Background

Naphthalene has been demonstrated to cause respiratory tumors in rats and mice, but the few available epidemiologic studies are inadequate to evaluate the potential for naphthalene to cause cancer in humans. In lieu of human studies, mechanistic information may be used to inform the potential carcinogenicity of naphthalene for human health risk assessment.

Multiple modes of action (MOAs) for naphthalene-induced carcinogenicity have been proposed based on animal and in vitro studies, including genotoxicity, cytokystosis, and sustained regenerative cell proliferation.

While these proposed MOAs may differ in specific key events, the formation of toxic naphthalene metabolites and the biological relevance of these toxic metabolites to humans has emerged as a key component in answering the question of applicability of carcinogenic risk to humans. There is a great deal of similarity between the rodent and human naphthalene metabolic pathways; however, the activity of the enzymes involved in naphthalene metabolism and therefore the number of metabolites and stereoisomers of the produced metabolites may differ between rodents to humans.

Here, concurrent with a broad systematic review of health effects related to naphthalene exposure, and animal and in vitro studies of the available mechanistic evidence was analyzed to (1) integrate the available evidence in vitro models on the formation and toxicity of each of the key toxic metabolites of naphthalene and (2) determine the biological plausability that each of these key metabolites could be generated in human tissue and increase human oncogenic risk.

Methods

Literature Search and Tagging: Mechanistic studies were identified by tagging studies during screening of the broad literature search focused on the potential human health impacts associated with naphthalene exposure.

Study evaluation: Studies tagged as mechanistic were evaluated using the SciRAP web tool (www.scirap.org) for either in vivo or in vitro study evaluation for factors rated to reporting quality, mechanistic quality, and relevance. SciRAP was selected for this evaluation because it has both in vivo and in vitro study evaluation tools available.

Evidence synthesis: For the specific question of metabolic relevance, we used the metabolic pathway for naphthalene (developed from rodent models) as a scaffold and then evaluated studies that addressed the applicability of this metabolic pathway to humans. Focusing on three key naphthalene metabolites (Figure 1): 1,2-naphthoquinone, 1,2-naphthaldehyde, and 1,2,4-naphthoquinone. Studies that had deficiencies in reporting critical important study data (e.g., missing experimental exposure data) were excluded.

The evidence regarding the formation, toxicity, and human relevance of these three key naphthalene metabolites was integrated in a tabular format describing the formation and toxicity of each metabolite, factors that increase strength of evidence, and factors that decrease strength of evidence (Table 1).

Evidence Synthesis

The evidence synthesis determined that the formation and toxicity of these human metabolites was not well studied and has also been supported by the results of studies that found these metabolites to be linked to cancer and other health effects in humans (Table 1).

The available evidence showed that 1,2,5-naphthaldehyde (the prevalent naphthalene metabolite in humans) is a highly reactive metabolite that is more toxic and metabolized more slowly than the 1R,2S enantiomer more commonly observed in mice, which may add more time to produce cytotoxicity.

1,2,5-naphthaldehyde can be metabolized to 1,2-naphthoquinone and 1,4-naphthoquinone (Figure 1), which have been shown to elicit cytotoxicity. These quinone metabolites bind to proteins and have been demonstrated in situ and across species (including non-human primate tissues) to form protein adducts. In addition, these quinones may also undergo protein adduction and disrupt normal cellular function by binding to CYF460 enzymes and to proteins involved in cell signaling and transduction.

The electrophile nature of 1,2- and 1,4-naphthoquinone cause these metabolites to undergo 1,4-Michael addition and covalently bind to DNA, forming depurinating N3Me and N7Gua adducts as well as stable adducts. Therefore, it is biologically plausible for the reactive naphthalene metabolites 1,2- and 1,4-naphthoquinone to form depurinating and stable DNA adducts.

Table 1 Evidence profile table describing a summary of the toxicological evidence for each of the known naphthalene metabolites

<table>
<thead>
<tr>
<th>Metabolite</th>
<th>Toxicity</th>
<th>Human Relevance</th>
<th>Additional Information</th>
</tr>
</thead>
<tbody>
<tr>
<td>1,2-naphthoquinone</td>
<td>Cytotoxicity, DNA adduct formation</td>
<td>Human relevance demonstrated</td>
<td>Linked to CYF460 enzymes and to proteins involved in cell signaling and transduction.</td>
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<td>Cytotoxicity, DNA adduct formation</td>
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Figure 1. Naphthalene Metabolic Pathway

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Targeted Mechanistic Evidence Synthesis to Inform Evidence Integration Decisions on the Potential Human Carcinogenicity of Naphthalene Exposure

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