		Rats		Mice		Rats		Mice	
	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F
Liver	x		x	x			x	x			x	x			x	x			x	x
Mononuclear-cell leukemia						x														
Malignant mesothelioma					x				x								x			
Mammary gland										x								x		
Lung							x													
Skin																	x			
Harderian gland																			x	
Large intestine									x	x										

*Monochloroacetic acid (MCA) exposure did not produce tumors in experimental animals.



Defer vote on carcinogenicity in experimental animals





Studies in Experimental Animals

Questions?





Reviewer Questions

- Comment on whether the scientific information from cancer studies in experimental animals for each of the haloacetic acids found as water disinfection by-products is clear, technically correct, and objectively presented.
 - Identify any information that should be added or deleted.
- Comment on whether the approach and assessment of the utility of the animal carcinogenicity studies (study quality and sensitivity, ie, the ability to detect a true effect or hazard) for informing the cancer evaluation is systematic, transparent, objective, and clearly presented (Appendix C, Sections 4.2).
- Comment on and provide any scientific criticisms of NTP's cancer assessment of the experimental animal studies of exposure to the haloacetic acids found as water disinfection by-products and on how findings from the scientific evidence across studies were synthesized (Section 4.4).