The Influence of Calcium Chloride on Soil Animals, Microorganisms, and Phytoplankton

Man Kyu Huh

ABSTRACT

Calcium chloride (CaCl₂) is an inorganic chemical from the chloride group, and it is widely used as one of snow removal agents on the road in winter. Laboratory experiments were conducted to examine the effects of the road salt (NaCl) on soil organisms (a soil animal, microorganisms, and plankton). The soil dwelling Armadillidium vulgare died from high concentration of calcium chloride treatment. The longer the processing time of A. vulgare at the high concentration of calcium chloride, the lower the survival rate. A. vulgare died 16% at a calcium chloride concentration of 1 mM. In the disc diffusion test, the diameter of the inhibition zone proportionally increased with the concentration of calcium chloride. Micrococcus sp. was slightly higher in calcium chloride inhibitory activity than those of three soil microorganisms (Bacillus subtilis, Pseudomonas fluorescens, and Xanthomonas maltophilia). About 90% of microorganisms (phytoplanktons) died when the 15.0 mM calcium chloride solution was used. In this study, high concentrations of calcium chloride affected the survival of soil animals, soil microorganisms, and water microorganisms. If chloride dissolves in water and flows into rivers or lakes, it can cause destruction of soil or aquatic ecosystems and threat to the survival of small organisms.

Keywords: Armadillidium Vulgare, Calcium Chloride, Phytoplankton, Soil Microorganism.

I. INTRODUCTION

In winter, the road surface is snowy, covered with ice and packed snow. The impact of snowstorms on safety and mobility can be mitigated by effective road maintenance operations, such as de-icing, snow removal, and snow relocation [1]. Snow removal agents can function as ice making by preventing slipping on roads where cars travel. When there is a lot of snow, the snow removal car sprays calcium chloride on the road. As snow accumulates, it melts to some extent due to temperature or pressure, and the molten water and calcium chloride react exothermic to release heat [2]. This heat melts the surrounding snow to make water, and the water reacts with calcium chloride again to continuously melt the snow.

Calcium chloride as a snow removal agent has many advantages: it is very effective deicing agent, it is easy to store, handle and spread, and it also has relatively low price. However, it is widely known that calcium chloride is highly corrosive, damaging rebars in cars or concrete, and causing small trees around the road to die [3]. In addition, snow removal agents can affect the soil under or around street trees.

Due to the increase in population and the resulting increase in industrial and agricultural activities [4], various chemicals flow into the natural environment, polluting the ecosystem and posing a major threat to public environmental hygiene [5]. In particular, many chemicals have been used as a base for snow removal agents. Snow removal agents in winter helped road conditions, but the soil ecosystem was harmed as much [6].

The soil is inhabited by a large number of microorganisms, which are invisible to the eye, and they act as decomposers there to decompose and recirculate various substances. Armadillidium vulgare may reach a length of 18 millimeters and is able to withstand drier conditions than many other woodlouse species. A. vulgare is less susceptible to cold during the night and may enter a state of dormancy during the winter in order to survive temperatures that would otherwise be lethal [7].

Phytoplankton are the foundation of the aquatic food web, the primary producers, feeding everything from microscopic, animal-like zooplankton to high organism such as fish. Plankton is itself directly affected by pollution, but it can also act as either a conduit to or a shield for higher animals [8]. Distribution patterns of phytoplankton are strongly correlated with environmental factors [9]. Phytoplankton cause mass mortality in other ways. Calcium chloride sprayed on the road eventually enters the water system and affects phytoplankton. While plankton populations are needed for thriving rivers and lakes, and marine ecosystems, too many plankton in one area can create a serious environmental problem.

Exogenous calcium could alleviate the physiological growth stress of plants caused by snow melt [10]-[12].

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M. K. Huh * Food Engineering and Technology Major, Dong-eui University, Busan, South Korea (e-mail: mkhuh@deu.ac.kr)
*Corresponding Author
Although there are various results on the use of calcium chloride and on shading and roadside plants, there have been not many basic experiments and studies on soil animals and microorganisms where street trees live among the environmental side effects that occur after use. This was investigated. 1) If calcium chloride works on soil microorganisms, can they live? 2) Is there an effect of calcium chloride on earthworms that live in the soil because it is thought to have an effect on animals? 3) Can they live if calcium chloride acts on plankton that live in rivers or streams?

II. MATH

A. Calcium Chloride Solution

Calcium chloride is solid but turns into a liquid when sprayed on snowy roads. In addition, calcium chloride sold on the market was not constant in purity, so reagents were purchased and treated by concentration. Since the molecular weight was 110.9, 110.9 g of distilled water was added to 1000 ml of distilled water to become 1 mol (M), but since the concentration was too strong, 110.9 g was dissolved in 1000 ml of distilled water to prepare 1.0 mol, and then diluted to lower the concentration.

B. Effects of Calcium Chloride on Armadillidium Vulgare

After securing enough soil in the place where Armadillidium vulgare lived, it was sterilized with steam and used for this experiment. I prepared several boxes of soil for 50 A. vulgare each. At this time, the degree of reaction to calcium chloride will vary depending on whether the A. vulgare is young or mature, so measure the young and the large on a scale and mix them as evenly as possible. Each of a 100 ml calcium chloride solution was treated in a box containing 50 animals and mix well. Because the A. vulgare escapes easily from the box, the top of the box is covered with a double net. The survival rate of the A. vulgare in the box was investigated by sifting the soil every day. The survival rate was calculated when the population of the A. vulgare remained constant within the error range.

C. Effect of Calcium Chloride on Soil Microorganism

When microorganisms in the soil were grown in the medium, many microbial groups (coronis) appeared. In this group, one type of pure microorganism could be obtained by coating it with platinum and placing it in a liquid medium. Soil microorganisms are collected by type and grown homogenously at 3°C in medium. The average temperature in Busan is 3°C in January. Pure separation is the cultivation of microorganisms to cultivate pure single colonies in mixed cultures using flat media. In samples containing various types of bacteria, only one desired strain is selectively grown in another medium and only one pure species is cultured. For example, growth conditions for Bacillus subtilis are first tried to grow on 0.5 LB agar plate (1% tryptone, 0.5% Yeast extract, 0.5% NaCl and Agar 1.5%), after getting growth on LB agar plate. Other soil microorganisms were also grown on suitable media for their cultures.

Distilled water, calcium chloride 1.0 mM, 5.0 mM, 10 mM, 15 mM, and 20.0 mM are added to four agar mediums to form a group treated with calcium chloride by concentration and a first control group not treated. The disk diffusion test is used to determine the susceptibility of isolates of soil to different calcium chloride solutions. The inhibitory disc measured each zone with the unaided eye while viewing the inhibitory disc of the petri dish using digital vernier caliper (Mitutoyo, Japan).

D. Effects of Calcium Chloride on Phytoplankton

Phytoplankton is made up of very small, usually unicellular, plants. When calcium chloride was sprayed as a winter snow removal agent, microorganisms were collected where calcium chloride melted and flowed into the river. Plankton is caught with a plankton net. Three replicate samples of a known volume of subsurface (5–40 cm) water were taken with a 10 L bucket and then filtered through a plankton net.

Quantitative analysis of plant plankton was identified after fixing the sample of 2 with a Lugol solution. After quantifying to 5% of plant plankton, calcium chloride solutions were treated by concentration to investigate whether they survived. Plankton are observed with a microscope (Olympus BH-2).

E. Data Analysis

Data was conducted using Microsoft Excel and SPSS 21.0 for Windows (Chicago, IL, USA). A one-way and a two-way analysis of variance (ANOVA) followed by the Tukey post hoc test were used to analyze statistical significance (p < 0.05).

III. RESULTS

The soil dwelling Armadillidium vulgare died from high concentration (20 mM) of calcium chloride treatment (Fig. 1). The longer the processing time of A. vulgare at the high concentration of calcium chloride, the lower the survival rate. A. vulgare died 16% at a calcium chloride concentration of 1 mM (Fig. 2). Half of A. vulgare died at a calcium chloride concentration of 5 mM. At calcium chloride concentrations of 10 mM and 20 mM, only 12% and 6% of A. vulgare survived, respectively.

![Fig. 1. a) Earthworm (armadillidium vulgare); b) Living individuals on control group (0.0 mM CaCl2), right: dead individuals on 20 mM CaCl2.]

Microorganisms cultured in liquid media. These pure separated individuals were sufficiently cultured. Cultures of Bacillus subtilis, Micrococcus sp., Pseudomonas fluorescens, and Xanthomonas maltophilia isolated principally from soil were tested for sensitivity to calcium chloride.
Calcium chloride concentrations were also dropped and mixed one drop at a time, placed in a 3°C thermostat for 30 minutes, and observed under the disc diffusion methods to observe whether growth inhibition occurred (Fig. 3). In the disc diffusion test, the diameter of the inhibition zone proportionally increased with the concentration of calcium chloride (Table I). Overall, Micrococcus sp. was slightly higher in calcium chloride inhibitory activity than those of three soil microorganisms (Bacillus subtilis, Pseudomonas fluorescens, and Xanthomonas maltophilia). However, there was no significant difference among four groups (p > 0.05).

Calcium chloride is an inorganic chemical from the chloride group and one of the most popular deicing agents used by snow removal and ice management experts globally. Calcium chloride is often used in food, though in small quantities. It is a compound safe for human consumption and addition a salty flavor. It can replace traditional road salt when used in a mixture of 80% quartz sand and 20% calcium chloride. Sodium chloride, regarded as a traditional road salt, has been used for years for de-icing road surfaces, driveways and pavements. It causes degradation of the natural environment, which is manifested by inhibition of the growth of plants growing near the roadway and the extinction of living organisms. Its negative effects can be reduced by using phosphate additives.

Calcium chloride also has a negative effect on the human body. The damage of calcium chloride through the dermis was assessed by histopathologic examination and hyperbaric oxygen (HBO) group showed a statistically significant preservation of epidermis and dermis with minimal necrosing findings of skin injury [17].

In the 20 mM calcium chloride treatment, all 100% of the microorganisms died, and some of the microorganisms were decomposed and only the remaining microorganisms were sparsely.

IV. DISCUSSION

There are five most frequent natural disasters in South Korea which is heavy rainfall, typhoon, wind waves, windstorm, and heavy snow [13]. Heavy snow damage is the third-highest natural disaster after heavy rain and typhoon damage. However, recently, the heavy snow also caused severe damages due to the climate change.

Rapid snow removal in roads is an essential chore during the winter months. Rapid snow removal is significantly important due to difficult occurrence estimation of heavy snowfall disasters, local snowfall pattern, and the expectation of the cycle by global warming and climate change [14]-[15].

Chloride-based salts are the most common chemicals used to serve as freezing-point depressants for winter road service applications such as sodium chloride (NaCl), magnesium chloride (MgCl₂), and calcium chloride (CaCl₂) [16].

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TABLE I: SIZE OF FILTER PAPER DISK ACCORDING TO CALCIUM CHLORIDE CONCENTRATION (mM)

<table>
<thead>
<tr>
<th>Species</th>
<th>Concentration (mM)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>Bacillus subtilis</td>
<td>0.4±0.03</td>
</tr>
<tr>
<td>Micrococcus sp.</td>
<td>0.7±0.02</td>
</tr>
<tr>
<td>Pseudomonas fluorescens</td>
<td>0.0±0.00</td>
</tr>
<tr>
<td>Xanthomonas maltophilia</td>
<td>0.0±0.00</td>
</tr>
</tbody>
</table>

Data represented the mean ± SD from three replicates.

Fig. 2. Earthworm (Armadillidium vulgare) survival rate according to calcium chloride concentration.

Fig. 3. Measuring zones of inhibition. Minimum inhibitory concentration experiment on soil microorganisms.

Fig. 4. Survival in plant plankton according to calcium chloride concentration: a) 1.0 mM; b) 10.0 mM; c) 15.0 mM.
In this study, high concentrations of calcium chloride affected the survival of soil microorganisms (Table 1). In a *Bacillus subtilis* mutagenicity assay the potential of calcium chloride to damage cellular DNA was examined at concentrations up to 0.5 M [18]. The result of the test was negative. In an *Escherichia coli* test the potential to induce an SOS response was tested at doses up to 1 mM, also giving a negative result [19]. Laboratory studies determined the effects of increasing NaCl concentrations on aquatic invertebrate drift, behavior, and survival [20]. Studies were made of the effect of calcium chloride acetate on soil [21], water, vegetation etc. The foremost potential environmental effect, which was found, and which was also referred to in the initial investigations was that even relatively low concentrations of calcium chloride can reduce the amount of oxygen in water. The bacteria which decompose acetate consume oxygen. In an investigation performed in test pools in the field, it was demonstrated that biochemical oxygen demand increased when calcium chloride was added [22]. Concentrations as low as 10 mg calcium chloride acetate/litre could cause significant reduction in the oxygen content of water. Oxygen depletion makes it difficult for aquatic organisms such as plankton to survive (Table I, Fig. 4).

It is better to use limited amount of calcium chloride as needed when there is a lack of labor force required for snow removal, or when it is not easy to remove snow from the physical force due to road conditions. However, in reality, the amount of calcium chloride is more than necessary due to the urgency of work. It not only corrodes roads, cars, steel structures, but also pollutes the roads by polluting the soil around the roadside trees, polluting the soil, water quality and even the atmosphere.

The chemical is currently of low priority for further work on the hazard to the environment. However, because of the effects of calcium chloride on soil dwelling organisms and plants and the exposure associated with the use of calcium chloride as a deicing agent in some countries, these countries may decide to assess the environmental risk related to this exposure scenario [23].

V. CONCLUSION

CaCl2 is intended for use as an ice and snow removal agent on highways, bridges, and other roadways. Using excessive fields as snow removal agents on roads in winter can pose a threat to the survival of small organisms or microorganisms that cannot move freely.

CONFLICT OF INTEREST

Author declares that he does not have any conflict of interest.

REFERENCES


