*Environmental Engineering and Management Journal*

November 2021, Vol. 20, No. 11, 1787-1795
http://www.eemj.icpm.tuiasi.ro/; http://www.eemj.eu

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# OZONE PRETREATMENT TO IMPROVE THE PHYSICO-CHEMICAL AND BIOLOGICAL PROPERTIES OF LIVESTOCK FECAL WATER

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## Abstract

Ozone is a strong oxidant which has a good degradation effect on organic matter and a good disinfection effect. In the present study, the effects of different ozone concentration and treatment time on total nitrogen, total phosphorus, total potassium, organic matter (OM), 5 day biological oxygen demand ($BOD_5$), chemical oxygen demand ($COD_{Cr}$) and hygiene indexes in pig manure water under the condition of 5% total solid concentration (TS) were studied. The current research aims at optimizing concentration and time of ozone treatment in pig manure water. Therefore, 5 different ozone concentrations (i.e. 0, 60, 120, 180 and 240 mg/L) are applied to treat fecal water for 10, 20, 30 and 40 minutes. The results showed that the pH, OM content, total phosphorus, total potassium and total nitrogen content of fecal water do not change significantly. On the other hand, with the increasing ozone concentration and treatment time, the contents of $BOD_5$, $COD_{Cr}$ and suspended solid (SS) in fecal water have a decreasing trend. While the mortality rate of eggs of *Ascaris lumbricoides* were 95%, the number of *E. coli* groups was $10^{-4}$ as the standards. According to the obtained data, the effect of ozone has enhanced with increasing ozone concentrations. The optimal treatment condition was applying ozone at 240 mg/L for 40 minutes.

*Key words:* disinfection, *E. coli*, fecal water, ozonation, organic matter

*Received: July, 2020; Revised final: February, 2021; Accepted: September, 2021; Published in final edited form: November, 2021*

## 1. Introduction

China is a biggest country in the world of animal husbandry. The amount of pig breeding accounts for about 50% of the total amount of pig breeding in the world (Xin et al., 2014). This means that a large amount of manure, both solid and liquid, is produced each year. This large amount can cause serious environmental pollution if not handled properly (Shi et al., 2011). At present, solid feces are well treated by composting on contrary to the fecal water which is more difficult to be treated although it accounts for more than half of livestock and poultry feces.

Various problems associated with fecal water including the high content of refractory organic matter, the imbalance of carbon and nitrogen ratio, the relatively concentrated discharge time, and the large change in water quality and quantity increased the difficulty of treatments (Field and Samadpour, 2007).

Traditionally, the livestock and poultry fecal water has been treated through long-term storage in biological ponds for anaerobic fermentation. However, it is inappropriate because of high investment cost, long storage time and unsatisfactory treatment effect (Zhang et al., 2019). It is particularly important to develop a new, efficient and environmentally suitable method to treat livestock

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fecal water. The method that aims to possess higher utilization of livestock and poultry waste resources.

Ozone can be used as an electrophile to react directly with other chemicals or indirectly react with hydroxyl radicals and compounds produced by it, hence suitable for wastewater treatment (Buffle et al., 2006). Meanwhile, ozone is a strong oxidant with a redox potential of 2.07 V. Due to its high oxidizing power, its use in the removal of hard or non-biodegradable and toxic pollutants in water and wastewater treatment processes has increasingly expanded (Ayoubi-Feiz et al., 2019; Chen and Wang, 2019). Many studies have confirmed that ozone has a good treatment effect on wastewater contaminants produced by pharmaceutical, printing, dyeing, papermaking and other industries (Huan et al., 2015; Shenyong et al., 2015). As has recently been shown by Xianhua (2014), ozone treatment (30 mg/L) has a significant effect on the chroma reduction of wastewater. Malakootian et al. (2020b) found ozone treatment (30 mg/L) has a significant effect on the chroma reduction of wastewater. In addition, ozone can also be used for disinfection. Ozone oxidation treatment can improve the sewage biochemical properties and effectively inactivated microorganisms (Arnold et al., 2018). Pines and Reckhow (2002) proposed that Co (II) and Fe (II)/UVA/ozone catalytic ozonation mechanism, improving the degradation rate of organic pollutants in wastewater. Shengyan (2018) shows that under the effect of acidic electrolysis of water-ozone mixture, the microbial killing effect can reach 85.25%. Ozone, as a high efficiency disinfectant, has been used in the treatment of industrial wastewater with the characteristics of high efficiency and saving, and its application mostly adopts the mode of catalytic combination to achieve high efficiency treatment.

The most important pollution of farm pollutants is fecal water, which contains urine, feed residue, mixed feces and enclosure washing water. The untreated fecal water contains a large number of organic pollutants. The water pollution caused by feces and urine mainly includes: first, feces and urine in the livestock house flow directly into the water environment; second, the long-term accumulation of pig manure is washed by rainfall or slowly infiltrates into the underground water source. After a large amount of organic substances are discharged into the natural water source. The dissolved oxygen in the water is greatly consumed, resulting in stink in the water and high concentration of nitrate and nitrite. Therefore, cause poisoning for people and animals if they drink it for a long time. The study found that the important source of copper, zinc and other heavy metals in the soil is pig manure and urine (Wang et al., 2011). The excrement of manure into the farmland may also lead to soil hardening and salinization, which greatly affects agricultural production (Tongxuan et al., 2019). If a large number of pathogenic microorganisms including eggs of parasitic worm remain in the wastewater for a long time. They can cause infection directly to livestock themselves and more likely to cause infectious diseases to spread from person to person. Serious circumstances will also lead to the occurrence of the epidemic (Caiying et al., 2011). Therefore, effective measures must be taken to treat the fecal water.

Meanwhile, ozone treatment can effectively reduce various industrial and organic contaminants in wastewater and improve the color of its to meet the discharging standards. Some studies (Jiangang., 2017; Yue., 2014) have shown that ozone treatment of wastewater can improve the biodegradability of wastewater and has a good inactivation effect on pathogenic bacteria. So, ozone can be used to treat the fecal water. This study focus on inactivation of pathogenic bacteria and reducing gas emissions, as well as keep organic and inorganic nutrients in fecal water to improve its fertilizing effect in farmland utilization. The present study intents to optimize concentration and time of ozone treatment of fecal water and provide reference for ozone treatment of livestock and poultry breeding fecal water. In view of this, the study was designed to use the pig breeding fecal water as the treatment object, and study the changes in fecal water composition under different ozone concentrations and different aeration time conditions.

**Novelty:** This study aims at application of ozone to treat fecal water directly through adjusting the ozone concentration and processing time. Additionally, the effect of different treatment time and ozone concentration of fecal water physical and chemical properties was explored. The optimal concentration and time of ozone treatment were obtained by measuring the sanitary index and returning farmland index of manure water after treatment, and combining previous studies (They have been shown in references.) on single index of ozone.

## 2. Research ideas and methods

### *2.1. Experimental setup*

This experiment started in July 2019 and was conducted in China Agricultural University (Yantai). The experimental materials were taken from a pig farm in Yantai City, Shandong Province, China.

There were 5 treatments of ozone concentrations in the experiment, i.e 0 (treatment 1 Control), 60 (treatment 2) 120 (treatment 3), 180 (treatment 4) and 240 mg/L (treatment 5). Each concentration of ozone was applied to 2 L of fecal water through the ozone generator (JDWX-MBOG-0.1KG, Jinda Wanxiang Environmental Protection Technology Co. Ltd., Beijing, China), rated ozone output is 100 g/L, and oxygen flux is 1 $Nm^3/h$. The ozone concentration can be controlled by adjusting the concentration control knob of the ozone generator and the effective area of the aerator is 332.84 $cm^2$, the outer diameter is 180 mm, and the height is 5 cm. The laboratory temperature was controlled by the air conditioning device to be 28°C which could keep the temperature of the water in a stable range. In this experiment, we use a self-designed ozone-fecal water treatment device (Fig. 1), which can continuously provide ozone to the aeration tank in the reaction process.

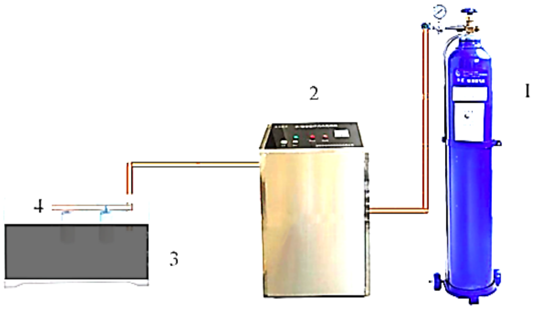


**Fig. 1.** Schematic diagram of ozone treatment system (1-Oxygen bomb; 2-Ozonation generator; 3-Aeration tank; 4-Aerator)

*2.2. Physico-chemical analysis*

After 10, 20, 30 and 40 minutes of ozonation, 50 ml of fecal water was collected for sample analysis. Total nitrogen was determined by Kjeldahl method (Kjeldahl, 1884), total phosphorus by spectrophotometry (Burton and Riley, 1956) and total potassium by flame photometry (Junsomboon and Jakmunee, 2011) in fecal water. We use spectrophotometry method to determine ammonia nitrogen content (Kunning et al., 2019). The suspended solids in fecal water were determined by gravimetric analysis (Yingping et al., 2018), and the organic matter content was determined by Wet Oxidation Method (Gong et al., 2009).

Biological oxygen demand ($BOD_5$) in fecal water was determined by five-day biochemical cultivation. As for chemical oxygen demand ($COD_{Cr}$), we use microwave digestion method to determine it (Rice et al., 2017).

*2.3. Biological analysis*

To confirm the *E. coli* in samples, a certain amount of water sample was taken in the medium of lactose peptone for the initial fermentation experiment, and then the plate separation and double negative fermentation experiment was identified. The most probable number (MPN) table was searched according to the number of tubes fermented at each dilution degree, and the number of coliform groups in each 10 mL or liter of water sample was obtained (Maheux et al., 2008).

Prior to the separation of *Ascaris lumbricoides* egg, 1600 ml of detergent (0.0025% T tween-20 solution) is added and 50 g of sludge sample is mechanically mixed over a long period of time (4 hours) to release eggs in the flocculate. Flotation was then carried out in a saturated $NaNO_3$ solution. Then centrifuge for 10 min (2500 g/min). The eggs were fixed on the filter and observed through a microscope at 200 times magnification. Eggs with significant deformities are considered dead (Zdybel et al., 2019). By detecting these indicators, firstly, the degree of pathogenic bacteria killing in fecal water can be detected, and secondly, whether the nutrients returned to farmland in fecal water react to disappear can be understood, and three samples were taken for chemical analysis.

*2.4. Statistical analysis*

We use Excel 2016 and SPSS 17.0 for data statistical analysis. For follow-up of ANOVA included the Tukey HSD Test.

**3. Result and discussions**

The indexes of initial fecal water are shown in Table 1.

**Table 1.** Physicochemical properties of initial fecal water

| ***Item*** | ***Numerical value*** | ***Item*** | ***Numerical value*** |
|---|---|---|---|
| pH | 7.774 | *E. coli* ($10^4$ cfu/mL) | 2.55 |
| TS (%) | 5 | Total nitrogen (%) | 0.061 |
| Organic matter (%) | 1.82 | Total phosphorus (%) | 0.029 |
| SS (mg/L) | 34960 | Total potassium (%) | 0.088 |
| $BOD_5$ (mg/L) | 5495.3 | Ammonia nitrogen (mg/L) | 0.07 |
| $COD_{Cr}$ (mg/L) | 29450 | Egg mortality of *Ascaris lumbricoides* (%) | 33.2 |

*3.1. Effect of ozone treatment on pH in fecal water*

The effect of varied ozone concentrations on the pH of fecal water is shown in Fig. 2. After ozone treatment, pH of fecal water remained basically unchanged. Because the initial pH of the control sample was not same, the pH of the fecal treated with different concentrations of ozone was basically similar to that of the control sample. After 60 mg/L ozone treatment, the pH of fecal water don't change significantly. The pH of feces treated with ozone for 10, 20, 30 and 40 min had no significant difference, and the pH value remained at about 7.8. The pH variation trend of 120 mg/L and 240 mg/L ozone treatment was similar to that of 60 mg/L ozone treatment. In the 180 mg/L ozone treatment, no significant difference can be seen in control, 20 min, 30 min and 40 min treatments, while the pH of 10 min treatment was significantly higher than that of other treatments. In general, when fecal water was treated with different concentration of ozone, pH was remained the same.

As the results indicated in Fig. 2, the increase of ozone concentration and the extension of treatment time did not show a significant change in pH of fecal water except 180 mg/L treatment. The non-significant in pH values may be due to limited ozonation process in which ozone reacts with nitrogen in the water (mainly nitrite) to produce a large number of hydrogen ions increasing the acidity of water (Changping et al., 2010), the limitations of ozonation process, in the present could explained by low nitrite content in raw water which resulting a small amount of ozone reactions that finished in a very short time, so negligible acidity was recorded in the fecal water. The influence of ozone decomposition into hydroxyl free radicals improves the reaction rate to positive, but significantly reduced the efficiency of ozone reaction medium (Gomes et al., 2012). The formation of these hydroxyl free radicals may also convert samples to neutral or weak alkaline, and produce little influence on the reaction rate. Some studies reported that with the decrease in water pH the rate of dissolved ozone concentrations was increases with time (Karnik et al., 2005). This indicate that pH level playing crucial role on the efficacy of ozone even at lower concentration.

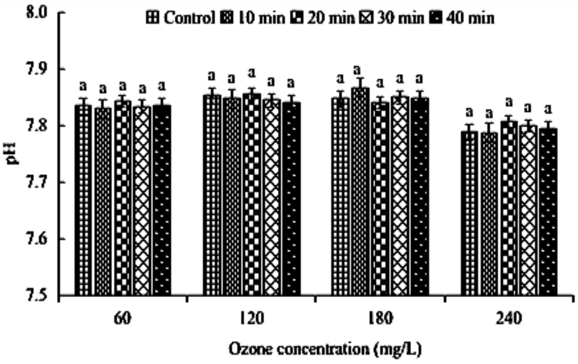


**Fig. 2.** Effect of ozone treatment on pH of fecal water (Explanation: Each value represents the mean of 3 replicates; The different lower-case letters stand for significant differences at 0.05 level between treatments. The same are as followings)

*3.2. Effect of ozone treatment on organic matter content in fecal water*

Effect of varied concentrations of ozone with different aeration time on organic matter content are shown in Fig. 3. After ozone treatment, the organic matter content of fecal water treated with ozone was significantly lower than that of control, and the lowest value was found in the treatment with ozone for 40 min. When ozone was applied at concentrations of 60 mg/L, 120 mg/L, 180 mg/L, 240 mg/L for 40 min, organic matter content of the treated fecal water was 54.2%, 50.8%, 43.2%, respectively. These values were all lower than the control (65.7%).

Experimental results showed that due to ozone treatment of fecal water organic matter contents reduce more apparently, with the increase of ozone concentration and prolonged treatment time. As shown in the Fig. 3, increasing ozone application time (10 min, 20 min, 30 min and 40 min) significantly decreased OM content of fecal water at all $O_3$ application rates under the experiment condition. The decrease in OM content after ozonation process could refer to the oxidation activity of $O_3$ react with organic compounds containing unsaturated bonds and destroy their structure (Westerhoff et al., 1999). Additionally, the increase dissolved ozone contents in fecal water encourages dissolved organic carbon into biodegradable dissolved organic carbon (by-products) such as organic acids, aldehydes and ketones acid leading to reduce the content of organic matter.

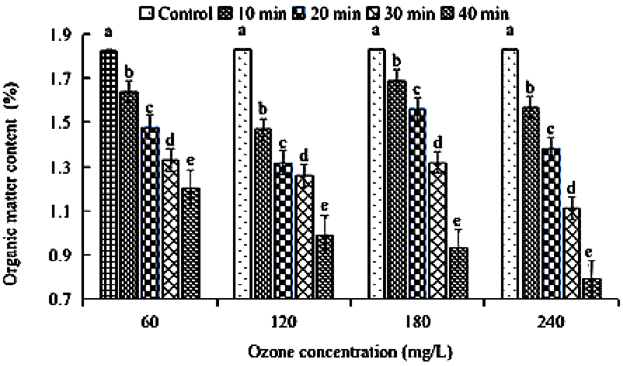


**Fig. 3.** Effect of ozone treatment on organic matter content in fecal water

However, properties of natural organic matter and its quantity could affect the oxidation rate (Ohlenbusch et al., 1998; Siddiqui et al., 1997). Ozonation transform the dissolved organic matter into low molecular weight substances. Previously, transformation of dissolved organic matter into dissolve organic carbon is reported by Galapate et al. (2001), further, low molecular organic carbon is some time completely degraded. However, complete decomposition of higher molecular weight organic substances is not commonly reported by ozonation.

*3.3. Effect of ozone concentration on total nitrogen, total phosphorus and total potassium contents of fecal water*

In the Table 2, we can see the influence of different ozone concentration treatments on total phosphorus, total potassium and total nitrogen content of fecal water. When the ozone treatment was 60 mg/L, the total nitrogen content of the ozone-treated fecal water decreased greatly compared to control (around 72.3% of the controls) at 20, 30 and 40 min of exposure.

On the other hand, total phosphorus contents of fecal water were increased significantly after ozone treatments at different time of exposure. This increase of total phosphorus were more rapid after higher concentrations of ozone i.e 180 and 240 mg/L. When ozone treatment was 60 mg/L, no significant difference can be seen in control and 10, 20, 30 and 40 minutes of ozone treatment, respectively. The variation trend of total potassium content in 120 mg/L,180 mg/L and 240 mg/L treatments was similar to that in 60 mg/L treatment. Overall, there was no significant change in total phosphorus, total potassium and total nitrogen content of fecal water.

After ozone treatment, total nitrogen did not change significantly with time and ozone concentration as it basically remained in a relatively stable range. It may be due to the process of ozone aeration in which granular organic nitrogen is continuously decomposed under the strong oxidation of ozone to be transformed into soluble organic nitrogen. This reaction is very rapid, so the changes in total nitrogen content will not change significantly with the increase of time. At the same time, ozone aeration has a significant effect on the form and content of phosphorus.

With the increase of aeration time, the contents of soluble total phosphorus, soluble inorganic phosphorus, total organophosphorus and granular organophosphorus decreases, while the contents of soluble organophosphorus, granular total phosphorus and granular inorganic phosphorus increase with the increase of aeration time, but the total phosphorus content has no significant variations. For potassium contents, the non-significant differences among treatments could be referring to formation of new substances containing potassium as a result of reaction of ozone with the substance containing potassium. Accordingly, potassium was retained in the fecal water as new substances (Jiangang, 2017; Yue, 2014). The contents of total phosphorus and total potassium in some ozone-treated fecal water were higher than those in control, but there was no significant difference with the control group, so it had no negative effect on the test results.

**Table 2.** Effect of ozone concentration on total phosphorus, total potassium

| ***Treatment*** | ***Total nitrogen (%)*** | | | | |
|---|---|---|---|---|---|
| | 0 min | 10 min | 20 min | 30 min | 40 min |
| ***Control*** | 0.061a | 0.061a | 0.061a | 0.061a | 0.061a |
| ***60 mg/L*** | 0.061a | 0.058a | 0.054a | 0.056a | 0.054a |
| ***120 mg/L*** | 0.061a | 0.058a | 0.061a | 0.059a | 0.056a |
| ***180 mg/L*** | 0.061a | 0.059a | 0.058a | 0.057a | 0.060a |
| ***240 mg/L*** | 0.061a | 0.060a | 0.058a | 0.054a | 0.060a |
| | ***Total phosphorus (%)*** | | | | |
| | 0 min | 10 min | 20 min | 30 min | 40 min |
| ***Control*** | 0.029a | 0.029a | 0.029a | 0.029a | 0.029a |
| ***60 mg/L*** | 0.029a | 0.027a | 0.028a | 0.030a | 0.028a |
| ***120 mg/L*** | 0.029a | 0.031a | 0.028a | 0.030a | 0.028a |
| ***180 mg/L*** | 0.029a | 0.032a | 0.028a | 0.031a | 0.031a |
| ***240 mg/L*** | 0.029a | 0.029a | 0.029a | 0.031a | 0.031a |
| | ***Total potassium (%)*** | | | | |
| | 0 min | 10 min | 20 min | 30 min | 40 min |
| ***Control*** | 0.088a | 0.088a | 0.088a | 0.088a | 0.088a |
| ***60 mg/L*** | 0.088a | 0.092a | 0.092a | 0.086a | 0.087a |
| ***120 mg/L*** | 0.088a | 0.089a | 0.088a | 0.088a | 0.088a |
| ***180 mg/L*** | 0.088a | 0.089a | 0.092a | 0.088a | 0.086a |
| ***240 mg/L*** | 0.088a | 0.086a | 0.088a | 0.086a | 0.086a |

*Explanation: Each value represents the mean of 3 replicates; The different lower-case letters stand for significant differences at 0.05 level between treatments. The same are as followings.*

### *3.4. Effect of ozone treatment on ammonia nitrogen content in fecal water*

Variation in the ammonia nitrogen content caused by ozone at different aeration times are shown in Fig. 4. When the ozone concentration was 60 mg/L, there were significant differences among all treatments, and the lowest value was recorded at 30 min, which was 86.1% of control. When the ozone concentration was 120 mg/L, the variation trend of ammonia nitrogen content was similar to 60 mg/L ozone treatment, and the lowest value was recorded at 40 min, which was 92.2% of control. When the ozone concentration was 180 mg/L and 240 mg/L, the variation trend was similar to 60 mg/L, and there were significant differences among all treatments. The lowest ammonia nitrogen content was found at 40 min and 30 min, which were 91.6% and 84.5% of control, respectively. In general, after ozone treatment, ammonia nitrogen content showed a decreasing trend, and the lowest content was 240 mg/L at 30 min.

As can be seen, ammonia nitrogen content has a decreasing trend as ozone concentration rises and treatment time lengthens. Ammonia nitrogen may be converted in two ways: first, it is volatized in a gaseous form, or it is oxidized to nitrate nitrogen by ozone. The ammonia nitrogen content measured in this study was relatively small and the reaction was relatively rapid, and its content did not change significantly with the increase of time and the increase of ozone concentration. this results may owing to the pH value and temperature of fecal water that affect the dissolution of ozone and the formation of $OH^-$, which weakens the reaction between ozone and ammonia nitrogen (Fernando, 2003; Jiangang, 2017; Vajnhandl and Le-Marechal, 2005; Yue, 2014). On ozonation of water ammonia nitrogen is decomposed into $NO_3^-$ ($NH_4^+ + 4O_3 \rightarrow NO_3^- + H_2O + 2H^+ + 4O_2$). These protons produce during decomposition of ammonia nitrogen were neutralized with $OH^-$to keep the pH unchanged (Krisbiantoro et al., 2020).

However, pH of wastewater plays crucial role in the complete composition of ammonia nitrogen upon ozonation. Further, different catalyst like cobbalt oxide, magnesium oxide and bromide were recommended for fast and effective removal ammonia nitrogen from wastewater (Ruffino and Zanetti, 2019; Krisbiantoro et al., 2020).

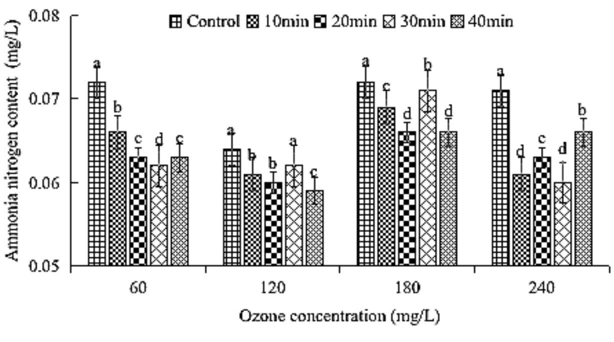


**Fig. 4.** Effect of ozone treatment on ammonia nitrogen content in fecal water

### *3.5. Effect of ozone treatment on suspended solids (SS), $COD_{Cr}$ and $BOD_5$ content in fecal water*

The influence of ozone treatment at different concentrations on suspended solids (SS) content of fecal water is shown in Fig. 5. After 10 min of ozone treatment, SS content of fecal water decreased slightly compared to control, among which the ozone concentration of 240 mg/L was the lowest. However, 180 mg/L treatment showed a little increase compared to control. Compared with ozone treatment for 10 min, SS content of fecal water after ozone treatment at different concentrations showed general decrease after 20 min. At 60 mg/L of ozone, SS found to be the lowest in fecal water. SS in controls samples remained unchanged with increase in time. However, at varying concentrations of ozone SS fluctuate rapidly in fecal water with increase in time. As shown in Fig. 5, the ozonation-treated fecal water showed significant decrease in SS content of all treatments compared to the control group.

Studies have shown that suspended solid (SS) contents in fecal water tend to decrease. As has recently been shown (Pak et al., 2016), along with the increase of the SS concentration ozone disinfection efficiency decline as SS consumes ozone to produce all kinds of free radicals. Kawara (1997) reported that ozone oxidation decomposed the high molecular weight compounds found in SS, and then further decomposition of low molecular weight compounds takes place. SS content reduced slightly in our study because of shorter processing time. As for the phenomenon that some SS content in Fig. 5 is higher than the control, it may be caused by the failure to mix the samples fully during the experimental sampling or detection, or the experimental errors.

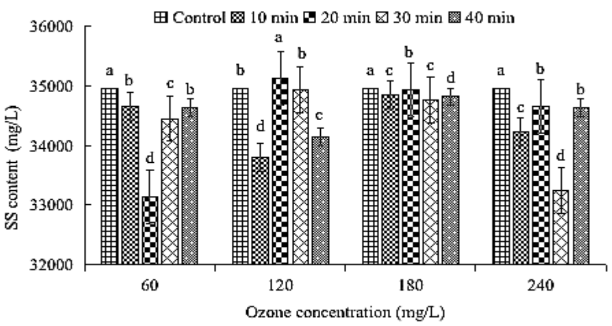


**Fig. 5.** Effect of ozone treatment on SS content in fecal water

In samples collected from ozone treated fecal water, the $COD_{Cr}$ of fecal water was significantly lower than controls and the lowest was observed at 40 min of exposure (Table 3). $COD_{Cr}$ in fecal water was decreased continuously with the increase in ozone concentration from 60 to 240 mg/L. Similar trend in the $BOD_5$ of fecal water was observed after treating with increasing concentration of ozone. $BOD_5$ in the fecal water was the highest in controls and the lowest fecal water samples treated with 240 mg/L of ozone at 40 min treatment, which was 9.8% of control. Time of exposure of ozone significantly affect the $BOD_5$ in the fecal water. Accordingly,the results showed that ozone treatments significantly reduce the content of $COD_{Cr}$ and $BOD_5$ in fecal water, especially at higher ozone concentration (240 mg/L).

This may be because when the ozone concentration rises to 240 mg/L, the pH of fecal water may be maintained at favorable conditions for the degradation of pollutants. Moreover, with the extension of treatment time, the treatment effect was significantly improved (Garcia-Morales et al., 2013; Jae-Wook et al., 2004; Xinchao et al., 2010). Recently, similar results of reduced (98% to 99%) $COD_{Cr}$ upon wastewater ozonation were observed by Khani et al. (2020) and Malakootian et al. (2020a).

### *3.6. Effect of ozone treatment on the number of E. coli flora groups and Ascaris lumbricoides egg mortality*

Ozone treatment was found effective in killing *Ascaris lumbricoides* egg and *E. coli* flora groups in fecal water (Table 4). With the increase of ozone concentration and the extension of treatment time, the killing effect was enhanced significantly. When ozone treatment of 60 mg/L was applied, colony forming units (CFU) of *E. coli* flora groups were significantly declined compared to control to reach its minimum (58.4% of control) after 40 min. Similarly, higher concentrations of ozone (120, 180 and 240 mg/L) showed more effective decline in CFU of *E. coli* flora group over the time. The lowest CFU was observed when fecal water treated with 240 mg/L of ozone for 40 min. Compared with control, eggs mortality of *Ascaris lumbricoides* in fecal water in all treatments increased significantly.

The results show that ozone treatment time has a significant effect on eggs mortality of *Ascaris lumbricoides*. When ozone treatment of 60 mg/L was applied, the highest value of eggs mortality of *Ascaris lumbricoides* was recorded at 40 min, which is 2.82 times of control. On all the tested concentrations, 40 min of exposure displayed maximum egg mortality. Similarly, egg mortality also increased greatly with the increase of ozone concentration. In general, all groups had significant differences with control, and the number of *E. coli* flora groups and *Ascaris lumbricoides* egg mortality dropped greatly.

**Table 3.** Effect of ozone treatment on $COD_{Cr}$ and $BOD_5$ in fecal water

| ***Treatment*** | ***$COD_{Cr}$ (mg/L)*** | | | | | ***$BOD_5$ (mg/L)*** | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | ***0 min*** | ***10 min*** | ***20 min*** | ***30 min*** | ***40 min*** | ***0 min*** | ***10 min*** | ***20 min*** | ***30 min*** | ***40 min*** |
| ***Control*** | 29450a | 29450a | 29450a | 29450a | 29450a | 5495.3a | 5495.3a | 5495.3a | 5495.3a | 5495.3a |
| ***60 mg/L*** | 29450a | 25700b | 21000cd | 17250d | 14900e | 5495.3a | 3106.3b | 1933.3cd | 1438.3e | 1373.3ef |
| ***120 mg/L*** | 29450a | 23290bc | 17840d | 17710d | 15970e | 5495.3a | 3269.8b | 2315.7c | 1984.3d | 980.1f |
| ***180 mg/L*** | 29450a | 27600b | 23350bc | 22400c | 13340ef | 5495.3a | 2989.6b | 2091.7c | 1681.7d | 717.7f |
| ***240 mg/L*** | 29450a | 25340b | 23410c | 17340d | 11680f | 5495.3a | 2679.6bc | 1881.7c | 1374.7e | 537.7g |

**Table 4.** Effect of ozone treatment on the number of *E. coli* flora groups and *Ascaris lumbricoides* egg mortality

| Treatment | *E. coli flora groups* ($10^4$ CFU/ml) | | | | | *Ascaris lumbricoides egg mortality* (%) | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | 0 min | 10 min | 20 min | 30 min | 40 min | 0 min | 10 min | 20 min | 30 min | 40 min |
| ***Control*** | 2.55a | 2.55a | 2.55a | 2.55a | 2.55a | 33.20g | 33.20g | 33.20g | 33.20g | 33.20g |
| ***60 mg/L*** | 2.55a | 2.26b | 1.99bc | 1.66cd | 1.49de | 33.20g | 68.70f | 82.33cd | 90.70b | 93.50a |
| ***120 mg/L*** | 2.55a | 1.98bc | 1.317e | 0.72ef | 0.397gh | 33.20g | 74.67e | 86.59c | 93.46b | 95.50a |
| ***180 mg/L*** | 2.55a | 1.43de | 0.89ef | 0.45g | 0.17hi | 33.20g | 79.27de | 89.37bc | 94.72ab | 95.90a |
| ***240 mg/L*** | 2.55a | 1.27e | 0.46g | 0.22h | 0.097i | 33.20g | 79.27de | 90.10bc | 94.90a | 97.48a |

This research has shown that ozone treatment has a significant effect on *Ascaris lumbricoides* egg mortality and the number of *E. coli* flora groups in fecal water. With the increase of treatment time and ozone concentration, the number of *E. coli* flora groups decreased significantly. Probably because ozone is highly oxidizing, that caused mortality of *E. coli* and the egg of *Ascaris lumbricoides*.

At higher concentrations, ozone interacts more rapidly with *E. coli* and egg of *Ascaris lumbricoides*, hence improve the effect of ozone sterilization. The mechanism of ozone sterilization may be acting on the bacterial cell membrane, destroying the cell membrane structure, causing metabolic disorders, inhibiting its growth and caused death (Szeto et al., 2020; Torrey et al., 2019).

## 4. Conclusions

Ozone aeration treatment of fecal water, due to its strong oxidation, has a good removal effect on organic matter, $COD_{Cr}$, $BOD_5$ as well as disinfection with *E. coli* groups and *Ascaris lumbricoides* egg mortality. Based on the results, ozone found to effectively degrade organic substances in livestock and poultry breeding fecal water, kill harmful bacteria and reduce environmental pollution and harm. Extending exposure time as well as increasing ozone concentration enhanced the efficacy of ozonation process.

Ozone oxidation is selective, and the degradation rate is affected by pH. Ozonation has no effect on the contents of total phosphorus, total potassium and total nitrogen content in fecal water. After ozone aeration treatment, the ammonia nitrogen content decreases slightly, but not significantly, while there is no significant change in the content of total nitrogen in fecal water with extending aeration time. It can be concluded that the optimal treatment time is 40 min and the optimal ozone concentration is 240 mg/L when the oxygen flux is $1Nm^3$ and the temperature is 28 ℃.

Therefore, according to the current situation of livestock and poultry dung water treatment, ozone aeration treatment can effectively clean and treat the breeding dung water, while retaining nutrients to meet the needs of returning to the field.

## Acknowledgements

This work was supported by Yantai Education Bureau Subject Development Project (2019XDRHXMXK25). We are equally indebted to Yantai Institute, China Agriculture University for providing research facilities.

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