A majority of U.S. households are supplied with disinfected water. Disinfection is necessary to destroy pathogenic organisms and prevent the outbreak of waterborne infectious diseases. Such diseases are largely under control in the United States, but waterborne outbreaks resulting in disease and mortality continue to occur (4). Although the benefits of water disinfection are well recognized, there is an undesirable side effect of producing various disinfection by-products (DBPs) when disinfectants such as chlorine and ozone react with organic and inorganic matter in water. Disinfection by-products (DBPs) are formed when disinfectants such as chlorine, chloramine, and ozone react with organic and inorganic matter in water. The observations that some DBPs such as trihalomethanes (THMs), di-/trichloroacetic acids, and 3-chloro-4-(dichloromethyl)-5-hydroxy-2(5H)-furanone (MX) are carcinogenic in animal studies have raised public concern over the possible adverse health effects of DBPs. To date, several hundred DBPs have been identified. To prioritize research efforts, an in-depth, mechanism-based structure–activity relationship analysis, supplemented by extensive literature search for genotoxicity and other data, was conducted for ranking the carcinogenic potential of DBPs that met the following criteria: a) detected in actual drinking water samples, b) have insufficient cancer bioassay data for risk assessment, and c) have structural features/alerts or short-term predictive assays indicative of carcinogenic potential. A semiquantitative concern rating scale of low, marginal, low-moderate, moderate, high-moderate, and high was used along with delineation of scientific rationale. Of the 209 DBPs analyzed, 20 were of priority concern with a moderate or high-moderate rating. Of these, four were structural analogs of MX and five were haloalkanes that presumably will be controlled by existing and future THM regulations. The other eleven DBPs, which included halonitrites (6), haloketones (2), halaldehyde (1), halonitrokanne (1), and dialdehyde (1), are suitable priority candidates for future carcinogenicity testing and/or mechanistic studies. Key words: DBPs, disinfection by-products, carcinogenic potential, drinking water, mechanism-based SAR analysis, prioritization, structure–activity relationship. Environ Health Perspect 110(suppl 1):75–87 (2002). http://ehpnet1.niehs.nih.gov/docs/2002/suppl-1/75-87/woolabstract.html

Disinfection by-products (DBPs) are formed when disinfectants such as chlorine, chloramine, and ozone react with organic and inorganic matter in water. The observations that some DBPs such as trihalomethanes (THMs), di-/trichloroacetic acids, and 3-chloro-4-(dichloromethyl)-5-hydroxy-2(5H)-furanone (MX) are carcinogenic in animal studies have raised public concern over the possible adverse health effects of DBPs. To date, several hundred DBPs have been identified. To prioritize research efforts, an in-depth, mechanism-based structure–activity relationship analysis, supplemented by extensive literature search for genotoxicity and other data, was conducted for ranking the carcinogenic potential of DBPs that met the following criteria: a) detected in actual drinking water samples, b) have insufficient cancer bioassay data for risk assessment, and c) have structural features/alerts or short-term predictive assays indicative of carcinogenic potential. A semiquantitative concern rating scale of low, marginal, low-moderate, moderate, high-moderate, and high was used along with delineation of scientific rationale. Of the 209 DBPs analyzed, 20 were of priority concern with a moderate or high-moderate rating. Of these, four were structural analogs of MX and five were haloalkanes that presumably will be controlled by existing and future THM regulations. The other eleven DBPs, which included halonitrites (6), haloketones (2), halaldehyde (1), halonitrokanne (1), and dialdehyde (1), are suitable priority candidates for future carcinogenicity testing and/or mechanistic studies. Key words: DBPs, disinfection by-products, carcinogenic potential, drinking water, mechanism-based SAR analysis, prioritization, structure–activity relationship. Environ Health Perspect 110(suppl 1):75–87 (2002). http://ehpnet1.niehs.nih.gov/docs/2002/suppl-1/75-87/woolabstract.html

National Toxicology Program (NTP), is conducting 2-year cancer rodent bioassays, transgenic mouse cancer assays, and medaka fish cancer assays, as well as tests for reproductive, developmental, immunologic, and neurologic toxicities on several DBPs. In addition, an information collection rule was promulgated (8) to collect national occurrence information on 32 DBPs (Table 1).

The chemicals in the Stage 1 DBPR are among the DBPs with the highest occurrence in drinking water. However, hundreds of other DBPs formed from treatment with various disinfectants, including chlorine, have been identified. There is a limited amount of information on most of these DBPs beyond their identification in water. In addition, there are many unidentified DBPs, as evidenced by measurements of total organic halides compared with known halogenated DBPs (5). The U.S. EPA believes that the standards in the Stage 1 DBPR will, to some extent, control these other known and unknown DBPs. The U.S. EPA must better define the risk from the DBPs identified in drinking water before it can determine whether they are adequately controlled by current standards. The two most important factors needed to characterize risk are occurrence and toxicity. For most DBPs that have been identified, few or no data are available in either area. Further research on these DBPs is therefore necessary. Because there are hundreds of DBPs for which there are few or no health or occurrence data, there is a need for prioritization before expensive toxicologic tests and occurrence monitoring studies are initiated.

Yin-Tak Woo,1 David Lai,1 Jennifer L. McLain,2 Mary Ko Manibusan,2 and Vicki Dellarco3

1Office of Prevention, Pesticides, and Toxic Substances, Risk Assessment Division, 2Office of Water, Office of Ground Water and Drinking Water; 3Office of Pesticide Program, Health Assessment Division, U.S. Environmental Protection Agency, Washington, DC, USA

Address correspondence to M.K. Manibusan, Office of Water, U.S. EPA, 1200 Pennsylvania Ave. NW, MC-4607M, Washington, DC 20460 USA. Telephone: (202) 564-5265. Fax: (202) 564-3767. E-mail: manibusan.mary@epa.gov

We thank M. Cox for his guidance and review, S. Richardson of the National Exposure Research Laboratory for providing the list of disinfection by-products, and A. Richard of the National Health and Environmental Effects Research Laboratory for technical advice.

This document has not been reviewed in accordance with U.S. Environmental Protection Agency policy. Mention of trade names or commercial products does not constitute endorsement or recommendation for use. The scientific views expressed are solely those of the authors and do not necessarily reflect those of the U.S. Environmental Protection Agency.

Received 13 September 2001; accepted 19 November 2001.
To achieve this goal, a tiered approach to prioritization was designed for evaluating which DBPs, if any, present a health concern sufficient to warrant additional research to better characterize the risk. Those DBPs considered to present a health concern would first be tested in a battery of appropriate in vitro or in vivo screening assays. The results could be used to decide which DBPs deserve further studies such as acute/subchronic specialized animal tests for neurotoxicity, immunotoxicity, developmental, reproductive, or system toxicity, and medium-term tests for cancer (e.g., transgenic mouse, medaka fish). Results from these studies could help prioritize more expensive long-term tests and mechanistic studies. In addition, occurrence studies and research on the development of analytical methods for these high-priority DBPs would also need to be addressed. In this article we discuss the process the U.S. EPA used to prioritize DBPs for future testing, in which DBPs were examined by expert structure–activity relationship (SAR) judgment with emphasis on genotoxic cancer potential.

The SAR analysis in this study addresses only the carcinogenic potential of DBPs. There are other ongoing efforts for predicting noncancer effects, including reproductive and developmental toxicity (9,10). Cancer has been raised as an end point of concern in epidemiologic studies (11–19). In addition, cancer is often the most sensitive health end point that is used to set drinking water standards. Most important, cancer concerns were addressed first because the predictive tools for evaluating the potential cancer risk are more developed than those for other end points such as reproductive and developmental effects.

Prioritization Approach

The U.S. EPA designed a simple prioritization scheme for determining which DBPs may require additional research (Figure 1). First, the U.S. EPA compiled a list of DBPs to consider for prioritization. More than 600 DBPs from various disinfectant combinations that have been identified and cataloged by the U.S. EPA (20) served as an important reference. Additional DBPs were subsequently added as new information became available (21,22). Of these, the U.S. EPA considered only those DBPs found or detected in actual drinking water samples. DBPs found only through laboratory experiments were excluded because these experiments are often performed under conditions that are not representative of actual water treatment practices. Thus, there is uncertainty as to whether DBPs identified in laboratory experiments can actually be found in drinking water samples. Several additional criteria included eliminating DBPs with incomplete chemical structure characterizations. In addition, chemicals believed to be impurities from processes other than disinfection, such as leachates from treatment plant materials and laboratory equipment (e.g., naphthalene, 3-ethyl styrene), were eliminated. The list of 252 remaining DBPs was peer reviewed by chemists with expertise in DBP formation and identification to ensure, to the extent possible, that the chemicals in the list were all actual or probable DBPs. After these criteria were applied, 239 DBPs remained for research prioritization.

In the next step, the U.S. EPA identified those DBPs that have or will have 2-year cancer bioassay data and occurrence data sufficient for making a hazard assessment, and those DBPs for which sufficient bioassay data are will be available but insufficient occurrence data currently exist. The criteria for judging if sufficient toxicity data exist to conduct a cancer assessment were as follows: a) there is an MCLG from the Stage 1 DBP rule or past drinking water rules; b) the NTP, the U.S. EPA, or others have conducted or will conduct a 2-year cancer bioassay; or c) there is an oral slope factor on the agency’s Integrated Risk Information System (IRIS) (23). The criteria for judging if sufficient occurrence data exist to derive a national estimate of exposure were as follows: d) there is an MCLG from the Stage 1 DBP rule or past drinking water rules, or e) the DBP is included in the information collection rule for DBPs that is collecting national occurrence data. Thirty DBPs (Table 2) were identified in this step and eliminated from SAR consideration.

As discussed in detail below, the remaining 209 DBPs were analyzed by expert judgment SAR analysis. After DBPs of low concern were identified, a literature search for mutagenicity and other toxicity data was performed for the remaining DBPs to provide additional input to the SAR analysis. The DBPs were categorized by a semiquantitative ranking scale of high (H), high–moderate (HM), moderate (M), low-moderate (LM), marginal (Mar), and low (L) concern. Because these concern levels are based on expert judgment relative to known carcinogens, there is no exact definition. As a guideline, the following narrative descriptions have been used (24): a) H = highly likely to be a potent multispecies, multitarget carcinogen even at low doses; b) HM = highly likely to be a moderately active multispecies/target carcinogen at moderate doses; c) M = likely to be a moderately active multispecies/target carcinogen at relatively high doses or active single species/target carcinogen at low doses; d) LM = likely to be weakly carcinogenic, or carcinogenic toward a single species/target at relatively high doses; e) Mar = likely to have marginal carcinogenic activity or may be weakly carcinogenic at doses at or exceeding maximum tolerated doses; f) and L = unlikely to be carcinogenic. Table 3 lists all 209 DBPs together with their assignment to their most appropriate structural chemical classes and categorization of concern level. The basic principles of mechanism-based SAR analysis used for categorizing concern levels of DBPs are discussed below.

Overview of Basic Principles of the Structure–Activity Relationship Approach

SAR analysis has been used to predict toxic potential of chemicals for which test data are limited or not available. SAR is an indispensable tool to help prioritize research and development on a compound by providing valuable initial information on its hazard potential. For organic chemicals the

Table 1. DBPs for which national occurrence data were collected in the information collection rule.

| DBP class                  | Individual DBPs                                                                 |
|----------------------------|----------------------------------------------------------------------------------|
| Trihalomethanes            | Chloroform, bromodichloromethane, dibromochloromethane, bromoform                |
| Haloacetic acids           | Tri-, di-, monochloroacetic acid, tri-, di-, monobromoacetic acid, bromochloro-, bromodichloro-, chlorodibromoacetic acid |
| Haloacetanilides           | Tri-, dichloracetanilide, bromochloracetanilide, dibromoacetanilide               |
| Haloacetones               | 1,1-Dichloropropanone, 1,1,1-trichloropropanone                                  |
| Aldehydes                  | Formaldehyde, acetaldehyde, propanal, butanal, pentanal, glyoxal, methyl glyoxal |
| Inorganics                 | Bromate, chlorite, chlorate                                                     |

Figure 1. Selection of DBPs for SAR analysis.
The predictive capability of SAR analysis combined with other toxicity information has been demonstrated (25–28). Currently, SAR analysis is well developed for chemicals and metabolites believed to initiate carcinogenesis through covalent interaction with DNA (i.e., DNA-reactive, -mutagenic, -electrophilic, or -proelectrophilic chemicals). At a more limited scale there is some SAR experience for predicting carcinogenicity that does not involve DNA reactive mechanisms but rather involves cellular toxicity, pathophysiologic parameters, or receptor-mediated mechanisms such as Ah receptor, peroxisome proliferation, and endocrine disruption (24,29–31).

Mechanism-based SAR analysis has been effectively used by the U.S. EPA for many years to assess the potential carcinogenic hazard of new chemicals, for which there are no or scantly data, under the Premanufacture Notification program of the Toxic Substances Control Act (3.2). The same approach has been used in design of safer chemicals (3.3) and pollution prevention (3.4). An expert system (OncoLogic) has been developed to systematize and codify the agency’s SAR expertise in predicting carcinogenic potential of chemicals (26). The principal authors of this present article have been involved in these program activities for more than a decade. The SAR predictions of the cancer potential of DBPs in this article are based mainly on human expert judgment, with some input from the OncoLogic expert system. A similar approach has been applied to prospective prediction of the outcome of NTP cancer bioassays (27). The predictive performance of our approach relative to other predictive methods has been affirmed by an independent evaluation (28).

**Mechanism-Based Structure–Activity Analysis**

Essentially, mechanism-based SAR analysis involves comparison of an untested chemical with structurally related compounds for which carcinogenic activity is known. Considering the most probable mechanism(s) of action, the structural features and functional properties of the untested compound are evaluated and compared with reference compounds. All available knowledge and data relevant to evaluation of carcinogenic potential of the untested chemical are considered. These include:  
1. **SAR knowledge base of the related chemicals;**
2. **toxicokinetics and toxicodynamics parameters** (including physicochemical properties, route of potential exposure, and mode of activation or detoxification) that affect the delivery of biologically active intermediates to target tissue(s) for interaction with cellular macromolecules or receptors; and
3. **supportive noncancer screening or predictive data** known to correlate to carcinogenic activity. A prediction of carcinogenic potential involves integration of all this available information with human expert intuition and judgment.

In evaluating the DBPs both structural and functional criteria were applied. The structural criteria and methodology for assessing carcinogenic potential of chemicals have been discussed in detail in previous reviews (26,27). Basically, the structural moieties or fragments that may contribute to carcinogenic activity through a perceived or postulated mechanism are identified, and the modifying role of the rest of the molecule to which the structural moiety/fragment is attached is evaluated. Whenever possible, comparison is made to a structurally related reference compound with known carcinogenic activity (tested preferably by the same route of administration as the chemical in question) to evaluate whether the difference in chemical structures may lead to an increase or decrease in carcinogenic activity. Electrophiles can interact with DNA and potentially lead to mutagenesis. The identification of electrophiles and their precursors is thus fundamental to the prediction of mutagenic carcinogens. Some of the commonly encountered electrophiles or electrophilic intermediates in carcinogenesis include carbonium ions (alkyl-, aryl-, benzylic), nitrenium ions, epoxides and oxonium ions, aldehydes, polarized double bonds (cyclic-unsaturated carboxyls or carboxylates), peroxides, free radicals, and acylating intermediates (27).

For compounds that are metabolically activated, resonance stabilization provides reactive intermediates a longer reactive lifetime. Structural features that may furnish resonance stabilization include conjugated double bonds, an aryl moiety (especially those capable of providing long resonance pathways), ring positions that allow several resonance forms, and structures that allow reversible cyclization of reactive intermediates.

The molecule to which a reactive moiety is attached may significantly affect its carcinogenic potential. Many potent carcinogens (e.g., aflatoxin B1, benzo[a]pyrene) have a relatively planar molecular size and shape favorable for DNA intercalation in addition to having a reactive functional group. Attachment of a reactive electrophilic group to normal cellular molecular constituents may also enhance carcinogenic activity (e.g., attaching the moderately carcinogenic nitrogen mustard to uracil yields a more potent carcinogen, uracil mustard), probably by serving as a carrier to reach the target macromolecule. Conversely, the presence of highly hydrophilic groups or bulky substituents that may affect metabolic activation or molecular planarity tends to decrease or eliminate carcinogenic activity.

The structural basis for identifying receptor-mediated carcinogens is considerably less understood and is dependent on the type of receptor believed to be involved. Some of the structural features useful in identifying these carcinogens include:

1. Planar tricyclic molecule with lateral ring substitution for Ah receptor–mediated 2,3,7,8-tetrachlorodibenzo-p-dioxin–related chemicals (35,36);
2. Nonmetabolizable acids (such as branching at the carbon next to the acid-bearing carbon) for peroxisome proliferator–type carcinogens (29); and
3. A molecular descriptor containing a phenolic group 6 angstroms away from a lipophilic moiety for at least some types of hormonal carcinogens (30).

In addition to the information on structural

**Table 2. DBPs that have or will have sufficient data for hazard assessment.**

| DBPs with cancer and occurrence data | DBPs with cancer data but lacking occurrence data |
|-------------------------------------|-----------------------------------------------|
| Aetaldehyde (48,71)                 | Benzaldehyde (47)                             |
| Bromate (48,71)                     | 1,4-Benzenedicarboxylic acid (terephthalic acid) (70) |
| Bromochloroacetic acid (a) (69,71)  | Benzyl chloride (47)                          |
| Bromochloroacetic acid (b) (69,71)  | 3-Chloro-4-(dichloromethyl)-5-hydroxy-2(5H)-furanone (IM) (50) |
| Bromochloromethane (47,48,71)       | Dichloromethane (47)                          |
| Bromiform (47,71)                   | 1,4-Dioxane (47)                              |
| Chloral hydrate (a) (69,71)         | Hexachloroethane (47)                         |
| Chlorate (a) (69,72)                | Hydrogen peroxide (48)                       |
| Chlorof orm (48,71)                 | 4-Methyl-2-pentanone (a) (69)                 |
| Chloropicrin (47,71)                |                                              |
| Dibromoacetic acid (a) (69,71)      |                                              |
| Dibromochloroacetic acid (a) (69,71)|                                              |
| Dibromochloromethane (47,71)        |                                              |
| Dichloroacetic acid (48,71)         |                                              |
| 2,4-Dichlorophenol (47,72)          |                                              |
| Formaldehyde (48,71)                |                                              |
| Glyoxal (a) (69,71)                 |                                              |
| Monochloroacetic acid (47,71)       |                                              |
| Trichloroacetic acid (48,71)        |                                              |
| 2,4,6-Trichlorophenol (47,48,73)    |                                              |

*Currently under study or selected for testing.*
| Chemical name | CAS number | Chemical class | Concern level |
|---------------|------------|----------------|---------------|
| 1. Acetone     | [64-64-1]  | Nonhalogenated ketones | L             |
| 2. Benzenacetaldehyde | [122-78-1] | Nonhalogenated aldehydes | Mar           |
| 3. 1,2-Benzenedicarboxylic acid | [88-99-3] | Nonhalogenated acids | L             |
| 4. 1,3-Benzenedicarboxylic acid (isophthalic acid) | [121-91-5] | Nonhalogenated acids | L             |
| 5. 1,2,3-Benzenetricarboxylic acid (hemimellitic acid) | [38382-97-7] | Nonhalogenated acids | L             |
| 6. 1,2,4-Benzenetricarboxylic acid (trimellitic acid) | [528-44-8] | Nonhalogenated acids | L             |
| 7. 1,3,5-Benzenetricarboxylic acid (trimesic acid) | [554-96-0] | Nonhalogenated acids | L             |
| 8. 1,4-Benzoquinone | [255378] | Nonhalogenated aromatics | LM            |
| 9. Benzoic acid | [65-85-0]  | Nonhalogenated aromatics | L             |
| 10. Benzotrione | [108-04-0] | Nonhalogenated aromatics | Mar           |
| 11. Benzyl cyanide | [140-24-0] | Nonhalogenated aromatics | L             |
| 12. 2,6-Bis-(1-dimethylalkyl)-2,5-cyclohexadiene-1,4-dione | [719-22-2] | Nonhalogenated aromatics | Mar           |
| 13. 1,2-Bis-(1-methylalkyl)-benzene | | | |
| 14. Bromoacetic acid | [79-08-2]  | Haloacids | Mar           |
| 15. Bromoacetonitrile | [509-17-0] | Haloacids/amides | LM            |
| 16. Bromoaniline | | Haloamines/amides | L             |
| 17. 2-Bromobenzohialdene | [2515-40-7] | Haloamines/amides | L             |
| 18. 2-Bromobutane (sec-butyl bromide) | [78-76-0]  | Haloamines/amides | LM            |
| 19. Bromochloroacetanilide | [83483-61-2] | Haloamines/amides | M             |
| 20. Bromochloroaniline | [77392-23-9] | Haloamines/amides | L             |
| 21. Bromochloroiodomethane | [39780-00-8] | Haloamines/amides | LM            |
| 22. Bromochloromethane | [749-75-7]  | Haloamines/amides | LM            |
| 23. Bromochloromethyl acetate | | Acetate of haloacids | LM            |
| 24. 1,1-Bromochloropropionate | [513-88-2] | Haloformates and related compounds | HM            |
| 25. 3-Brom-4-(dibromomethyl)-5-hydroxy-2(3H)-furanone (BMX-3) | | | |
| 26. Bromochloroacetaldehyde | [34818-29-9] | Haloaldehydes | LM            |
| 27. Bromochloroacetone | [60523-73-1] | Haloaldehydes | LM            |
| 28. Bromochloroform | [918-07-4]  | Haloaldehydes | LM            |
| 29. 1-Bromo-1,1-dichloropropionate | [18995-35-0] | Haloaldehydes | LM            |
| 30. Bromopropin | [484-10-8]  | Haloaldehydes | LM            |
| 31. 3-Bromopropyl-chloromethyl ether | | Haloformates Mar (or), HM (inhaled) | |
| 32. Butanal | [123-72-8]  | Nonhalogenated aldehydes | L             |
| 33. Butanediol | [838-37-9]  | Nonhalogenated aldehydes | M             |
| 34. Butyric acid | [107-92-6]  | Nonhalogenated acids | L             |
| 35. Butanone | [78-93-3]   | Nonhalogenated ketones | L             |
| 36. cis-Butenedioic acid (maleic acid) | [110-16-7]  | Nonhalogenated acids | L             |
| 37. trans-Butenedioic acid (fumaric acid) | [110-17-8]  | Nonhalogenated acids | L             |
| 38. 2-(2-Butoxyethoxy)-ethanol (diethylene glycol (ethyl ether) | [112-34-5]  | Other nonhalogenated organics | |
| 39. 2-(2,2-Butoxyethoxy) ethoxy-ethanol | | Other nonhalogenated organics | L             |
| 40. 2,2-Dibutoxyethanol | [143-22-6]  | Other nonhalogenated organics | L             |
| 41. Chlorite | [14988-27-7] | Inorganics | LM            |
| 42. Chloroacetaldehyde | [107-20-0]  | Haloaldehydes | LM            |
| 43. Chloroacetone | [107-14-2]  | Haloaldehydes | LM            |
| 44. 3-Chloro-4-bromochloromethyl-5-hydroxy-2(3H)-furanone (BMX-1) | | Haloformates and related compounds | HM            |
| 45. Chlorobutane | [16045-92-4] | Haloacids | Mar           |
| 46. Chloro-2-butanol | [563-94-0]  | Other nonhalogenated organics | M             |
| 47. Chloro-2-butanol acetate | [51482-23-0] | Acetate of haloalcohols | Mar           |
| 48. Chlorobutenedioic acid | | Haloaldehydes | LM            |
| 49. Chlorocyclohexanone | [822-97-7]  | Haloaldehydes | LM            |
| 50. Chlorodibromobutane | | Haloaldehydes | LM            |
| 51. 3-Chloro-4-(dichloromethyl)-5-hydroxy-2(3H)-furanone (BMX-1) | | Haloformates and related compounds | HM            |

(Continued)
| Chemical name | CAS number | Chemical class | Concern level |
|---------------|------------|----------------|---------------|
| Dodecanic acid | [143-07-7] | Nonhalogenated acids | L             |
| 4-Dodecyl-5-ethyl-3-(E)-furanone | | Other nonhalogenated organics | L             |
| 1-Ethoxy-1-hydroxybutane | | Other nonhalogenated organics | L             |
| 2-Ethyl-3-methylmaleic acid | | Nonhalogenated acids | L             |
| Glyoxylic acid | [298-12-4] | Nonhalogenated acids | L             |
| Hexenoic acid | [2363-71-5] | Nonhalogenated acids | L             |
| Heptenedioic acid, dioctyl ester | | Other nonhalogenated organics | L             |
| Hexenedioic acid (adipic acid) | [124-04-9] | Nonhalogenated acids | L             |
| 9-Hexadecanoic acid | [10030-73-6] | Nonhalogenated acids | L             |
| Hexachloropropane | [116-16-5] | Haloalkanes/alkenes | L             |
| Heptanoic acid | [111-14-8] | Nonhalogenated acids | L             |
| Heptanenitrile | [629-08-3] | Other nonhalogenated organics | L             |
| Heptanal | [111-71-7] | Nonhalogenated aldehydes | L             |
| Heptane | [111-16-6] | Nonhalogenated aldehydes | L             |
| Heptanenitrile | [111-14-8] | Other nonhalogenated organics | L             |
| Hexanoic acid | [142-62-1] | Nonhalogenated acids | L             |
| Hexenal | [505-57-7]-cis | Nonhalogenated aldehydes | LM            |
| Hexanal | [86-25-1] | Nonhalogenated aldehydes | L             |
| Hexanedioic acid (acidic acid) | [124-04-9] | Nonhalogenated acids | L             |
| Hexanedioic acid, dioctyl ester | | Other nonhalogenated organics | L             |
| Hexane | [57-10-3] | Nonhalogenated aldehydes | L             |
| Hexene | [10030-73-6] | Nonhalogenated acids | L             |
| Hexanal | [86-25-1] | Nonhalogenated aldehydes | L             |
| Hexanoic acid | [142-62-1] | Nonhalogenated acids | L             |
| Hexenal | [505-57-7]-cis | Nonhalogenated aldehydes | LM            |
| Hexanal | [86-25-1] | Nonhalogenated aldehydes | L             |
| Hexanoic acid | [142-62-1] | Nonhalogenated acids | L             |
| Hexenal | [505-57-7]-cis | Nonhalogenated aldehydes | LM            |
| Hexanal | [86-25-1] | Nonhalogenated aldehydes | L             |
| Hexanoic acid | [142-62-1] | Nonhalogenated acids | L             |
| Hexenal | [505-57-7]-cis | Nonhalogenated aldehydes | LM            |
| Hexanal | [86-25-1] | Nonhalogenated aldehydes | L             |
basis of receptors, functional criteria using short-term test data can also be used.

Functional criteria involve consideration of all the available short-term noncancer predictive data and pharmacologic and toxicologic capabilities correlated or associated with carcinogenic activity. Functional criteria complement structural criteria because structural considerations alone cannot forecast entirely new types of carcinogens. Furthermore, functional criteria may serve as a means to confirm or cast doubt on the mechanistic assumptions made in applying structural criteria. Information that is highly useful for predicting carcinogenic potential includes data on oncogenes, tumor suppressor genes, genotoxicity and/or ability to bind covalently to DNA, apoptosis, cellular proliferation, immunosuppression, and subchronic toxicity end points that are indicative or suggestive of carcinogenic potential. Ideally, all of the available data should be evaluated with respect to predictive capability, strength of evidence, and relevance to the carcinogenic process and then integrated. Positive predictive tests and data covering all aspects of the carcinogenic process (initiation, promotion, and progression) should be given more weight than multiple tests detecting the same mechanistic end point (24).

Conditions of Hazard Expression (Routes of Exposure)

An individual may be exposed to DBPs by different routes of exposure (e.g., inhalation from showering, dermal from bathing, oral from tap water consumption). In evaluating the carcinogenic potential of a compound, it is important to consider the route of exposure because the hazard and risk posed by a compound may vary by exposure route (37). Delivery of the reactive intermediate to target macromolecules such as DNA is crucial for carcinogenic activity, and exposure routes such as inhalation and injection are often required for maximal activity for direct-acting reactive chemicals. For example, electrophiles such as aldehydes are DNA reactive, but this reactivity also means they are readily detoxified by cellular-protective nucleophiles such as glutathione (GSH). Their toxicity, therefore, tends to be localized to the port of entry. Thus aldehydes, which are of cancer concern via inhalation, pose a lower cancer concern via the oral route because they are readily oxidized to acids before they can react with DNA. However, functional or genetically diminished capability to detoxify aldehydes may be at higher risk. The SAR predictions presented in this document focus mainly on the hazard potential via ingestion of drinking water, a major route of exposure to DBPs. Inhalation exposure to some volatile DBPs may occur through bathing or showering. In general, for the purpose of ranking hazard potential, DBPs that require metabolic activation (e.g., THMs) should have similar hazard potential whether via oral or inhalation, whereas DBPs that are highly reactive direct-acting chemicals (e.g., α-haloethers if they could actually remain reactive through the water delivery system) are expected to have higher concern via inhalation than via oral route.

Literature Search Approach

In support of the SAR analysis of DBPs of greater than low concern, a literature search was performed using chemical abstract numbers. There were several DBPs for which a literature search was not performed because Chemical Abstracts Service (CAS) registry numbers could not be found in the CAS Scientific and Technical Network online database (38). For some DBPs, information on closely related compounds was searched.

Because the present SAR study emphasized predicting genotoxic carcinogens, selected databases were used. Both the Environmental Mutagen Information Center-Front and Back Files (EMIC/EMICBACK) (39) were searched. EMICBACK, developed and maintained by the Oak Ridge National Laboratory, is a bibliographic database on compounds tested for genotoxic activity. The database contains literature published from 1950 to 1990 and includes some references published before 1950. EMIC covers publications from 1989 to the present. The Chemical Carcinogenesis Research Information System (CCRIS) (40), developed and sponsored by the National Cancer Institute, was also searched. CCRIS contains information from carcinogenicity, mutagenicity, tumor promotion, and tumor inhibition studies that have been evaluated for acceptability by experts in carcinogenesis. CCRIS contains 7,000 chemical records. Additionally, the NTP (41), IRIS (23), and the Agency for Toxic Substances and Disease Registry (ATSDR) toxicological profiles (42) were searched for availability of cancer bioassay data. Although the EMIC/EMICBACK and CCRIS databases were searched, some information on mutagenicity and carcinogenicity may have been missed, in particular, information on metabolism and mode of action (e.g., cell proliferation, apoptosis).

Mutagenicity and carcinogenicity data were gathered from either abstracts or actual publications and compiled into a summary table listing the chemical name, CAS number, test, strain, method, result, dose, and publication reference. This information was then used to assist in the SAR predictions for DBPs of greater than low concern.

Structure–Activity Relationships Cancer Prediction for Disinfection By-Products

Prior to the SAR analysis the U.S. EPA determined that those DBPs ranked as moderate, high-moderate, or high would be the priority candidates for future testing. This was decided because of the large number of DBPs involved. If a chemical with few occurrence data was determined to be of a higher concern, then further toxicity research on the chemical might be justified. If, however, a chemical was determined to be of a lower concern, some occurrence data beyond mere identification would have to be obtained before testing would be warranted.

SAR predictions were made for 209 DBPs (Table 3). The DBPs were first reviewed to identify chemicals with low concern. Judgments of low cancer concern were based on structural similarity to chemicals with negative cancer data, a lack of structural alert for genotoxicity, or presence of structural features suggestive of low cancer risk via the oral route (26,27,33). Once the DBPs of low concern were removed from the list, a literature search was done for the remaining DBPs. It should be noted that literature was not found in EMIC/CCRIS/NTP/IRIS/ATSDR databases for many of the DBPs. Thus, the mechanism-based SAR predictions relied heavily on expert judgment and experience. SAR assumptions and conclusions for concern levels and specific classes of DBPs are discussed below.

Distribution of Disinfection By-Products within Structure–Activity Relationship Concern Levels and Structural Classes

Table 4 summarizes the structural class and concern level distribution of the 209 DBPs. Of the 209 DBPs examined, none are considered to be of high concern. Only 20 (<10%) are predicted to have a concern level of moderate or high-moderate. With one exception, all these compounds are halogenated, with most of them belonging to the structural classes of haloaromatics, haloalkanes/alkenes, haloalcohols, and haloesters. A detailed analysis of these four classes will be presented. Haloacids would have constituted a major class of concern. However, because several haloacids have already been tested or selected for testing (Tables 1 and 2), they are not considered in detail in the present study. Outside of the four major classes of concern, one haloaldehyde (dichloroacetaldehyde), one haloacetate (dibromonitromethane), and
one nonhalogenated aldehyde (butanedia) are considered of moderate concern. Dichloroacetaldehyde has been given a moderate concern because it is a potential cross-linking agent. It can also be readily oxidized to dichloroacetic acid, which has been shown to be a rodent carcinogen with multiple mechanisms of action (43–45). Dibromonitr reimethane has been given a moderate concern because the corresponding dichloronitr reimethane is believed to be the proximate mutagen of chloropirin (46). The replacement of chlorine by bromine should make it a more potent mutagen because bromine is a better leaving group. The structurally related nitromethanes, particularly tetratin reimethane, are carcinogenic, whereas chloropirin (trichloronitr reimethane) is noncarcinogenic in mice and inconclusive in rats (47). Butanedia is the only nonhalogenated DBP given a moderate concern in the present study. This compound has two terminal reactive aldehydes separated by two methyl groups, which should make it a highly favorable cross-linking agent.

The majority (131/209) of the DBPs in this study are considered to have low (98/209) or marginal (33/209) cancer concern. Most of these compounds are nonhalogenated carboxylic acids, ketones, aldehydes, and miscellaneous organic compounds. Nonhalogenated hydrophilic carboxylic acids are not of concern because they are unlikely to be absorbed and, even if absorbed, are rapidly excreted. High-molecu lar-weight nonhalogenated carboxylic acids are also a low concern because they have no structural alerts, and many are natural products and nutrients, U.S. Food and Drug Administration food additives, and synthetic flavorings. Several medium-size (6–10 carbons) carboxylic acids with branching at the carbon next to the carboxylic group (omega-1 carbon) were considered potential rodent carcinogens because of potential peroxisome-proliferating activity but were given a marginal concern rating because of uncertain human significance. A number of nonhalogenated aldehydes, particularly those with high molecular weight, are given low or marginal concern because they are unlikely to have significant dose via drinking water; this subject will be further discussed below. With the exception of α,β-unsaturation or closely spaced dicarbonyl groups, nonhalogenated ketones are mostly of low concern because they lack electrophilic activity and are generally not associated with carcinogenicity. Halogenated aliphatic amines are a low concern because of structural analogy to chloramine, which has negative cancer bioassay data (47).

The remainder (58/209) of the DBPs fall into the low-moderate concern category and represent a wide variety of classes, both halogenated and nonhalogenated. In general, these DBPs are considered to have a concern level lower than moderate because they have a less active chlorine/bromine group or contain structural features that are not as favorable for carcinogenic activity. These DBPs include certain haloacids, haloalkdehydes, haloethers, haloamides, nonhalogenated aromatics, and reactive ketones. Additionally, a large number (35/209) of haloaldehydes, halofuranones, haloalkanes, haloamines, and nonhalogenated aldehydes are considered of low-moderate concern. The rationale for their assignments as well as the SAR information available on these classes are discussed in more detail below.

Table 4. Structural class and concern level distribution of DBPs under evaluation.

| Structural class of the DBP | Total no. of DBPs | Concern level |
|----------------------------|------------------|--------------|
|                            |                  | HM | M  | LM | Mar or L |
| Halofuranones MX-related   | 10               | 3 (30%) | 1 (10%) | 4 (40%) | 2 (20%) |
| Haloalkanes/haloalkanes     | 14               | – | 5 (36%) | 6 (43%) | 3 (21%) |
| Halonitrites               | 15               | – | 6 (40%) | 7 (47%) | 2 (13%) |
| Haloaldehydes              | 18               | – | 2 (11%) | 14 (88%) | 2 (11%) |
| Haloalcohols               | 6                | – | 1 (17%) | 5 (83%) | – |
| Halonitroxoalkanes         | 4                | – | 1 (25%) | 3 (75%) | – |
| Halkoacids                 | 11               | – | 2 (18%) | 9 (82%) | – |
| Acetate of haloalcohols    | 2                | – | 1 (50%) | 1 (50%) | – |
| Haloesters                 | 3                | – | 1 (33%) | 2* (67%) | – |
| Haloamines/haloamides      | 6                | – | 2 (33%) | 4 (67%) | – |
| Haloaromatics              | 4                | – | 2 (50%) | 2 (50%) | – |
| Other halo-organics        | 3                | – | 1 (33%) | 2 (67%) | – |
| Nonhalogenated ketones     | 16               | – | 2 (12%) | 14 (88%) | – |
| Nonhalogenated aldehydes   | 20               | – | 1 (5%) | 4 (20%) | 15 (75%) |
| Nonhalogenated acids       | 54               | – | 2 (22%) | 7 (70%) | – |
| Nonhalogenated aromatics   | 5                | – | 2 (40%) | 3 (60%) | – |
| Other nonhalo-organics     | 12               | – | 2 (100%) | – | – |
| Inorganics                 | 2                | – | 2 (100%) | – | – |
| Total                     | 209              | 3 (2%) | 17 (8%) | 58 (28%) | 131 (62%) |

*Excluding several dihalo- and trihaloacetic acids previously selected for testing. **Compounds expected to hydrolyze in water instantaneously. Concern would be HM if exposure were via inhalation.

Halofuranones, MX, and Related Compounds

Within the halofuranones class, 3-chloro-4-(dichloromethyl)-5-hydroxy-2(5H)-furanone (MX) is the most well-known chemical. MX is the most potent, direct-acting mutagenic DBP ever tested in the Ames test (48). On a molar basis MX alone can account for up to 30–50% of the mutagenicity of chlorinated water (49). It is also a potent multitarget carcinogen in the rat (50). The upper-bound cancer risk per unit dose (oral slope factor) for lifetime exposure to MX (based on thyroid follicular adenomas in the rat) was estimated (51) to be 3.7 (mg/kg-day)-1. This number is not as high as would be expected from its bacterial mutagenic potency, indicating that MX may be readily detoxified in the body. The structure–mutagenicity relationships of MX and related compounds have been extensively studied using Ames Salmonella assay (49,52,53). MX is an extremely potent, direct-acting bacterial mutagen; its mutagenic activity can be substantially decreased by inclusion of S-9 mix. MX can undergo reversible cyclization between its closed-ring and open-ring forms, depending on the pH of the aqueous medium. In general, MX and related compounds, which are capable of undergoing cyclization reactions, are considerably more mutagenic than their corresponding compounds, which remain predominantly in the open-ring forms. For example, MX is at least 10 times more potent than (E)-2-chloro-3-(dichloromethyl)-4-oxobutenoic acid (EMX), the geometric isomer of the open-ring form of MX with limited capacity to cyclize (49). The hydroxy group at the 5 position, which facilitates the cyclization reaction, also has a profound effect on determining the mutagenicity. Elimination of the 5-OH group from MX (yielding 3-chloro-4-(dichloromethyl)-2-(5H)-furanone (red MX)) reduces the mutagenicity by 100-fold (49). Apparently, the closed-ring form, which is less hydrophilic than the open-ring form, may be required for optimal membrane penetration. It appears that the ultimate mutagen of MX-related compounds inside the cells may be their open-ring form, but they need to cyclize to closed-ring form outside the cells to facilitate membrane penetration. Substitution of chlorine by bromine has no appreciable effects on mutagenicity, as indicated by comparable mutagenicity among MX, 3-chloro-4-(bromochloromethyl)-5-hydroxy-2(5H)-furanone (BMX-1), 3-chloro-4-(dibromomethyl)-5-hydroxy-2(5H)-furanone (BMX-2), and 3-bromo-4-(dibromomethyl)-5-hydroxy-2(5H)-furanone (BMX-3) (53), whereas replacement of the 4-dichloromethyl group of MX by...
4-chloromethyl generates a less potent mucochloric acid (52, 54, 55).

On the basis of this SAR information, the cancer concern levels of the 10 MX-related DBPs in this study are summarized in Table 5, along with rationale and available genotoxicity data. The three chlorobromo analogs of MX (BMX-1, BMX-2, and BMX-3) are all given a high-moderate rating whereas mucochloric acid is given a moderate rating. Despite the lack of toxicity data, 2-MX are considered to be of low-moderate favorability cyclizing capacity, EMX and red-MX are given a high-moderate rating whereas mucochloric acid is given a moderate rating. On the basis of weaker mutagenicity and less favorable cyclizing capacity, EMX and red-MX are considered to be of low-moderate concern. Despite the lack of toxicity data, 2-MX are considered to be of low-moderate concern because the oxidation of the aldehyde group is expected to eliminate cyclizing capacity and may render the compounds too hydrophilic.

### Haloalkanes and Haloalkenes

Numerous haloalkanes and haloalkenes have been tested for carcinogenic and mutagenic activities; the SARs have been extensively studied (56). In general, the genotoxic potential is dependent on the nature, number, and position of halogen(s) and the molecular size of the compound. Short-chain monohaloalkanenogenated (excluding fluoride) alkanes and alklenes are potential direct-acting alkylating agents, particularly if the halogen is at the terminal end of the carbon chain or at an allylic position. Dihalogenated alkanes are also potential alkylating or cross-linking agents (either directly or after GSH conjugation), particularly if they are vicinally substituted (e.g., 1,2-dihaloalkane) or substituted at the two terminal ends of a short to medium-size (e.g., 2–7) alkyl moiety (i.e., α,ω-dihaloalkane). Fully halogenated haloalkanes tend to act by free radical or nongenotoxic mechanisms (such as generating peroxisome-proliferative intermediates) or undergo reductive dehalogenation to yield haloalkenes that in turn could be activated to epoxides. Haloalkanes are of concern because of potential to generate genotoxic intermediates after epoxidation. The concern for haloalkanes may be diminished if the double bond is internal or sterically hindered.

On the basis of the above SAR information, the cancer concern levels of the 14 haloalkanes and haloalkenes in this study are summarized in Table 6 along with rationale and available screening cancer bioassay.

#### Table 5. Halo furanones and MX-related compounds.

| DBP | Structure | Concern level | Rationale |
|-----|-----------|---------------|-----------|
| 1. 3-Chloro-4-(bromochloromethyl)-5-hydroxy-2(5H)-furanone (BMX-1) | ![Structure](image1) | LM | Structural analogy to MX with Cl 5-hydroxy-2(5H)-furanone expected to be less reactive. Positive genotoxicity data (Ames, E. coli, sister chromatid exchange in Chinese hamster ovary cells) but less active than MX (54, 55). |
| 2. 3-Chloro-4-(dibromomethyl)-5-hydroxy-2(5H)-furanone (BMX-2) | ![Structure](image2) | HM | The rationale above applies to BMX-2. |
| 3. 3-Bromo-4-(dibromomethyl)-5-hydroxy-2(5H)-furanone (BMX-3) | ![Structure](image3) | HM | The rationale above applies to BMX-3. |
| 4. 2,3-Dichloro-4-oxobutenoic acid (mucochloric acid; 3,4-dichloro-5-5-hydroxy-2(5H)-furanone) | ![Structure](image4) | LM | The diastereoisomer of the open-ring form of MX with limited capacity to cyclize. Positive Ames assay but much less potent than MX (49). |
| 5. (E)-2-Chloro-3-(dichloromethyl)-4-oxobutenoic acid (EMX) | ![Structure](image5) | LM | Structural analogy to MX but lacking the important 5-OH group. Positive Ames assay but much less potent than MX (52, 74). |
| 6. 3-Chloro-4-(dichloromethyl)-2-(5H)-furanone (red-MX) | ![Structure](image6) | LM | Structural analogy to MX which has been shown to be a multitarget carcinogen in the rat (50). Positive mutagenicity data in the Ames test for all three compounds with potency comparable to that of MX (53). |
| 7. Dihydro-4,5-dichloro-2(3H)-furanone | ![Structure](image7) | LM | Active chlorine, possibly acylating agent, possible GSH-mediated activation of vicinally substituted chlorine, may generate haloaldehyde after ring opening. |
| 8. 5-Hydroxy-5-trichloromethyl-2-furanone | ![Structure](image8) | LM | α,β-Unsaturated lactone that may undergo conjugate Michael addition with nucleophiles. Structural analogy to carcinogenic β-angelicalactone (69). |
| 9. 2-Chloro-3-(dichloromethyl)butenedioic acid (ox-MX) | ![Structure](image9) | Mar | Structural analogy to the open-ring form of MX, but oxidation of the aldehyde group eliminates cyclizing capability and may render the compound too hydrophilic. |
| 10. (E)-2-Chloro-3-(dichloromethyl)butenedioic acid (ox-EMX) | ![Structure](image10) | Mar | Structural analogy to the open-ring form of EMX, but oxidation of the aldehyde group eliminates cyclizing capability and may render the compound too hydrophilic. |
(pulmonary adenoma assay) and genotoxicity data. Five brominated and iodinated methanes and ethene derivatives are given a moderate rating. Beyond the fact that bromine and iodine are better leaving groups than chlorine, there is also evidence that brominated THMs may be preferentially activated by a theta-class glutathione S-transferase (GSTT1-1) to mutagens in Salmonella even at low substrate concentrations (57,58). Furthermore, there are human carcinogenicity implications because of polymorphism in GSTT1-1. Human subpopulations with expressed GSTT1-1 may be at a greater risk to brominate THMs than humans who lack the gene (57). Six, two, and one haloalkanes/ haloalkene(s) are given low-moderate, marginal, and low concern, respectively, with detailed rationale summarized in Table 6.

**Halonitriles**

There are basically three types of halonitriles detected as DBPs: a) halogenated acrylonitrile and higher congeners, b) halogenated acetonitriles and higher congeners, and c) cyanogen halides. The predicted concern levels of these compounds are summarized in Table 7 along with rationale and available screening data.

Three DBPs in this class are chlorinated acrylonitriles (cis-and trans-2,3,4,5-tetrachloro-2-butenenitrile and trichloropropenenitrile); they have all been given a moderate concern rating. Acrylonitrile is a well-known genotoxic rodent carcinogen (59). The introduction of halogens to acrylonitrile may reduce the potential to undergo Michael addition or epoxidation, but the terminal chlorine in cis- and trans-2,3,4-trichloro-2-butenenitrile may introduce an additional reactive terminal chlorine. Trichloropropenenitrile is of concern because of its structural analogy to tetrachloroethene.

Acetonitrile is not carcinogenic in rodents and is only weakly or marginally mutagenic (60). The introduction of halogen to α- and terminal carbons is expected to increase genotoxic potential by making it an alkylating/cross-linking agent. Halogenated acetonitriles have been tested in various cancer and genotoxicity screening assays. Table 8 summarizes and compares the available data. On the basis of alkylating activity, the brominated compounds are expected to be more reactive than chlorinated compounds. On the basis of data for chlorinated acetonitriles, and consistent with chemistry of halogenated compounds, increasing halogenation tends to decrease alkylating activity. Essentially mixed results have been observed in the screening assays. Despite their higher alkylating activity, monohalogenated acetonitriles tend to be inactive in a number of in vitro genotoxicity assays, probably because of complication by their higher cytotoxicity. There is some evidence that, in Comet, Chinese hamster ovary, and newt micronucleus assays, increasing chlorination increases the genotoxic potency (Table 8). However, this pattern is not seen in lung adenoma assay and skin tumor initiation studies in SENCAR mice. Probably the only consistent pattern seen across various assays is the higher activity of dibromoacetonitrile and bromochloroacetonitrile. Dibromoacetonitrile

### Table 6. Haloalkanes and haloalkenes.

| DBP                        | Structure | Concern level | Rationale |
|----------------------------|-----------|---------------|-----------|
| 1. Dibromomethane          | CH₂Br₂    | M             | Structural analogy to dichromethane, which is a rat carcinogen [47]. The brominated compound is expected to be more hazardous than the chlorinated compound because of more favorable leaving tendency and GSH-mediated activation. Positive genotoxicity (Ames, ara forward mutation, E. coli) data (75–78). |
| 2. Bromochloromethane      | CH₂BrCl   | M             | Structural analogy to dichromethane, which is a rat carcinogen [47]. The brominated compound is expected to be more hazardous than the chlorinated compound because of more favorable leaving tendency and GSH-mediated activation. |
| 3. Bromochloriodomethane   | CHBrCl    | M             | Structural analogy to bromodichromethane, which is a rodent carcinogen [47]. The iodo group is expected to be a better leaving group than chloro group. |
| 4. Dichloriodomethane      | CHClI     | M             | Structural analogy to bromodichromethane and chloroform, which are both carcinogenic [47]. The iodo group is expected to be an even better leaving group than the chloro/bromo group. |
| 5. 1,1,1-Tribromo-2-bromo-2-chloroethane | Br₂C–CHBrCl | M | Structural analogy to pentachloro-ethane, which is a mouse carcinogen [47], and 1,1,2,2-tetra-bromomethane, which is hepatotoxic. |
| 6. Chloromethane            | CH₃Cl     | LM            | Structural analogy to iodomethane [56] and chloroethane [47], which are both carcinogenic. Positive Ames assay [56]. |
| 7. 2-Chlorobutane (sec-butyl bromide) | CH₂CH₂CHBrCH₃ | LM | Positive lung adenoma assay and positive Ames assay [56]. The internal location of bromine may limit its genotoxic potential. |
| 8. 2,3-Dichlorobutane      | CH₂CH₂CHClCH₃ | LM | Vicinal dichloro substitution may lead to GSH-mediated activation, but internal location of chlorine may limit its genotoxic potential. |
| 9. 3,3,3-Trichloro-2-methyl-1-propane | Cl₃CCl₂CH₃ | LM | Structural analogy to 1,3-dichloropropane, which is a mouse carcinogen [47] but not as favorable because of steric hindrance by methyl and marginally active trichlor group. |
| 10. 1,2-Dichloro-2-methyl-butan | Cl₂CH₂O(C)(CH₂)CH₂CH₃ | LM | Structural analogy to 2-chloroisobutane, which is positive in the lung adenoma assay [58]. Vicinal substitution may lead to GSH-mediated activation, but methyl substitution may lead to steric hindrance. |
| 11. Tetrachlorocyclopropane | Cl₂ClClCl | LM | Limited structural analogy to hexachloropentadiene, which has negative bioassay data [47]. However, this compound may have some genotoxic potential. One of the chlorines at the bridged carbon may leave and generate a carbenion that can be stabilized by the ring by resonance stabilization. |
| 12. 1,1,5,5-Tetrachloropentane | Cl₃(C)(CH₂)CH₂Cl₂ | Mar | Potential alkylating agent, but its genotoxic potential may be reduced because the potentially reactive terminal carbons are both dichlorinated, making them not as favorable as mono chlorine as leaving groups. |
| 13. 1-Chlorooctane           | ClO₂H₂ (CH₂)Cl₂H₃ | Mar | Despite the presence of a terminal chlorine, this compound is expected to be a weak alkylating agent because of its high molecular weight and its saturated chain. |
| 14. 2-Chlorododecane        | CH₂CH₂Cl (CH₂)Cl₂H₃ | L | Expected to be a very weak alkylating agent because of its high molecular weight and its saturated chain. |
Table 7. Halonitriles and cyanogen halides.

| DBP | Structure | Concern level | Rationale |
|-----|-----------|---------------|-----------|
| 1. cis-2,3,4-Trichloro-2-butene nitrile | Cl\_2\_CN | M | This compound is a substituted acrylonitrile, a probable human carcinogen (79). Although the substitutions may reduce potential to undergo Michael addition or epoxidation, the terminal active chlorine may provide additional genotoxic potential. |
| 2. trans-2,3,4-Trichloro-2-butene nitrile | Cl\_2\_CN | M | This compound is a substituted acrylonitrile, a known rodent carcinogen (79). Although the substitutions may reduce potential to undergo Michael addition or epoxidation, the terminal active chlorine may provide additional genotoxic potential. |
| 3. Bromochloroacetonitrile | BrCl\_CN | M | This compound has active halogen. Positive skin tumor initiator (80), positive in lung adenoma assay (81), and positive genotoxicity (SOS, Ames, sister chromatid exchange, nev induced micronucleus data) (47, 69, 82–84). |
| 4. 2,3-Dichloro-3-bromopropanenitrile | BrCl\_CH(CN) | M | This compound has vicinally substituted active halogens and may have genotoxic potential via GSH-mediated activation. Concern level at the low end of M. |
| 5. 3,4-Dichlorobutanenitrile | Cl\_CH\_CN | M | This compound has vicinal dichloro substitution and may have genotoxic potential via GSH-mediated activation. Concern level at the low end of M. |
| 6. Trichloropropenenitrile | Cl\_2\_CN | M | This compound is a substituted acrylonitrile, a probable human carcinogen (79), but the chlorines may reduce the potential to undergo Michael addition or epoxidation. Because cyano group may be considered a pseudo halogen, the compound is also a structural analog of perchloroethylene, a rodent carcinogen (47). |
| 7. Bromoacetonitrile | Br\_CN | LM | This compound has an active bromine but negative or mixed genotoxicity data (69, 83, 84), due possibly to its cytotoxicity. |
| 8. Chloroacetonitrile | Cl\_CN | LM | This compound has an active chlorine. Positive skin tumor initiator (80) and positive in lung adenoma assay (81) but negative or mixed genotoxicity data (69, 83, 84) due to possibly to its cytotoxicity. |
| 9. Dichloroacetonitrile | Cl\_2\_CN | LM | This compound is structurally related to trichloroacetonitrile, with bromine expected to be a better leaving group than chlorine. |
| 10. Tribromoacetonitrile | Br\_3\_CN | LM | This compound is structurally related to chloroacetonitrile, with bromine expected to be a better leaving group than chlorine. |
| 11. Bromodichloroacetonitrile | Br\_Cl\_CN | LM | This compound is structurally related to chloroacetonitrile, with bromine expected to be a better leaving group than chlorine. |
| 12. Dibromochloroacetonitrile | Br\_2\_Cl\_CN | LM | This compound is structurally related to chloroacetonitrile, with bromine expected to be a better leaving group than chlorine. |
| 13. Trichloroacetonitrile | Cl\_3\_CN | LM | This compound is positive in skin tumor initiation and lung adenoma assays (80, 81) but has mixed genotoxicity data (69, 83, 84). |
| 14. Cyanogen chloride | ClCN | L | This compound is known to be readily metabolized to cyanide in the body. The expected high acute toxicity should limit significant exposure. There is also no structural alert suggestive of cancer concern. |
| 15. Cyanogen bromide | BrCN | L | This compound is expected to behave in the same way as cyanogen chloride. |

Table 8. Comparison of chemical, biochemical, and biologic properties of haloacetonitriles.

| Chemical/biochemical/biologic properties relevant to assessing carcinogenic potential | Relative order of potency of haloacetonitriles tested | Reference |
|-----------------------------------------------|-----------------------------------------------|-----------|
| Alkylating activity 4-|\_lp-nitrobenzyl|lp|nitrobenzyl|pyridine reaction) | Br\_2\_3 \_Cl \_Cl \_Cl \_Cl | (82) |
| Inhibition of glutathione S-transferase | Cl\_3 > Br\_3 > Br\_2 > Cl\_2 > Cl (inactive) | (69, 83) |
| E. coli/SOS chromotest | BrCl > Br\_2 > Cl\_2 > Cl (inactive) | (83) |
| Ames or Ames fluctuation tests | Cl\_2 > Br\_2 > Br\_Cl > Br\_Cl > Br\_Cl > Cl (inactive) | (69, 84) |
| DNA single-strand breaks in HeLa cell (comet assay) | Br\_2 > Br\_Cl > Br\_Cl > Cl (inactive) | (69, 84) |
| Sister chromatid exchanges in Chinese hamster ovary cells | Br\_2 > Br\_Cl > Br\_Cl > Cl (inactive) | (69, 84) |
| Neutrophil microneucleus assay | Br\_2 > Br\_Cl > Br\_Cl > Cl (inactive) | (63) |
| In vivo mouse micronucleus assay | Br\_2 > Br\_Cl > Cl (all inactive) | (80) |
| Lung adenoma assay in strain A mice | Cl\_2 > Br\_Cl > Br\_Cl > Cl (inactive) | (81) |
| Skin tumor initiation in SENCAR mice | Br\_2 > Br\_Cl > Br\_Cl (inconsistent) > Cl (inactive) | (80) |

halogenation at both α-carbons may lead to unstable compounds. The stability of several chlorinated ketones in aqueous solutions follows this order: 1,3- dichloro > pentachloro > hexachloro (61). A variety of haloketones have been tested in various screening assays. Consistent with

Haloketones

Haloketones with monosubstitution with chlorine or bromine at the α-carbon or terminal carbon are expected to be potential alkylating agents. Haloketones with active halogen at both ends of the aliphatic chain are expected to be cross-linking agents. The leaving tendency of halogen decreases with an increase in the degree of halogenation as the electron-withdrawing effect of the second and/or third halogen diminishes the leaving potential of the first halogen (22, 48, 56). On the other hand, haloketones

has already been selected for testing (Table 2). In this study, bromochloroacetanilide has been given a moderate concern, whereas all other halogenated acetanilides have been given a low-moderate concern. Two higher homologs of bromochloroacetanilides (2,3-dichloro-3-bromopropanenitrile and 3,4-dichlorobutanenitrile) have also been considered of moderate concern because of SAR consideration, although they should be at the low end of the moderate category.

Cyanogen chloride and cyanogen bromide have been given a low concern. They are known or expected to be metabolized to cyanide in the body. The expected high acute toxicity should limit significant exposure. There are also no structural alerts suggestive of carcinogenic potential.

Table 7. Halonitriles and cyanogen halides.

| DBP | Structure | Concern level | Rationale |
|-----|-----------|---------------|-----------|
| 1. cis-2,3,4-Trichloro-2-butene nitrile | Cl\_2\_CN | M | This compound is a substituted acrylonitrile, a probable human carcinogen (79). Although the substitutions may reduce potential to undergo Michael addition or epoxidation, the terminal active chlorine may provide additional genotoxic potential. |
| 2. trans-2,3,4-Trichloro-2-butene nitrile | Cl\_2\_CN | M | This compound is a substituted acrylonitrile, a known rodent carcinogen (79). Although the substitutions may reduce potential to undergo Michael addition or epoxidation, the terminal active chlorine may provide additional genotoxic potential. |
| 3. Bromochloroacetonitrile | BrCl\_CN | M | This compound has active halogen. Positive skin tumor initiator (80), positive in lung adenoma assay (81), and positive genotoxicity (SOS, Ames, sister chromatid exchange, nev induced micronucleus data) (47, 69, 82–84). |
| 4. 2,3-Dichloro-3-bromopropanenitrile | BrCl\_CH(CN) | M | This compound has vicinally substituted active halogens and may have genotoxic potential via GSH-mediated activation. Concern level at the low end of M. |
| 5. 3,4-Dichlorobutanenitrile | Cl\_CH\_CN | M | This compound has vicinal dichloro substitution and may have genotoxic potential via GSH-mediated activation. Concern level at the low end of M. |
| 6. Trichloropropenenitrile | Cl\_2\_CN | M | This compound is a substituted acrylonitrile, a probable human carcinogen (79), but the chlorines may reduce the potential to undergo Michael addition or epoxidation. Because cyano group may be considered a pseudo halogen, the compound is also a structural analog of perchloroethylene, a rodent carcinogen (47). |
| 7. Bromoacetonitrile | Br\_CN | LM | This compound has an active bromine but negative or mixed genotoxicity data (69, 83, 84), due possibly to its cytotoxicity. |
| 8. Chloroacetonitrile | Cl\_CN | LM | This compound has an active chlorine. Positive skin tumor initiator (80) and positive in lung adenoma assay (81) but negative or mixed genotoxicity data (69, 83, 84) due to possibly to its cytotoxicity. |
| 9. Dichloroacetonitrile | Cl\_2\_CN | LM | This compound is structurally related to trichloroacetonitrile, with bromine expected to be a better leaving group than chlorine. |
| 10. Tribromoacetonitrile | Br\_3\_CN | LM | This compound is structurally related to chloroacetonitrile, with bromine expected to be a better leaving group than chlorine. |
| 11. Bromodichloroacetonitrile | Br\_Cl\_CN | LM | This compound is structurally related to chloroacetonitrile, with bromine expected to be a better leaving group than chlorine. |
| 12. Dibromochloroacetonitrile | Br\_2\_Cl\_CN | LM | This compound is structurally related to chloroacetonitrile, with bromine expected to be a better leaving group than chlorine. |
| 13. Trichloroacetonitrile | Cl\_3\_CN | LM | This compound is positive in skin tumor initiation and lung adenoma assays (80, 81) but has mixed genotoxicity data (69, 83, 84). |
| 14. Cyanogen chloride | ClCN | L | This compound is known to be readily metabolized to cyanide in the body. The expected high acute toxicity should limit significant exposure. There is also no structural alert suggestive of cancer concern. |
| 15. Cyanogen bromide | BrCN | L | This compound is expected to behave in the same way as cyanogen chloride. |
Table 9. Haloketones.

| Compound | Structure | Concern level | Rationale |
|----------|-----------|---------------|-----------|
| 1. 1,3-Dichloropropane | ClCH2(O)CH2Cl | M | This compound is a potential cross-linking agent via its active chlorine at both termini. Positive skin tumor initiator (63) and positive genotoxicity data (Ames, SOS, newt micronucleus) and strong GSH depletor (48,86,87). |
| 2. 1,1,3-Trichloropropane | ClCH(O)CH2Cl | M | This compound is a potential cross-linking agent. Negative skin tumor initiator (63) but consistently positive genotoxicity data (48,88). Concern level at the low end of M. |
| 3. 1,1,3,3-Tetrachloropropanone | ClCH(O)CHCl2 | LM | This compound has the potential to be a cross-linker via its chlorines at both termini, but the disubstituted chlorines are not as reactive as monosubstituted chlorine. Positive Ames (48). |
| 4. 1,1,1,3-Tetrachloropropanone | ClCCl(O)CHCl | LM | This compound has the potential to be a cross-linker via its chlorines at both termini, but the trisubstituted chlorines are only marginally active. |
| 5. Chloropropane | ClCH2(O)CH3 | LM | This compound has an active chlorine and is a good GSH depletor (67). Negative skin tumor initiator (63); mixed genotoxicity (+SOS, +/-Ames, –newt micronucleus) data (48,86,87). |
| 6. 1,1-Dichloropropanone | ClCH(O)CH3 | LM | Structural analogy to 1,1-dichloropropanone but with chlorine expected to be a better leaving group than chlorine. |
| 7. 1,1-Dibromopropanone | BrCH(O)CH3 | LM | Structural analogy to 1,1-dichloropropanone but with bromine expected to be a better leaving group than chlorine. |
| 8. Pentachloropropanone | Cl5CO(O)CH3 | LM | Marginal activity. Ames test (48), but the extensive chlorine substitution makes the compound unstable at high pH (68). |
| 9. 1,1,1-Trichloropropanone | Cl3CC(O)CH3 | LM | The chlorines at the trisubstituted terminus are only marginally active. Negative skin tumor initiator (63). Weak or mixed genotoxicity (+SOS, +/-Ames, –newt micronucleus) data (48,88). Concern level at low end of LM. |
| 10. 1-Bromo-1,1-dichloropropanone | BrCl2CC(O)CH3 | LM | Structural analogy to 1,1,1-trichloropropanone. The halogens are only marginally active. Concern level at low end of LM. |
| 11. Hexachloropropanone | Cl3CC(O)CCl3 | LM | Marginally active chlorines. Ames test is negative or inconsistent and solvent dependent (48,89). The extensive chlorine substitution makes the compound unstable even at near neutral pH (89). Concern level at low end of LM. |
| 12. 1,1-Dichloro-2-butanone | CH3CH2OCH2Cl | LM | Structural analogy to 1,1-dichloropropanone. The chlorines are somewhat active. Concern level at low end of LM. |
| 13. 1,1-Dichloropropanone | ClCH(O)CH2Cl | LM | The chlorines are somewhat active. Negative skin tumor initiator (63). Weak or mixed genotoxicity (+SOS, +/-Ames, –newt micronucleus) data (48,88,87). Concern level at low end of LM. |
| 14. 1,1,1-Trichloro-2-butanone | CH3CH2OCCl3 | LM | Structural analogy to 1,1,1-trichloropropanone. The chlorines at the trisubstituted terminus are expected to be marginally active. Concern level at low end of LM. |
| 15. 2,2-Dichloro-1,3-cyclohexadienone | O | LM | This compound is an α,β-unsaturated ketone with at least one β-position available for Michael addition. The pentenedione chlorines at 2-position may be somewhat active. Concern level at low end of LM. |
| 16. 2-Chlorocyclohexanone | | LM | The unsaturated cyclohexanone is a weak to marginally active carcinogen (90). The introduction of active chlorine at the a-carbon expected to increase genotoxic potential, but the rigid ring may limit its potential. Concern level at low end of LM. |
| 17. 3,3-Dichloro-2-butane | ClCH2Cl2CHCl2 | Mar | The chlorines are only marginally active and are not terminal. |
| 18. 2,2-Dichloro-3-pentane | ClCH2ClCHCl2CH3 | Mar | The chlorines are only marginally active and are not terminal. |

their potential chemical reactivity as alkylating agents, three chloropropanones have been shown to react directly with GSH. Their relative potency follows this order: 1,3-dichloro > monochloro > 1,1-dichloro (62). Among five chloropropanones (mono-, 1,1-, 1,3-, 1,1,1-, and 1,1,3-) tested for skin tumor-initiating activity in SENCAR mice, only 1,3-dichloropropanone showed clearly positive results (63). With the exception of 1,1,1,3-tetrachloropropanone, all congeners of chloropropanones have been tested for mutagenicity in the Ames test. Among the mutagenic chloropropanones (mostly direct-acting), the relative mutagenic potency follows this order: 1,3- > 1,1,3,3- > penta- > 1,1,3- > 1,1,1- > 1,1- with the potency of 1,3- being about 100 to 1,000 times higher than that of 1,1- (48,62). Inconsistent results have been observed in the Ames test on monochloropropanone because of its high cytotoxicity (which to some extent can be attenuated by inclusion of 59% mix) and on hexachloropropanone, which is relatively unstable in water (61). 1,3-Dichloro and, to a lesser extent, 1,1,3-trichloro congeners have also been consistently found to be more mutagenic than monoo-, 1,1-, and 1,1,1-congeners in E. coli SOS chromotest for DNA damage (SOS), Ames fluctuation, and newt micronucleus tests (64).

Based on the above SAR and screening data, the cancer concern levels of 19 haloketones are summarized in Table 9 along with rationale and available data. Only 1,3-dichloropropanone and, to a lesser extent, 1,1,3-trichloropropanone have been given a moderate concern. Most of the other haloketones have been given a low-moderate concern, although there may be slight differences within the low-moderate category as detailed in the rationale of individual compounds.

**Nonhalogenated Aldehydes**

As a class, aldehydes have been given special attention tailored to drinking water consideration. Essentially, aldehydes are electrophilic, reactive chemicals that may form DNA–protein cross-links and induce carcinogenesis/mutagenesis. A variety of aldehydes have been tested for carcinogenic activity (65). By the inhalation route formaldehyde and, to a much lesser extent, acetaldehyde are carcinogenic, whereas isobutyraldehyde is not carcinogenic even at doses that cause irritation to the respiratory tract. There is
some suggestive evidence that acetaldehyde may be a potential ultimate carcinogen in alcoholics with genetically deficient detoxifying capabilities; however, the subject remains to be resolved. By the oral route, the α,β-unsaturated aldehyde, crotonaldehyde, is carcinogenic, whereas acrolein is equivocal, probably because it is too reactive.

Numerous aldehydes have been tested for mutagenic activity. In general, only short-chain aldehydes (e.g., formaldehyde, acetaldehyde) have been clearly shown to be mutagenic. The genotoxic potential of aldehydes decreases substantially with an increase in molecular size. The introduction of hydrophilic groups generally decreases activity, whereas α,β-unsaturation tends to increase the genotoxic potential provided that the β-position is not sterically hindered (66).

Although short-chain aldehydes such as formaldehyde and acetaldehyde are carcinogenic in animals by inhalation, their carcinogenic potential by the oral route may be limited unless exposure occurs in high doses that overwhelm the detoxification mechanisms or to susceptible individuals. There is some evidence that hexamethylenetetramine, which is known to be hydrolyzed to formaldehyde and shown to induce local sarcomas by injection, has no carcinogenic activity when tested by the oral route (65). With the exception of α,β-unsaturated aldehydes, our assessment of the cancer hazard potential of aldehydes is based on the assumption that the principal route of exposure is oral and that the general population has adequate capacity to detoxify environmentally levels of aldehydes. Humans are known to have genetic polymorphism in aldehyde dehydrogenase-2 (ALDH-2), and there is some suggestive (67) but inconsistent (68) evidence that subpopulations with deficient ALDH-2 may be at a higher cancer risk to acetaldehyde generated from consuming alcohol.

Among the nonhalogenated aldehydes considered in this study, butanediol is the only compound that has been given a moderate concern. Despite the lack of toxicity data, butanediol has been given a higher concern than the rest of the compounds because it has two terminal reactive aldehydes separated by two methylene groups, which should make it a highly favorable cross-linking agent. Four aldehydes (methyl glyoxal, cyanoformaldehyde, 2-hexenal, and propional) are considered to be of low-moderate concern if they could be found in water in significant amounts. Higher molecular-weight aldehydes are not of significant concern by SAR consideration and comparison to isobutyraldehyde, which is not carcinogenic even by the inhalation route.

Summary and Conclusions

Determining appropriate drinking water DBP regulations is a complex problem. Disinfectants are necessary to protect against waterborne pathogens, and thus DBPs are unavoidable. Source water quality and constituents vary widely throughout the United States. Combined with the assortment of disinfectants available, this means that DBPs differ from site to site in both occurrence and concentration. Along with a number of DBPs that have some occurrence data, there are hundreds of chemicals that have been identified as DBPs but that have no quantitative occurrence data beyond this single identification. The conundrum presented by these hundreds of identified DBPs is how to determine research priorities. Two important factors to consider in setting regulations are the toxicity of the chemical and the concentration at which the chemical is found. For the majority of the chemicals in this article, no data were available on either factor. Gathering occurrence data and toxicity testing are both expensive and time-consuming activities. SAR analysis is essential in narrowing down health research priorities because it is time and cost effective. The U.S. EPA efforts are ongoing to gather occurrence data for a number of DBPs of higher concern.

It is encouraging from a public health standpoint that although more than 200 DBPs were analyzed, only 20 were of moderate or higher concern for carcinogenic potential. Of these, four are structurally related to MX, which is believed to occur at very low levels (nanograms per liter), and are thus likely not of great concern. Five others are halogenated alkanes, which presumably will be controlled by existing and future THM regulations. As a result of this analysis, the most suitable candidates for testing are the halonitrites and haloketones that are in the moderate concern category, dibromomethane and butanediol.

REFERENCES AND NOTES

1. Hughes JM. Infectious diseases transmitted by drinking water in the United States: Perspectives of the Centers for Disease Control and Prevention. In: Safety of Water Disinfection: Balancing Chemical and Microbial Risks (Craun GF, ed, Washington, DC:ISIL Press, 1993:11–16.
2. Rook JJ. Formation of haloforms during chlorination of natural waters. Water Treat Exam 23:234–245 (1974).
3. Beller TA, Lichtenberg JJ, Kroner RC. The occurrence of organohalides in chlorinated drinking water. J Am Water Works Assoc 66:703–706 (1974).
4. U.S. Environmental Protection Agency. (U.S. EPA). National Interim Primary Drinking Water Regulations; Control of Trihalomethanes in Drinking Water. Fed Reg 41(23):66824–66870 (1974).
5. U.S. Environmental Protection Agency. 40 CFR Parts 9, 141, and 142. National Primary Drinking Water Regulations: Disinfectants and Disinfection Byproducts; Final Rule. Fed Reg 60(35):60399–60476 (1995).
6. U.S. Environmental Protection Agency. 40 CFR Parts 9, 141, and 142. National Primary Drinking Water Regulations: Interim Enhanced Surface Water Treatment; Final Rule. Fed Reg 63(24):68478–69521 (1998).
7. Public Health Service Act. Title XIV [Safe Drinking Water Act](PL 104–182).
8. U.S. Environmental Protection Agency. National Primary Drinking Water Regulations: Monitoring Requirements for Public Drinking Water Supplies; Final Rule. Fed Reg 61(94):24304–24398 (1996).
9. Moudgil C.J, Rice G, Bruce RM, Teuchler L, Richardson S. Estimating risk posed by drinking water disinfection byproducts. Toxicologist 60:142 (2001).
10. Boorman GA, Delarco V, V, Dinnick JK, Chapin RE, Hunter S, Hauchman F, Gardner H, Cox M, Sils RC. Drinking water disinfection byproducts: review and approach to toxicity evaluation. Environ Health Perspect 107(suppl 1):207–217 (1999).
11. Cantor KP, Houper R, Hartge P. Drinking water source and bladder cancer: a case-control study. In: Water Chlorination: Chemistry, Environmental Impact and Health Effects, Vol 5 (Jolley RL, Bull RJ, Davis WP, eds). Chelsea, MI:Lewis Publishers, 1985:145–152.
12. Cantor KP, Houper R, Hartge P, Mason TJ, Silverman DT, Altman R, Austin DF, Child MA, Key CR, Marrett LD. Bladder cancer, drinking water source and tap water consumption: a case control study. J Natl Cancer Inst 79:1269–1279 (1987).
13. Cantor KP, Lynch CF, Hildesheim M, Dossmeci M, Lubin J, Alavanja M, Craun GF. Drinking water source and chlorination byproducts. I: Risk of bladder cancer. Epidemiology 9:21–28 (1998).
14. Crable DL, Shy CM, Sturrock RJ, Siff EJ. A case-control study of colon cancer and chlorine nitration in North Carolina. In: Water Chlorination: Chemistry, Environmental Impact and Health Effects, Vol 5 (Jolley RL, Bull RJ, Davis WP, eds). Chelsea, MI:Lewis Publishers, 1985:153–159.
15. Doyle TJ, Sheng W, Chen JR, Hong CP, Sellers TA, Kushi LH, Folsom AR. The association of drinking water source and chlorine nitration byproducts with cancer incidence among postmenopausal women in Iowa: a prospective cohort study. Am J Public Health 87(7):1166–1176 (1997).
16. Freedman M, Cantor KP, Lee NL, Chen LS, Lee HH, Ruhl CE, Wang SS. Bladder cancer and drinking water: a population-based case-control study in Washington County, Maryland (United States). Cancer Causes Control 8:738–744 (1997).
17. Hildesheim ME, Cantor KP, Lynch CF, Dossmeci M, Lubin J, Alavanja M, Craun GF. Drinking water source and chlorination byproducts: risk of colon and rectal cancers. Epidemiology 9(1):29–35 (1998).
18. King WD, Marrett LD. Case-control study of bladder cancer and chlorination byproducts in treated water (Ontario, Canada). Cancer Causes Control 7:596–604 (1996).
19. McGehee MA, Reif JS, Becher JC, Mangione EJ. Case-control study of bladder cancer and water disinfection methods in Colorado. Am J Epidemiol 138:492–501 (1993).
20. Richardson SD, Thruston AD Jr, Caughran TV, Chen PH, Collette TW, Floyd TL, Schenck KM, Lykins BW Jr, Sun G, Majetic G. Identification of new ozone disinfection byproducts in drinking water. Environ Sci Technol 33:3398–3377 (1999).
21. Suzuki N, Nakamishij K. Brominated analogues of MX (3-chloro-4-(dichloromethyl)-5-hydroxy-2(5H)-furanone) in chlorinated drinking water. Chemosphere 36(6):1557–1564 (1998).
22. Richardson SD. Drinking water disinfection byproducts. In: Encyclopedia of Environmental Analysis and Remediation, Vol 3. Meyers RA, ed). New York:John Wiley & Sons 1998:1396–1421.
23. U.S. Environmental Protection Agency Integrated Risk Information System. Available: http://www.epa.gov/irisweb/iris/index.html (cited 2 January 2002).
24. Woot Y-T, Lai DY, Argus MF, Arcos JC. Development of an integrated approach of combining mechanistically complementary short-term predictive tests as a basis for assessing the carcinogenic potential of chemicals. Environ Carcinog Ecotoxicol Rev (Part C of J Environ Sci Health) C16(2):101–122 (1998).
25. Ashby J, Tennant RW. Prediction of rodent carcinogenicity for 44 chemicals: results. Mutagenesis 11(1):15–19 (1994).
26. Woot Y-T, Lai DY, Argus MF, Arcos JC. Development of structure-activity relationship rules for predicting carcinogenic potential of chemicals. Toxicol Let 79:219–228 (1995).
27. Woot Y-T, Lai DY, Arcos J, Argus M, Cinomo M, Devoito S, Keller L. Mechanism-based structure-activity relationship (SAR) analysis of carcinogen potential of nitro aromatics. Environ Carcinog Ecotoxicol Rev (Part C of J Environ Sci Health) C15(2):139–160 (1997).
28. Richard AM, Besigni R Al and SAR approaches for pre-
