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Ecotoxicity risk assessment of amines used in 'switchable water' and CO_2-capturing processes†

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Utilizing amines and alkanolamines as CO_2-capturing agents and water-soluble ionogens in 'switchable water' systems is an intensively explored research area. However, the potential risks of such amine derivatives to the environment have been poorly evaluated. In this work, we report on the ecotoxicological effect of relevant amines and alkanolamines in an aqueous environment on various classes of organisms such as bacteria (*Aliivibrio fischeri*), vascular plants (*Spirodela polyrhiza*), and invertebrates (*Daphnia magna*). The measured half maximal effective concentration (EC_{50}) data indicate that all tested alkanolamines and most amines have EC_{50} values over 100 mg L^{-1} and can be classified as practically harmless or harmless. On the other hand, tetramethyl-1,3-propane diamine afforded EC_{50} values between 61 and 73 mg L^{-1}, indicating moderate toxicity towards invertebrates and vascular plants. Moreover, we observed a good agreement between the experimental results and the ECOSAR predictive model. Thus, our work indicates that hydrophilic amines and alkanolamines utilized in emerging CO_2-mediated processes can generally be considered harmless or practically harmless in an aqueous environment towards bacteria, vascular plants, and invertebrates, except more lipophilic diamines, which may need careful consideration.

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Environmental significance

Many CO_2-capturing and 'switchable water' processes rely on amines' ability to absorb CO_2 gas. These processes are an essential part of the more sustainable chemical and energy sector, but the involvement of large amounts of amines may pose risks to aquatic and terrestrial ecosystems. The work provides data on the ecotoxic effects on various classes of organisms, such as bacteria (*Aliivibrio fischeri*), vascular plants (*Spirodela polyrhiza*), and invertebrates (*Daphnia magna*). Our risk assessment enables the design of safer chemicals and processes, minimizes long-term environmental harm, and supports regulatory compliance, ensuring that amine-mediated emerging technologies contribute to both emission control and ecosystem protection.

Introduction

The need for green transition has sparked wide interest in utilizing amines as CO_2-capturing (CC) agents and, more recently, also as water-soluble ionogens in switchable water (SW) systems. The aim of these technologies is to capture gaseous CO_2 and, in the case of the SW process, to reduce the energy requirements associated with the separation of organic products from water. Therefore, they are crucial in moving towards more sustainable chemical processes.

In both processes, a key intermediate is a soluble bicarbonate salt, which forms due to an acid–base reaction between hydrated CO_2 and an amine (Fig. 1).[1]

In the case of amine-mediated CO_2 capturing technology (Fig. 1a), gaseous CO_2 is first absorbed into an aqueous amine solution, where CO_2 reacts with the amines to form carbamates or bicarbonates, depending on the type of amine (primary, secondary, or tertiary). The CO_2 can be released upon heating, and the water and amines can be subsequently recovered and reused in the process.[2–5] It is a well-established process that offers operational convenience, rapid and high-capacity CO_2 absorption, and recyclability.

On the other hand, the SW process facilitates the removal of water-soluble organic compounds (*e.g.*, EtOH, *etc.*) from the water. This is particularly relevant in emerging chemical production processes from biomass, where the conventional separation techniques, such as distillation, have high energy requirements and can thus negate the environmental benefits of biomass conversion.[6] In this process, an amine is added to the mixture of water and an organic product.[7–13] Upon subsequent introduction of CO_2 gas into the system, the amine reacts to form its bicarbonate salt (Fig. 1b), inducing the precipitation of the organic product if it is a solid or facilitating its separation in the form of an "organic-rich" liquid phase if it is a liquid. After separating the organic component, the aqueous phase can be decarbonated, and the removed CO_2 can be re-used. Decarbonation reverses the reaction, converting the amine back to its

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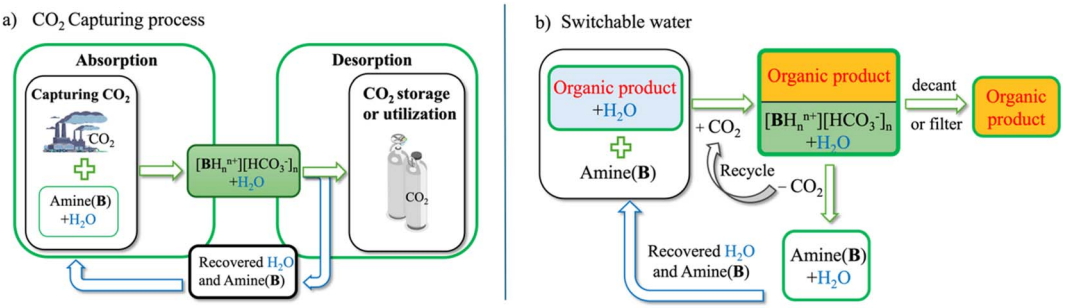


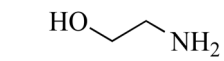
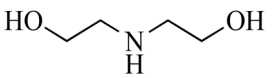
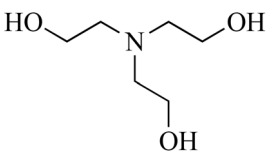
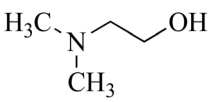
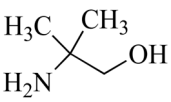
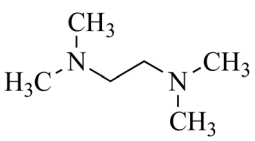
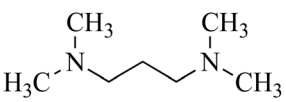
Fig. 1 Schematic illustration of using amines in the (a) CO_2 capture and (b) switchable water (SW) processes.

neutral state. Finally, the amine can be recovered from the decarbonated water *via* reverse osmosis or by filtration and reused for another cycle.

The development of any new technology requires a critical assessment of its potential environmental impact. Hence, the environmental assessment of amines, particularly those used in emerging technologies, is imperative due to their potential environmental exposure and biological effects. Although the amines used in these processes are recycled, the unintended leaks into the environment cannot be excluded. Moreover, such an assessment during the early stages of development identifies the hotspots directly and guides the process development towards lower risks.

Studies on the ecotoxicity of amines have primarily focused only on a few alkanolamines, such as monoethanolamine (**MEA**), diethanolamine (**DEA**), and triethanolamine (**TEA**; for structures, see Table 1).[14–19] Only limited and divergent information is available, mainly for some decomposer, producer, and first-level consumer organisms. These amines have been evaluated previously toward single-celled organisms (*Entosiphon sulcatum* and *Chilomonas paramecium*), bacterium (*Vibrio fischeri*), invertebrate (*Daphnia magna*) and alga (*Skeletonema*

Table 1 List of compounds evaluated in this study

Entry	Name/amine type	Formula	CAS no.	MW (g mol^{-1})	log K_{ow}[a]	Structure
1	Monoethanolamine (**MEA**)/primary	C_2H_7NO	141-43-5	61.08	−1.6	
2	Diethanolamine (**DEA**)/secondary	$C_4H_{11}NO_2$	111-42-2	105.14	−1.7	
3	Triethanolamine (**TEA**)/tertiary	$C_6H_{15}NO_3$	102-71-6	149.19	−2.5	
4	Dimethylethanolamine (**DMEA**)/tertiary	$C_4H_{11}NO$	108-01-0	89.14	−0.9	
5	Amino-2-methyl-1-propanol (**AMP**)/hindered	$C_4H_{11}NO$	124-68-5	89.14	−0.7	
6	Tetramethylethylenediamine (**TMEDA**)/diamine	$C_6H_{16}N_2$	110-18-9	116.24	−0.3	
7	Tetramethyl-1,3-propanediamine (**TMPDA**)/diamine	$C_7H_{18}N_2$	110-95-2	130.23	0.2	

[a] Data taken from QSAR prediction analysis (ECOSAR).

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costatum).[17,19,20] Generally, the findings from these studies suggest a low toxicity level of alkanolamines towards most tested species. On the other hand, amines developed more recently, especially for the SW process, have not been evaluated. Moreover, the potentially harmful effects of amine emissions, such as the formation of nitrosamines and nitramines *via* photooxidation in the atmosphere, which could harm human health and the environment, have been observed.[21] Hence, there is a gap in the knowledge about the toxicity data of amines employed more recently in SW or CC processes. It is also generally known that even minor modifications in chemical structure can lead to substantial changes in biological activity and environmental fate.[22] The lack of comprehensive data on the environmental toxicity of many of these compounds motivates us to carry out wider ecotoxicological testing.

In this work, we report on the ecotoxicity of structurally diverse amines utilized in emerging SW and CO_2 capture technologies. The list includes primary monoethanolamine (**MEA**), secondary diethanolamine (**DEA**), tertiary triethanol – (**TEA**) and dimethylethanolamine (**DMEA**), structurally hindered amino-2-methyl-1-propanol (**AMP**), and two diamines – tetramethylethylenediamine (**TMEDA**) and tetramethyl-1,3-propanediamine (**TMPDA**). Such amines are often used for SW and CC processes due to their ability to absorb gases efficiently.[23] **MEA**, **DEA**, **TEA**, **AMP**, and **DMEA** are alkanolamines used in both processes.

The ecotoxicity was assessed on aquatic organisms with varying biological complexity, *i.e.*, bacteria, vascular plants, and invertebrates.

Experimental

Test compounds

The tested compounds are listed in Table 1: monoethanolamine (**MEA**) (Alfa Aesar, purity 98%), diethanolamine (**DEA**) (Lach-Ner, purity 99%), triethanolamine (**TEA**) (Alfa Aesar, purity 98%), dimethylethanolamine (**DMEA**, reagent grade), 2-amino-2-methyl-1-propanol (**AMP**, reagent grade), tetramethylethylenediamine (**TMEDA**) (Sigma Aldrich, purity 99%), tetramethyl-1,3-propanediamine (**TMPDA**) (Sigma Aldrich, purity 99%). All tested compounds were liquids and miscible with water.

Ecotoxicity testing

The toxicity assessment was performed using three standardized tests. The level of toxicity of compounds was determined by establishing the half-maximal effective concentration, EC_{50}, the concentration of substances in the environment that will affect 50% of the organisms in the test population under specified conditions. For bacterial tests, the range of concentration of each sample was 1–5000 mg L^{-1}, and for vascular plants and invertebrates, the range was 1–1000 mg L^{-1}. A fresh stock solution with the highest tested concentration was prepared for each test, thus ensuring concentration accuracy throughout the studies. Serial dilutions of the stock solution were then performed to obtain the lower-concentration solutions. All samples were tested in triplicate for each assay to ensure test reproducibility. The stability of **TMPDA** and **TMEDA** was checked by incubating the compounds under tested conditions without organisms and the results were confirmed by ^{1}H and ^{13}C NMR (Fig. S1–S12†).

WaterTOX™ STD: bacterial luminescence inhibition test using *Aliivibrio fischeri*. Toxicity of samples towards the bioluminescent marine bacterium *A. fischeri* was measured by comparing initial and final light emission after 15 min according to the ISO standard 11348-3:2007.[24] The toxic effect caused by a decrease in cellular metabolism is expressed as a decrease of the luminescence intensity. Tests were carried out at 15 °C in the kit's standard diluent. Potassium dichromate [12.5–100 mg L^{-1}] was used as a reference positive control. A series of dilutions were prepared for each sample according to the manufacturer's instructions.

Growth inhibition test with vascular plants. *Spirodela polyrhiza* is based on the measurement of growth retardation of the germinated dormant vegetative buds (turions) after 3 days of exposure to samples according to the ISP standard.[25] The tests were carried out on a 48-well plate containing a dilution series of tested samples at 25 °C in the plant growth chamber, using an illumination system enabling at least 6000 lux. A digital image of the multiwell plate was taken at the start of the test and after the incubation to measure the growth inhibition by Image Analysis (Image J, National Institute of Mental Health, Bethesda, Maryland, USA, software for image processing and analysis) to determine the size of the vegetative buds (turions) before and after incubation. Next, by comparing the data obtained from the test plate, the growth of the duckweeds was calculated by subtracting the mean of the "initial" size of the first frond from the mean "final" size, in the control and at various concentrations of diluted samples. The 72 h EC_{50} concentration of the compound was obtained from the percentage of growth inhibition of the duckweed.

Crustacean toxicity screening test for freshwater using *Daphnia magna*. This test determines the lethal effects of toxicants on the *D. magna* after 48 h exposure. The 48 h immobilization test was performed in a multi-well test plate using neonate *D. magna* hatched from ephippia based on the ISO standard.[26] Ephippia hatching was initiated before the start of the toxicity test in a Petri dish with standard freshwater medium at 25 °C for 72 h, under continuous illumination (at least 6000 lux). Two hours before collecting the neonates for the test, they were pre-fed with *Spirulina powder*. The test incubation was carried out on a multiwell plate containing a dilution series of the tested samples in darkness for 48 hours. The number of immobilized (dead) organisms was counted after 48 h under a microscope (magnification 10–12×). The obtained data was used to determine the EC_{50} values.

Prediction of EC_{50} for amines. Modelling was done using the Ecological Structure Activity Relationships (ECOSAR) predictive model (Tracy Wright, U.S. EPA Existing Chemicals Risk Assessment Division), software for estimating a chemical's acute (short-term) toxicity and chronic (long-term or delayed) toxicity to aquatic organisms.

Statistical analysis. A comparison of the tested substrates was analysed using one-way ANOVA, and a *post hoc* pairwise

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comparison *via* a Tukey test in R (version 3.6.0) using aov( ) and TukeyHSD( ) on EC_{50} data (Table S2†). The linear trends of $\log(1/EC_{50})$ against $\log K_{ow}$ for each tested organism were found based on the performed tests.

Results and discussion

The results of the toxicity measurements of the tested compounds are visualized in Fig. 2. The tertiary amine **TEA** showed the highest EC_{50} values (*i.e.*, the lowest toxicity) across all tested organisms and can be rated as non-toxic (EC_{50} > 1000 mg L^{-1}, Fig. 2a–c). Compared to **TEA**, the secondary **DEA** exhibited lower EC_{50} values, followed by the primary **MEA** but both can still be categorized as practically harmless towards bacteria [$EC_{50(\mathbf{DEA})}$ = 468 (95% CI: 280; 656) mg L^{-1}; $EC_{50(\mathbf{MEA})}$ = 227 (95% CI: 150; 304) mg L^{-1}], vascular plants [$EC_{50(\mathbf{DEA})}$ = 549 (95% CI: 479; 619) mg L^{-1}; $EC_{50(\mathbf{MEA})}$ = 358 (95% CI: 262; 454) mg L^{-1}] and invertebrates [$EC_{50(\mathbf{DEA})}$ = 367 (95% CI: 288; 447) mg L^{-1}; $EC_{50(\mathbf{MEA})}$ = 260 (95% CI: 203; 317) mg L^{-1}]. Thus, the toxicity of these aminoalcohols increases in the following order: **TEA** < **DEA** < **MEA**. These results align with the previous ecotoxicity studies of **MEA**, **DEA**, and **TEA** in seawater environments, where the toxicity also decreased when alkyl substituents were added to the nitrogen atom.[20,27] Similarly, Finlay and Callow have investigated the impact of alkyl chain length and branching on aquatic organisms.[14] They found that alkylamines with shorter and less-branched alkyl chains exhibited higher toxicity than those with longer and more highly branched chains.

Compared to **TEA**, another tertiary alkanolamine, **DMEA**, which has two ethanol groups replaced by methyl substituents, exhibited somewhat lower EC_{50} values towards all three groups of organisms. **DMEA** showed a practically harmless level of toxicity towards *S. polyrhiza* [EC_{50} = 229 (95% CI: 188; 269) mg L^{-1}], *D. magna* [EC_{50} = 223 (95% CI: 213; 234) mg L^{-1}] and *A. fischeri* [EC_{50} = 437 (95% CI: 293; 581) mg L^{-1}].

Primary aminoalcohol **AMP** is a structural isomer of **DMEA** with different arrangements of substituents. The toxicity levels of these two isomers were also relatively similar, although the EC_{50} values for **AMP** were slightly higher towards the tested

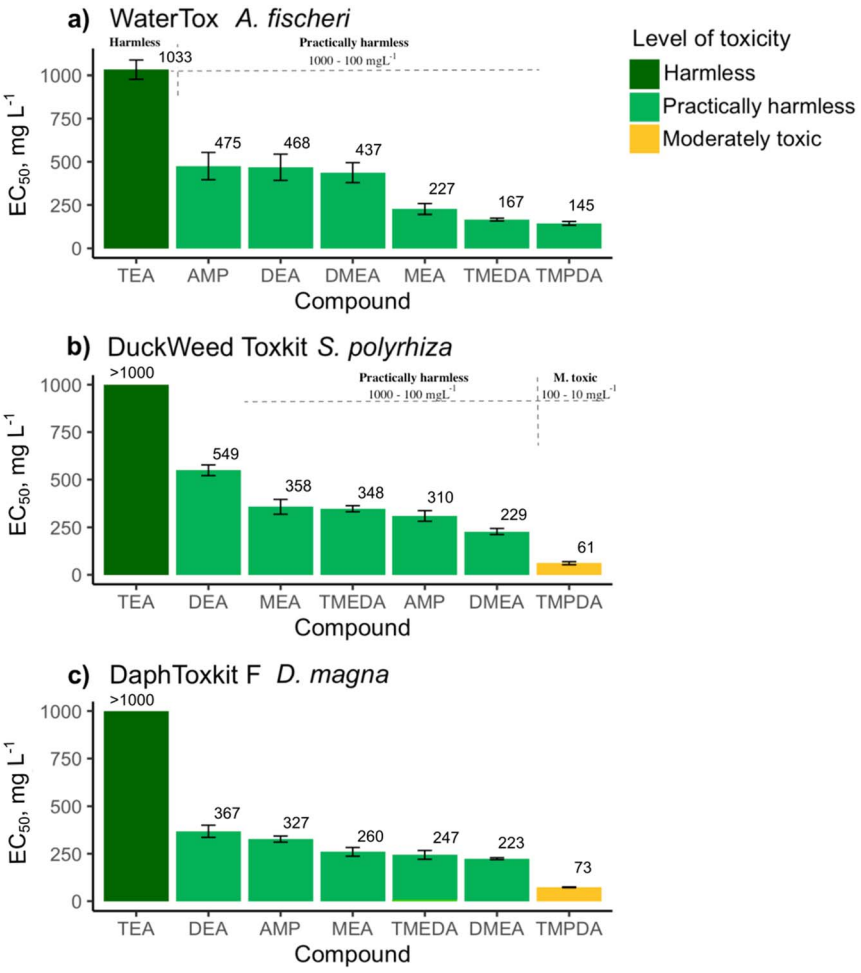

Fig. 2 Values of the tested compounds' mean effective concentrations (EC_{50}, mg L^{-1}) towards (a) *A. fischeri*, (b) *S. polyrhiza*, (c) *D. magna*. The compounds are ordered according to the level of toxicity: dark green represents the harmless (EC_{50} values > 1000 mg L^{-1}) compounds; light green represents the practically harmless (EC_{50} values 100–1000 mg L^{-1}) compounds; yellow denotes the moderately toxic (EC_{50} values of 10–100 mg L^{-1}) compounds. For numerical values, see (Table S1†).

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organisms, *i.e.*, EC_{50} = 475 (95% CI: 279; 671) mg L^{-1} towards *A. fischeri*, EC_{50} = 310 (95% CI: 240; 380) mg L^{-1} towards *S. polyrhiza* and EC_{50} = 327 (95% CI: 287; 367) mg L^{-1} towards *D. magna*.

Finally, we evaluated two very similar structures, **TMEDA** and **TMPDA**, which differ only by the length of the carbon spacer between the two nitrogen atoms. While diamine **TMEDA** with an ethylene spacer showed a practically harmless effect toward *A. fischeri* [EC_{50} = 167 (95% CI: 149; 186) mg L^{-1}], *S. polyrhiza* [EC_{50} = 348 (95% CI: 308; 388) mg L^{-1}] and *D. magna* [EC_{50} = 247 (95% CI: 192; 302) mg L^{-1}], in contrast, **TMPDA** with a propylene spacer was moderately toxic to *S. polyrhiza* [EC_{50} = 61 (95% CI: 41; 81) mg L^{-1}] and to *D. magna* [EC_{50} = 73 (95% CI: 69; 77) mg L^{-1}]. Moreover, **TMPDA** was the only tested compound with EC_{50} values in the moderate toxicity range, although towards *A. fischeri* it can still be categorized as practically harmless [EC_{50} = 145 (95% CI: 117; 172) mg L^{-1}].

We correlated our results with the octanol–water partition coefficient ($\log K_{ow}$; for calculated values, see Table 1). This coefficient measures a compound's hydrophobicity and shows the distribution between a hydrophobic (octanol) and a hydrophilic (water) phase.[28] This parameter is often used to estimate the potential environmental risk of compounds to the aquatic environment. Chemicals with high $\log K_{ow}$ values are less soluble in water, which increases bioaccumulation and potential toxicity levels.[29] In our study, tertiary alkanolamine **TEA** is the only compound that showed a harmless level of toxicity towards all tested organisms. The $\log K_{ow}$ for **TEA** is −2.5, which is the lowest value compared to other alkanolamines that have $\log K_{ow}$ values in the range from −0.7 to −1.7 and a practically harmless level of toxicity. The diamine **TMPDA**, which has the highest $\log K_{ow}$ value among all tested compounds ($\log K_{ow}$ = 0.2), showed moderate toxicity toward *S. polyrhiza* and *D. magna*. Such correlation, where more hydrophilic compounds (**TEA**, **DEA**, **MEA**, **DMEA**, **AMP**, **TMEDA**) exhibit lower toxicity compared to the more hydrophobic compounds (**TMPDA**), also aligns with our previous ecotoxicology study with isosorbide-based compounds.[30]

There could be several potential mechanisms responsible for amine toxicity. One of the reported mechanisms is the induction of oxidative stress by the generation of reactive oxygen species, which leads to lipid peroxidation and cytotoxicity.[31,32] Additionally, reactive amine-derived species, such as nitrosamines or imines, can cause DNA damage.[33]

We also evaluated the aquatic toxicity of the same test compounds towards *Daphnia magna* and green algae using the Ecological Structure Activity Relationships (ECOSAR) predictive model (Table 2). Compared to our experimental results, the ECOSAR predicts similar trends, and in many cases, even the individual EC_{50} values are comparable. For example, **TEA** and **TMPDA** are the least and most toxic compounds, respectively, by both methods, and their EC50 values fall into the same range. Additionally, mono-, di-, and triethanolamines exhibit the same order of toxicity and similar EC_{50} values. Hence, for such types of amines and alkanolamines, the ECOSAR can be considered a valuable tool for initial screening.

Table 2 ECOSAR prediction results for amines

	D. magna 48 h	Green algae 48 h
Short name	EC_{50} mg L^{-1} (ECOSAR)	EC_{50} mg L^{-1} (ECOSAR)
TEA	1771	4092
DEA	430	834
MEA	217	411
DMEA	123	199
AMP	94	145
TMEDA	63	87
TMPDA	35	44

Additionally, we carried out liner regression analysis between $\log K_{ow}$ and toxicity for amines to each tested organism $\log(1/EC_{50})$ (Fig. S13†). The resultant regressions are: $\log(1/EC_{50}) = 0.32 \log K_{ow} - 2.1$ ($R^2 = 0.76$, p-value = 3.03×10^{-7}) for *D. magna*; $\log(1/EC_{50}) = 0.35 \log K_{ow} - 2.1$ ($R^2 = 0.74$, p-value = 5.33×10^{-7}) for *S. polyrhiza*; and $\log(1/EC_{50}) = 0.33 \log K_{ow} - 2.6$ ($R^2 = 0.62$, p-value = 2.01×10^{-5}) for *V. fischeri*. After analyzing the obtained equations, we conclude that there is no statistical difference between the tested organisms (Fig. S14†). Compared to the reported linear regression between $\log K_{ow}$ and $\log(1/EC_{50})$ for non-amine compounds,[34] our results indicate that the amine compounds showed a similar or slightly lower level of ecotoxicity (Fig. S15†).

Conclusion

The toxicity measurements of various amines reveal a clear trend related to their structural and hydrophilicity properties. Tertiary amine **TEA** exhibited the highest EC_{50} values, indicating the lowest toxicity among the tested compounds, and is categorized as harmless. Secondary amine **DEA** and primary amine **MEA**, while slightly more toxic, are still considered practically harmless to the tested organisms. Tertiary **DMEA**, despite being somewhat more toxic than **TEA**, also falls into the almost harmless category. The structurally hindered **AMP** showed similar toxicity levels to **DMEA**.

The ecotoxicity results of diamines **TMEDA** and **TMPDA** demonstrated that even small structural changes, such as the length of the carbon spacer between nitrogen atoms, can impact toxicity. **TMEDA** remained practically harmless, while **TMPDA** showed moderate toxicity towards *S. polyrhiza* and *D. magna* organisms, likely due to its lower hydrophilicity. Overall, the results suggest that increased hydrophilicity in these amines correlates with reduced toxicological impact, probably due to enhanced excretion and decreased bioaccumulation.

Data availability

Data supporting this article have been included as a part of the ESI.†

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Author contributions

Alina Ismagilova: investigation, data curation, formal analysis, validation, visualization, writing – original draft. Veljo Kisand: supervision, methodology, writing – review & editing. Lauri Vares: conceptualization, funding acquisition, supervision, methodology, writing – review & editing.

Conflicts of interest

The authors declare no conflict of interest.

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