# Diagnosing the Common Causes of Regulatory Toxicity Involving Pulp and Paper Mill Effluents

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## Abstract

Effluent discharges from pulp and paper mills in Canada are regulated for toxicity. The regulation requires 50% survival of rainbow trout (*Oncorhyncus mykiss*) and *Daphnia magna* exposed to full-strength (i.e., 100%) effluent in 96 h and 48 h toxicity tests, respectively. Approximately 25% of mills have one or more toxicity episodes per year. For these mills, understanding the cause of effluent toxicity is an essential requirement for its prevention and remediation. Since 1996, Paprican has investigated over 80 toxicity episodes involving mill effluents with an approximately 70% success rate. Based on this experience, a diagnostic strategy was devised to help others troubleshoot the most common causes of toxicity episodes and thus facilitate the correct remedial steps for returning to compliance. The overall strategy is a two-step process that involves formulation of a hypothesis regarding the suspected cause of toxicity followed by diagnostic tests for confirmation purposes. The selection of the suspected cause of toxicity is based on toxicological properties of causative agents as well as an understanding of how mill operating conditions can influence these properties, criteria from the toxicity test results, and a review of the mill operating conditions. The strategy is expected to be sufficient for correctly diagnosing about 70% of the effluent toxicity episodes.

## Keywords

PULP MILLS, PAPER MILLS, EFFLUENTS, TOXICITY, REGULATIONS, DIAGNOSIS, TESTING, FISHES.

## Introduction

Effluent discharges from pulp and paper mills in Canada are regulated for toxicity [1]. The regulation requires 50% survival of rainbow trout (*Oncorhyncus mykiss*) and *Daphnia magna* exposed to full-strength (i.e., 100%) effluent in 96 h and 48 h toxicity tests, respectively. Approximately 25% of mills have one or more toxicity episodes per year [2]. For these mills, understanding the cause of effluent toxicity is an essential requirement for its prevention and remediation.

The U.S. Environmental Protection Agency (EPA) has issued protocols [3-5] for toxicity identification evaluation (TIE). However, these are not specific for any particular industrial effluent and, as such, some effort may be spent on searching for unlikely causative agents. Hence, there is a need for a simplified diagnostic strategy that is industry specific and could be routinely used to troubleshoot at least the most common causes of toxicity episodes.

Since 1996, Paprican has investigated over 80 toxicity episodes involving biotreated mill effluents [6]. In about 70% of the cases the cause of toxicity could be traced to resin acids, ammonia, carbon dioxide,

polymeric formulations, copper, manganese, dimethyl disulfide and, in one case, possibly nitrite. Knowing the potential causes of mill effluent toxicity provided an opportunity to streamline the existing U.S. EPA TIE protocols for the pulp and paper sector. This paper describes a streamlined strategy for identifying the most common causes of toxicity.

## Overall Strategy

The overall strategy for diagnosing the common cause(s) of regulatory effluent toxicity is a two-step process that involves the formulation of a hypothesis regarding the suspected cause of toxicity followed by simple diagnostic tests for confirmation purposes. The starting point for the formulation of the hypothesis is the list of possible causative agent(s) identified in our investigation of over 80 toxicity episodes [6]. Then, selection criteria, including the toxicological properties of the common causative agents, observations from toxicity tests and details of mill operating conditions, are used to determine which of these could be responsible for toxicity. The subsequent diagnostic tests involve chemical analysis and certain aspects of the generic U.S. EPA TIE protocols [3-5]. The details of the strategy are described below. If the two-step process fails to identify the toxicant, then it is necessary to address such cases according to more elaborate TIE approaches.

## Details of the Diagnostic Strategy

### Common Causes of Effluent Regulatory Toxicity (Table 1)

A summary of common causes of regulatory toxicity involving biotreated mill effluents as well as their source is shown in Table 1. The causative agents in Table 1 can originate from the wood furnish, from the biotreatment plant, from additives used by the mill and from process derivatives (compounds formed during the manufacturing process).

TABLE 1
List of causes of regulatory effluent toxicity episodes identified from 84 Paprican case studies (Kovacs et al. 2003).

| Cause of Toxicity | Source |
|---|---|
| Resin acids | Wood furnish: softwoods |
| Ammonia/nitrite | Nutrient addition to biotreatment system |
| Carbon dioxide | Metabolic end product of microorganisms in sealed biotreatment systems |
| Polymeric formulations | Additives used for retention, drainage and flocculation |
| Dimethyl disulfide | Kraft pulping process derivative |
| Trace metals (copper, manganese) | Wood, additives, contamination, process chemicals |

### Selection Criteria (Tables 2 and 3)

The criteria for selecting a suspected cause of toxicity in Table 1 are based upon knowledge of toxicological properties (Table 2), toxicity test observations and consideration of mill operating conditions (Table 3).

**Toxicological properties (Table 2)**

The key toxicological properties of the common causative agents of mill effluent toxicity are listed in Table 2. Except for most polymeric formulations, the other causative agents in Table 2 are more toxic to trout than to *D. magna.* The toxicity thresholds and the role of important modifying factors listed in Table 2 are based on tests done at Paprican as well as information from the literature. The toxicity of these agents is greatly dependent on the modifying effects of the effluent matrix, particularly pH and hardness. The effluent matrix, in turn, is influenced by the quality of the water used by the mill and mill operating conditions.

The effluent pH is a critical factor for the toxicity of acidic and basic compounds. For these, toxicity depends on the degree of ionization, with the unionized molecules being more toxic than the ionized forms. Because pH affects the degree of ionization, it also affects toxicity. Typically, the pH of most effluents increases during toxicity tests. This occurs mainly because of the loss of carbon dioxide from solution [17]. The change in pH tends to be insignificant for effluent from mills that use soft waters with low alkalinity (carbonate/bicarbonate content). When a pH increase does occur, it tends to be faster in trout tests than in *Daphnia* tests because the former is aerated and the latter is not. Aeration increases the loss of carbon dioxide from solution.

Hardness refers to the concentration of multivalent cations in water, mainly calcium and magnesium [18]. These cations have important physiological functions at the surface of gills in fish. Trace metals, such as copper, compete with calcium at the gill level [19]. As such, in waters with low hardness, even minute amounts of copper can affect the physiological functioning of the gill and result in toxicity [19].

**Toxicity test observations (Table 3)**

The crucial information from toxicity test reports includes time of mortality, species sensitivity (that is, was the effluent more toxic to trout or *Daphnia* or equally toxic to both species?), symptoms exhibited by the exposed organisms, and certain effluent characteristics such as pH and hardness. The symptoms of fish in Table 3, including the timing of mortality, are based on our observations of tests with specific compounds in the concentration range most likely to be found in mill effluents. The symptoms may be different for these compounds at more extreme concentrations. The symptoms in Table 3 are what we consider to be the most distinguishing for the agents listed and do not include a full list of symptoms that can occur. Some symptoms for *D. magna* are also given in Table 3.

**Mill operating conditions (Table 3)**

The final criterion for formulating a hypothesis involves knowledge of normal operating conditions and an audit of changes that may have occurred prior to the toxicity episode. Success depends on the availability of accurate information and dedication of the mill staff. The basic information that is required includes the wood furnish, type and performance of the biotreatment system, kinds and amounts of additives used, and records of spills or accidental release of certain chemicals in substantial quantities. The effluent toxicity may be affected by the type of wood species used for pulping as well as the time between cutting and use by the mill. The extractives in wood can be toxic to aquatic life and the concentrations and type of extractives are wood species related [20,21]. The extractives content can be reduced with chip storage time [22]. The major types of biotreatment systems in use are aerated stabilization basins, activated sludge, oxygen activated sludge and sequential batch reactors. The various systems require different operating conditions, most importantly in terms of nutrient addition [23], and this can result in differences concerning toxicity (e.g., in terms of ammonia). The oxygen activated sludge treatment plants also pose unique problems with respect to effluent toxicity as these plants are covered and sealed [24]. This results in high carbon dioxide concentrations and relatively low effluent pH (6 to 6.5).

TABLE 2
Toxicological properties of common causative agents of effluent toxicity.

| Compound(s) | Approximate Toxicity Threshold | | | Key Modifying Factors |
|---|---|---|---|---|
| | Rainbow trout | Daphnia magna | Test Condition | |
| Resin acid [7-9] | >1 mg/L | >4 mg/L | pH <7.5 | pH [Increased pH decreases toxicity] |
| Ammonia [10,11] | 0.4 to 0.6 mg/L* | 0.8 to 1.2 mg/L* | — | pH [Increased pH increases toxicity] |
| Nitrite [12]** | >6 mg/L | >10 mg/L | Chloride: <20 mg/L | Chloride [Increased chloride decreases toxicity] |
| Carbon dioxide [13] | >125 mg/L | >180 mg/L | pH 6–6.5 | pH [Increased pH decreases toxicity] |
| Dimethyl disulfide | >10 mg/L | >50 mg/L | — | — |
| Polymeric Formulations [14,15] | Variable: 1 to 50 mg/L | Variable: 0.04 to >100 mg/L | — | — |
| Metals [16]: | | | | |
| Copper | >10 µg/L | Variable | Hardness: <20 mg/L as $CaCO_3$ | Hardness [Increased hardness decreases toxicity] |
| Manganese | >2 mg/L | >18 mg/L | Hardness: <40 mg/L as $CaCO_3$ | Hardness [Increased hardness decreases toxicity] |

* Expressed in terms of un-ionized ammonia concentration
** Expressed as $NO_2$-N

TABLE 3
Criteria for selection of suspected causes of effluent toxicity.

| Causative Agent | Toxicity Test Information | | | | Mill Operating Conditions | | |
|---|---|---|---|---|---|---|---|
| | Species Sensitivity | Effluent pH | Rainbow Trout | | Pulping | Biotreatment | Additive |
| | | | Approx. Time of Mortality, h | Symptoms[1)] | | | |
| Resin Acids | Trout > Daphnia | <7.5 | <24 | • Gasping at surface[2)]<br>• Erratic darting interspersed with resting on the bottom<br>• Distended abdomen (swim bladder) | • Softwood in furnish<br>• Water with low alkalinity | • Upset or overload<br>• OAS | NA |
| Ammonia | Trout > Daphnia | 8.0–9.0 | 48–96 | • Prolonged lethargy, sluggishness<br>• Loss of equilibrium interspersed with recurring bouts of erratic swimming | NA | • Nutrient addition or endogenous decay of biomass | NA |
| Nitrite | Trout > Daphnia | NA | 48–96 | • Prolonged lethargy and sluggishness, but no loss of equilibrium | • Only mechanical mills | • Incomplete nitrification | NA |
| Carbon dioxide | Trout > Daphnia | 6.0–6.5 | <6 | • Instantaneous violent swimming, attempts to jump out, gasping at surface interspersed with resting on the bottom<br>• Greatly increased gill ventilation<br>• Fish not killed in the first few hours recover | — | • OAS | NA |
| Dimethyl disulfide | Trout > Daphnia | NA | <24 | • Lethargy and sluggishness<br>• Surfacing | • Only kraft mills | • OAS | NA |
| Polymeric[3)] formulations | Daphnia > Trout[4)]<br>Trout > Daphnia<br>Trout = Daphnia | NA | Variable | • Variable and intermittent<br>• Surfacing, loss of equilibrium and lethargy is common | • Mostly at mills making paper | • Mostly activated sludge | Yes |
| Trace metals (Cu and Mn) | Trout > Daphnia[5)] | 6.0–7.5 | >24 h | • Mostly fish appear normal<br>• Recurring bouts of brief equilibrium loss and occasional darting resulting in light reflection off sides of the fish | • Soft process water (<50 mg/L as $CaCO_3$) | NA | Yes |

NA: not applicable; OAS: oxygen activated sludge

[1)] This table lists some distinguishing symptoms at concentrations likely to be present in mill effluents.
[2)] Based on observations of tests with dehydroabietic acid.
[3)] Distinguishing symptoms for Daphnia include: at surface, often stuck together; debris attachment; outer shell sometimes distended; non-concentration dependent mortality
[4)] For about 80% of the formulations
[5)] In mill effluents

**Diagnostic Tests (Table 4)**

Once the above criteria have implicated suspected causative agent(s), simple diagnostic tests are available for confirmation purposes. These could include only analysis for a suspected toxicant or specific tests summarized in Table 4. The diagnostic tests essentially involve some kind of effluent manipulation that is meant to selectively reduce/remove particular effluent component(s) or to alter their chemical structure, such as the degree of ionization. The results of toxicity tests done with effluent samples before and after these manipulations provide the evidence for confirmation or repudiation of the suspected causative agent. As the diagnostic tests are mainly taken from the U.S. EPA protocols [3-5], these can be consulted by those interested in specifics concerning methods of filtration, pH adjustment and so on.

Because the diagnostic procedure is hypothesis driven and is meant to be used as a quick tool for assessing only for common causes of regulatory toxicity, there is less of a requirement for full confirmation tests as recommended by the U.S. EPA protocols [5]. Of course, confirmation steps, such as spiking a non-toxic effluent with a suspected toxicant, may be done if there is a need and resources are available.

Trout tests require substantial volumes of effluents, often exceeding 10 liters. Such volumes may be problematic for the effluent manipulation steps (e.g., filtration, cation exchange). There may be a temptation to use a surrogate species requiring smaller volumes for the diagnostic tests. However, in our experience this is not advisable even when the surrogate species is another species of fish, such as fathead minnow larvae. It is much better to modify the trout test in terms of volume and number of fish used. We have had good success with using three fish in a two-liter volume [6] instead of using ten fish in volumes  10 L as per the regulatory protocol [25]. In virtually all cases, the results of these small-volume tests were very similar to the results of trout tests done according to regulatory protocols. Nevertheless, before the start of any investigation, it may be worthwhile to compare the results of small-volume and regulatory tests.

## Summary

- A simple diagnostic strategy, intended for use by mill staff, was developed for troubleshooting regulatory effluent toxicity episodes involving the most common toxicants.
- The strategy involves formulating a hypothesis as to the cause of toxicity followed by chemical analysis and specific diagnostic tests, taken from generic U.S. EPA TIE protocols, which include effluent manipulation. The selection of the suspected cause of toxicity is based on the toxicological properties of causative agents, understanding how mill operating conditions may influence these properties, criteria from the toxicity test results and a review of the mill operating conditions.
- The strategy is expected to correctly diagnose about 70% of the effluent toxicity episodes. For the remaining 30% of the cases, more elaborate toxicity identification evaluation procedures will be needed.

TABLE 4
Diagnostic tests for suspected causes of effluent toxicity.

| Suspected Cause | Chemical Analysis | Diagnostic Tests | Expected Outcome |
|---|---|---|---|
| Resin acids | 1 mg/L | - pH adjustment | - Reduction in toxicity if pH is increased |
| Ammonia | Un-ionized ammonia 0.4 to 0.6 mg/L | - pH adjustment<br>- Cation exchange | - Reduced toxicity with reduced pH<br>- Reduced toxicity after cation exchange |
| Nitrite | 6 mg/L | - Chloride addition | - Decreased toxicity with increased chloride levels |
| Carbon dioxide | 125 mg/L | - pH adjustment<br>- Air sparging | - Decreased toxicity with increased pH<br>- Decreased toxicity after sparging |
| Dimethyl disulfide | 10 mg/L | - Air sparging | - Decreased toxicity after sparging |
| Polymeric formulations | — | - Filtration<br>- Solvent extraction<br>- Sublation* | - Decreased toxicity to Daphnia<br>- Toxicity captured in solvent phase<br>- Toxicity captured in residue phase |
| Metals (copper, manganese) | Copper: >10 µg/L; Manganese: >2.5 mg/L | - EDTA addition<br>- Hardness adjustment | - Decreased toxicity<br>- Decreased toxicity with increased hardness |

* Sublation: removal of surface-active material by sparging with air in a glass cylinder [3].

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