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Original Article

Toxic Effects Promoted by a Commercial Detergent on the Germination and Initial Development of Cucumber Seedlings (*Cucumis sativus* L.)

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Abstract

Detergent is a significant pollutant that poses serious risks to natural ecosystems. Its components can penetrate soil and bodies of water from various sources, negatively impacting fauna and flora and causing eutrophication, foam formation, as well as altering important parameters such as temperature, salinity, turbidity, and pH. Considering these factors, the aim of this study was to evaluate the toxic potential of a commercial detergent using cucumber (*Cucumis sativus* L.) as a model organism. Four concentrations (1, 2, 4, and 8 g L^{-1}) were evaluated, and the toxicity test was carried out according to ISO 18763 standards with minor modifications. The parameters evaluated were: percentage of germination, germination speed index, root length, and shoot length of seedlings. The test results showed that the detergent had no toxic effects on seed germination percentage. However, all concentrations affected the germination speed index. Regarding root length, this parameter was affected since the concentration 2 g L^{-1}, with this concentration promoting an 11.26% reduction compared to the control. Shoot length was affected by all concentrations, being the most affected parameter by the different treatment, as revealed by the calculation of the toxicity index. The mean inhibitory concentration (IC_{50}) that reduced root length by 50% was 3.03 g L^{-1}, while for shoot length, was 1.56 g L^{-1}. These results suggest that the excessive use or improper disposal of commercial detergents can have negative effects on living organisms, highlighting the importance of control and regulation measures to mitigate the environmental risks associated with their use.

Keywords: Plant bioassays. Water contamination. Ecotoxicology. Surfactants.

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INTRODUCTION

Detergents play a fundamental role in various industrial processes, being considered the most important cleaning product worldwide, with an annual investment of approximately US $ 60 billion (Bianchetti et al., 2015). They are widely used as fuel additives (Grinstead, 1994), in the synthesis of biological molecules (Dauty et al., 2001), and in personal, household, and laundry cleaning industries (Corey, 1994).

However, several studies have pointed out the adverse effects of detergents on the environment due to their ability to accumulate in organisms (McWilliams; Payne, 2002), low biodegradability (Verdia et al., 2016), and higher concentration of solids compared to other cleaning compounds (Anastas; Lankey, 2000). In light of this, there is a growing demand for the development of more sustainable cleaning products, such as "biodegradable" ones that use improved chemical compositions. However, even with these efforts, low has been done in terms of reducing toxicity compared to conventional washing compounds, and the negative environmental impacts of detergents are still significant (Arthur et al., 2012).

Over time, the presence of cleaning products in the environment has been a major concern, as their impact on the ecosystem is clearly visible. Among the various effects of detergents on natural ecosystems, eutrophication can be highlighted, which is the excessive growth of algae, and the decrease in oxygen and light in the water, affecting its quality. In addition, detergents have the potential to modify pH and salinity in the receiving bodies, potentially impacting aquatic environments' fauna and flora (Abd El-Gawad, 2014; Rajan, 2015).

To assess the toxic potential and possible environmental impact of detergents, a viable alternative is the use of bioassays that employ plant organisms as indicators. Plant bioassays have been widely used in ecotoxicology studies because they are sensitive and present rapid responses to environmental changes. Moreover, they are relatively simple and inexpensive, allowing for large-scale experiments (Da Silva, 2023; Iqbal et al., 2019). In this sense, the use of plant bioassays can be a valuable tool for evaluating the toxicity of detergents and contributing to the development of more sustainable and less harmful cleaning products to the environment.

One of the plants widely used in bioassays to assess the toxicity of chemical substances like detergents is the cucumber (*Cucumis sativus* L.). Cucumber is a plant commonly grown worldwide and is known for its sensitivity to various chemical compounds. The use of cucumber as an indicator organism in bioassays offers several advantages, including ease of cultivation, production of a large number of seeds, and availability at different times of the year (Guevara et al., 2019).

Considering the problem exposed, the main objective of this work is to evaluate the toxic potential of a commercial detergent using cucumber (*Cucumis sativus*) seeds as biological indicators, with germination and initial development of seedlings being employed as toxicological parameters.

MATERIALS AND METHODS

Preparation of solutions

The liquid detergent used in this study was purchased from a local market in the city of Lavras, located in Minas Gerais. Its chemical composition included chelating agents, preservatives, surfactants, dyes, adjuvants, fragrances, linear alkylbenzene sulfonate, and water, according to the specifications of the Safety Information Sheet for Chemical Products (FISPQ, 2018). To prepare the solutions, the detergent was diluted in distilled water, evaluating four different concentrations, corresponding to 1, 2, 4, and 8 g L^{-1}. In addition, distilled water was used as a control.

Toxicity test using cucumber seeds (*Cucumis sativus*)

The toxicity test, as well as the parameters evaluated in this study, were outlined in Figure 1 and followed the protocols established by the ISO 18763:2016 standard, with some specific modifications. The experiment was conducted using a completely randomized design, in which five different treatments (control, 1, 2, 4, and 8 g L^{-1}) were tested, with five replicates for each of them.

The *Cucumis sativus* seeds used in the experiment (Top Seed) were purchased from a local agricultural store. For the test, 20 seeds per replicate were placed in Petri dishes containing filter paper moistened with 3 mL of the test solution. The plates were sealed with plastic wrap and kept in a germination chamber at a temperature of 24 ± 2 ºC. Germination was evaluated at intervals of 12, 24, 36, 48, 60, and 72 hours to determine the percentage of germination (PG%) and the germination speed index (GSI). After 72 hours of exposure to the treatments, the length of the root and shoot of each seedling was measured using a digital caliper.

For assessing detergent toxicity in relation to the analyzed parameters (germination percentage, germination speed index, root and shoot length), the toxicity index was employed. This index was calculated through the relative mean, derived by dividing the mean values of the test treatment by the mean values of the control, then multiplying the outcome by 100. Furthermore, the average inhibitory concentration (IC_{50}) was determined using GraphPad Prism software.

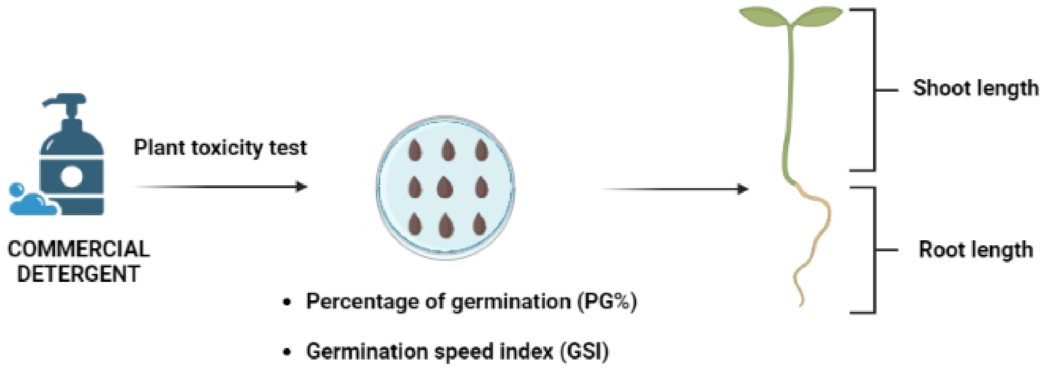

Figure 1 - Toxicity test and parameters employed to evaluate the toxic potential of a commercial detergent in the plant model *Cucumis sativus*. Source: Authors, 2023.

Data analysis

The data obtained the evaluated parameters, such as germination percentage (PG%), germination speed index (GSI), root and shoot length, were subjected to analysis of variance and the means were compared by Scott-Knott test, with 5% probability, using the SISVAR software. For graph elaboration, GraphPad Prism software was used.

RESULTS

Effects on germination

The commercial detergent, at the evaluated concentrations, did not affect the germination percentage (PG%) of cucumber (*Cucumis sativus*) seeds, as shown in Table 1. However, all tested concentrations had a significant effect on the germination speed index (GSI). The lowest evaluated concentration (1 g L^{-1}) resulted in a 10% reduction of this parameter, whereas the highest concentration (8 g L^{-1}) caused a significant decrease of 20.38% in the same parameter compared to the control group.

Table 1 - Percentage of germination (PG%) and germination speed index (GSI) of cucumber seeds (*Cucumis sativus*) exposed to different treatments. Means followed by the same letters in the column do not differ statistically among themselves (Sott-Knott Test 5%).

Treatments	PG%	GSI
Control	100 a	15,40 a
1 g L^{-1}	100 a	13,86 b
2 g L^{-1}	100 a	12,60 c
4 g L^{-1}	100 a	12,40 c
8 g L^{-1}	100 a	12,26 c

Source: Authors, 2023.

Effects on the initial development of cucumber seedlings (*Cucumis sativus*)

The detergent affected the development of cucumber seedling roots starting from a concentration of 2 g L^{-1}, causing an 11.26% reduction in root length compared to the control group. The most significant results were observed at the concentration of 8 g L^{-1}, which reduced this parameter by 90.37% (Figure 2). Regarding the length of the aerial part, all concentrations of detergent significantly reduced this parameter. At the lowest concentration (1 g L^{-1}), there was a reduction of 33.54%, while at the highest concentration (8 g L^{-1}), the reduction was 81.83% (Figure 2).

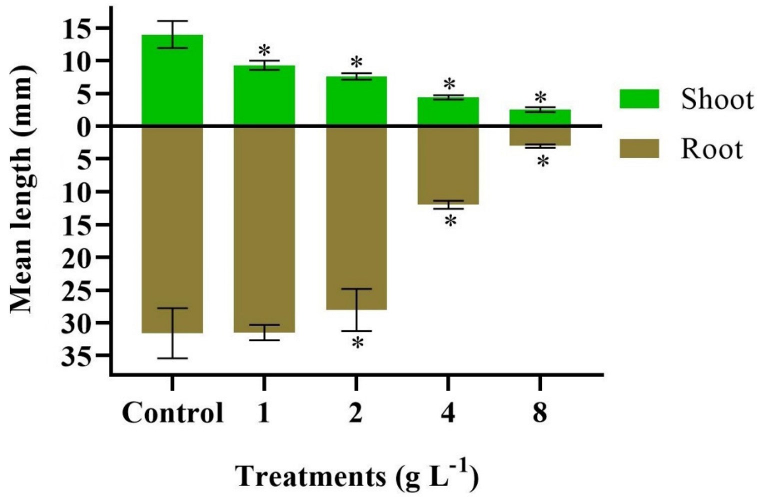

Figure 2 - Effects of different concentrations of the commercial detergent on the initial development of cucumber seedlings (*Cucumis sativus*). Data are expressed as mean + standard deviation.

* Differs statistically from the control group.

Source: Authors, 2023.

Ecotoxicity data of the commercial detergent

The toxicity index calculation revealed that different concentrations of the evaluated detergents did not show toxicity to the percentage of cucumber seed germination. However, the germination speed index was affected by 17.01%, and root length and shoot length were reduced by 41% and 57.38%, respectively (Figure 3).

In relation to the determination of the median inhibitory concentration (IC_{50}), it was not possible to calculate it for the parameters of germination percentage and germination speed index since none of the evaluated concentrations reduced these parameters by 50%. However, for root length, the IC_{50} value was determined to be 3.03 g L^{-1}. As for shoot length, the IC_{50} was identified as 1.56 g L^{-1} (Table 2).

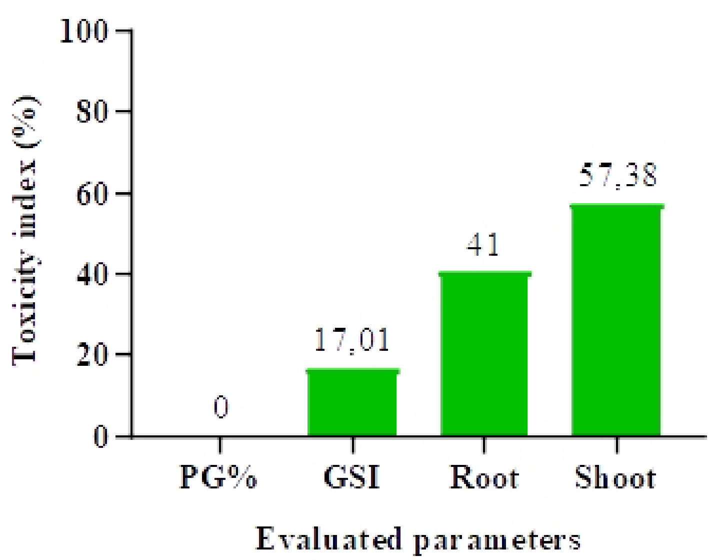

Figure 3 - Toxicity index of the commercial detergent on the evaluated parameters (percentage of germination (PG%), germination speed index (GSI), root and shoot length).

Source: Authors, 2023.

Table 2 - Mean inhibitory concentration (IC_{50}) obtained for each evaluated parameter using cucumber (*Cucumis sativus*) as a plant model.

Evaluated parameters	Mean inhibitory concentration (IC_{50})
Percentage of germination (PG%)	n.d
Germination speed index (GSI)	n.d
Root length	3,04 ± 0,2
Shoot length	1,56 ± 0,17

n.d = not determined

Source: Authors, 2023

DISCUSSION

The results from multiple studies employing plant models to assess the toxic potential of chemical agents have consistently shown that germination percentage is the least sensitive parameter to changes (Da Silva, 2023; Carvalho, 2019). This conclusion is supported by the calculation of the toxicity index (Figure 3). Despite its relative insensitivity, germination percentage has the capacity to comprehensively enrich the understanding of the effects triggered by such agents, especially when associated with other response indicators, such as germination speed index and seedling growth.

A previous study showed that concentrations higher than 4 g L^{-1} of detergent were able to reduce the germination percentage of lettuce seeds (*Lactuca sativa* L.), while concentrations of 2 g L^{-1} reduced the germination percentage of corn seeds (*Zea mays* L.) (Da Silva, 2023). However, in the present study, such reduction was not observed, since each plant species presents a different behavior regarding potentially toxic substances. In another study conducted by Uzma *et al*., 2018, the authors sought to evaluate the effect of different concentrations (1, 10, 50, 100, and 500 mg L^{-1}) of detergent on the germination of corn, and at these concentrations, there were also no significant effects on the germination percentage.

In the current study, the evaluated detergent did not exert an influence on germination percentage; however, it had a significant impact on the germination speed index. Various chemical substances, such as detergents, lack the ability to directly affect germination percentage. Nonetheless, they can induce disruptions in seed metabolism, leading to a delay in the germination process and consequently impeding proper seedling development (Rodrigues, 2009). This phenomenon, observed within the scope of this study, is supported by the data presented in Table 1 and illustrated in Figure 2.

The observed effect on the germination speed index might be closely linked to the detergents' ability to induce oxidative stress. This phenomenon can trigger lipid peroxidation, which in turn increases the permeability of the cell membrane as a response to exposure to toxic ions generated by the detergents (Heidari, 2013). Additionally, the interaction between detergents and seeds inhibits water absorption due to an increase in osmotic potential. This interaction can consequently lead to significant disturbances in the seed's inherent metabolic pathways (Ehilen et al., 2017).

The evaluated detergent also contained surfactants and tensioactives. These components are used to reduce the surface tension of water, which can interfere with the natural germination process and prevent seeds from breaking dormancy (Penteado et al., 2009). This condition, in turn, may have contributed to the observed germination delay in the present study.

In the present study, the toxicity of the commercial detergent was confirmed, as evidenced by the reduction in the root and shoot length of cucumber seedlings (Figure 2). The calculation of the toxicity index revealed that the shoot and root were most affected parameters by the different concentrations of the detergent (Figure 3). Studies show that root and shoot length are the most sensitive parameters to evaluate the toxic potential of substances, as complex physiological reactions occur during the development of these organs that can be inhibited by toxic substances (Lapa, 2014).

The reduction in cucumber seedling length can be attributed to the presence of salts and surfactants in the chemical composition of the evaluated commercial detergent. High salinity is known to inhibit water absorption by plants (Rahimi et al., 2006). Moreover, elements like sodium present in water contaminated by the detergent can inhibit specific enzymes, resulting in imbalanced nutrient absorption by the plants and consequently negatively impacting their physiological development (Ehilen et al., 2017).

Another component present in the detergent that might have contributed to the growth inhibition of seedlings is surfactants. These substances can disrupt the ionic balance of exposed seedlings, leading to increased concentrations of ions such as sodium (Na^+), potassium (K^+), and calcium (Ca_2^+), which directly affects water and nutrient uptake by the plants. Additionally, the reduction in seedling length is closely related to the detergents' ability to cause injuries to plant tissues. This property is associated with the interaction of surfactants with various proteins present in the cell membrane, affecting physiological and biochemical processes of the cell, as well as its interaction with lipids, resulting in modification of fatty acid compositions (Uzma et al., 2018).

It's important to highlight that detergents have the potential to negatively affect cell viability, as demonstrated in the literature by cytotoxic effects on meristematic cells of *Allium cepa* L. (Iqbal et al., 2017). Considering that plant growth relies on cell division occurring in actively mitotic meristems (root and stem) (Da Silva et al., 2023), it can be inferred that detergents had an adverse impact on the cell division process in these areas, resulting in the formation of smaller cucumber seedlings.

Additionally, studies demonstrate that exposure to detergents has an adverse impact on the photosynthetic pigments of seedlings, resulting in a reduction of their photosynthetic rate (Hassouna et al., 2007; Uzma et al., 2018). Beyond this effect, these substances have the ability to induce ATP depletion in cells and inhibit subsequent ATP synthesis (Azizullah et al., 2012). This combined set of effects can have significant implications on the healthy growth of seedlings.

The ecotoxicity data presented in Table 2 demonstrate that the parameters assessed in the present study were affected by low concentrations of the commercial detergent. In a previous research, the researchers evaluated the inherent toxicity of two commercially used detergents (referred to as D1 and D2), using the plant species *Vicia faba* L. as the model organism. In this context, the average inhibitory concentrations (IC_{50}) related to root length were measured, and it was found that the IC_{50} for detergent D1 was 12 g L^{-1}, whereas for detergent D2, it reached a value of 16 g L^{-1} (Roccotiello; Viale; Cornara, 2011).

There were no data found in the literature that demonstrate environmentally relevant concentrations for detergent. However, it is important to highlight that the average amount of detergent consumed daily per person is 15.3 grams, which amounts to a total of 5.6 kilograms per year (Quevedo and Paganini, 2016). This value is considerably higher than the IC_{50} obtained in the present study, conducted with cucumber seeds. It is essential to emphasize that the excessive use of detergents can result in their inadequate disposal in the environment, leading to contamination of water and soil. Therefore, it is crucial to adopt awareness-raising measures and sustainable practices to reduce the environmental impact caused by using detergents.

CONCLUSION

In the present study, the detergent did not influence the germination percentage of Cucumis sativus seeds at any of the evaluated concentrations. However, toxic effects were observed on the germination speed index of the seeds at all tested concentrations. Furthermore, beginning from a concentration of 2 g L^{-1}, the seedlings' root length was significantly reduced, while the shoot length was affected at all concentrations under examination.

Based on the toxicity index and the median inhibitory concentration (IC_{50}), it can be affirmed that the various concentrations of the detergent exerted an extremely toxic effect on seedling development, with the shoot length being the most impacted parameter. The concentration that caused a 50% reduction in root length was calculated to be 3.04 g L^{-1}, whereas for shoot length, this effect was observed at a concentration of 1.56 g L^{-1}.

Therefore, it is evident that the introduction of detergents into the ecosystem can cause significant harm to plant development. It's worth noting that these organisms are present at the base of the food chain in both terrestrial and aquatic environments. Thus, adopting a conscientious and responsible approach is of the utmost importance, both in the utilization and disposal of detergents, with the aim of minimizing, to the greatest extent possible, any potential adverse impacts on the environment.

CREDIT AUTHOR STATEMENT

Study design: FJS; LMS.

Data curation: FJS; LMS.

Data collection: FJS; LMS.

Data analysis: FJS.

Original manuscript writing: FJS, LMS.

Review and editing manuscript: FJS; LMS.

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