RESEARCH ARTICLE

Investigation of microalgal treatment for poultry slaughterhouse wastewater after the dissolved air flotation unit

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ABSTRACT

Meat and meat products are some of the primary consumption products required for the continuation of life. The world population accessed over 7.5 billion that means the demand for food is increasing every day. Slaughterhouses and integrated meat facilities are being rapidly developed and established to satisfy meat and meat product requirements. In slaughterhouse poultry plants, high amounts of water are utilized for the meatpacking process. The poultry slaughterhouse wastewaters contain high levels of organic solids such as fat, blood, suspended matter, and dissolved protein, which can be treated using physical, chemical, and biological treatment methods. In this study, the treatment of poultry slaughterhouse wastewater preliminarily treated by dissolved air flotation, with microalgae culture (Chlorella Vulgaris) development, unlike traditional treatments, was investigated. Chemical oxygen demand and total suspended solids parameters for wastewater treatment have been monitored for 15 days of incubation. 0.8, 4, 8, 12, and 20\% by volume algae were applied for slaughterhouse wastewater, and the optimum amount of algal inoculation was determined after 15 days. When the removal efficiencies were examined, the most appropriate amount of inoculation rate with 76\% chemical oxygen demand removal and 87\% algal growth (as total suspended solids) was selected as 12\%.

Keywords: Chlorella Vulgaris, microalgae, poultry slaughterhouse wastewater

1. INTRODUCTION

Industry and technology have been developing day by day in our country. As a result, the population increases and the need for meat and meat products arises. According to OECD-FAQ, while meat consumption is 34.4 kg person\textsuperscript{-1} in the World, 32.4 kg person\textsuperscript{-1} is in Türkiye in 2019 [1]. As of 2019, 2335 thousand tons of poultry meat are produced in Turkey [2]. Therefore, slaughterhouse and meat integrated facilities numbers increasing day by day. However, meat processing in slaughterhouse and meat integrated facilities requires high amounts of water. When the wastewater of the integrated meat and slaughterhouse industry is discharged to the receiving medium without any treatment, it causes a decrease in the dissolved oxygen value of the receiving medium and the deterioration in the quality of the water environment [3].

The poultry slaughterhouse industry is also one of the rapidly developing industries. The poultry slaughterhouse industry wastewater contains high organic solids such as fat, blood, suspended matter, and dissolved proteins [4,5]. The organic substance content of the poultry slaughterhouse industry wastewater is high, and its temperature varies between 20-35 °C, and that can be treated in treatment plants where physical, chemical, and biological methods are used [5–11].

Microalgae's are heterotrophic and/or autotrophic, unicellular or multicellular organisms that grown in aquatic environments. Microalgae's are photosynthetic organisms and, CO\textsubscript{2}, water, and nutrients (e.g., nitrogen, phosphorus, and potassium) are necessary for their growth [12–14]. They can continue to live by adapting to adverse environmental conditions because they can overgrow. According to the types of algae and the culture conditions, the...
structure of algae (mainly lipids, carbohydrates, proteins) can alternate [14–17].

Advanced treatment systems, in which ammonia, nitrate, and phosphate can be removed, are high-cost systems compared to the traditional treatment systems. Microalgal treatment systems are favorable advanced treatment systems due to inorganic nitrogen and phosphorus degradation [18]. This study is an example to prove the feasibility of microalgae treatment to the slaughterhouse wastewater after the air flotation unit. Slaughterhouse wastewater contains a high amount of oil-grease and suspended solids that prevent light transmission and air transfer in water. Microalgae require light and optimum air transfer to develop, and slaughterhouse wastewater includes excessive level suspended solids for algal growth before air flotation. After the dissolved air flotation unit, the inhibitory organisms, oil-grease, total suspended solids are eliminated roughly %99 in slaughterhouse wastewater which makes a suitable environment for microalgae existence.

This study aims to investigate the treatment of poultry slaughterhouse wastewater coming from the pre-treatment and dissolved air flotation units by microalgae’s (Chlorella Vulgaris) at a laboratory scale. These wastewaters have a suitable medium for the growth of microalgae. Air floated poultry wastewater different inoculum rates of total wastewater volume 0.8, 4, 8, 12 and 20 % were investigated for 15 days. Chemical Oxygen Demand (COD) and Total Suspended Solid parameters were measured to determine the treatment of wastewater. While the studied optimum inoculum rate was specified as 12 %, in the same ratio, COD removal 76% and 87% algae growth as mass was observed.

2. MATERIALS AND METHOD

2.1. Reagents and chemicals

All chemicals and reagents have been supplied by Merck Millipore Company.

2.2. Microalgae cultivation

Chlorella Vulgaris has been preferred as a convenient algae culture to realize the objectives of the current research. The algae culture cultivated in Sakarya University, Department of Engineering, Sakarya, TURKEY. Chlorella Vulgaris were cultured before for this study at BG-11 medium (NaNO3, 1.5 g L–1; K2HPO4, 0.04 g L–1; MgSO4·7H2O, 0.075 g L–1; CaCl2·2H2O, 0.036 g L–1; citric acid, 0.006 g L–1; ferric ammonium citrate, 0.006 g L–1; EDTA (disodium salt), 0.001 g L–1; Na2CO3, 0.02 g L–1; 1 mL trace elements solution (in g L–1: H3BO3 2.86; MnCl2·4H2O, 1.81; ZnSO4·7H2O, 0.222; NaMoO4·2H2O 0.39; CuSO4·5H2O, 0.079; Co(NO3)2·6H2O, 0.0494 at pH 7±2). The BG-11 media and all glassware were autoclaved at 15 Psi and 120 °C for 20 minutes for sterilization.

2.3. Wastewater samples

Air floated poultry slaughterhouse wastewater (AFPSWW) has rich content; it provides a suitable environment for algal culture development [6]. The PSWW samples were obtained from a local poultry slaughterhouse in Sakarya, Turkey. PSWW was pre-treated with a dissolved air flotation unit.

The characterization of the Air Floated PSWW was made by using the wastewater sample taken from the air flotation unit before starting the study. The results are as shown in Table 1.

| Parameters (mg L–1) | COD  | S.Solid | PO4  | TOC  | TN  |
|---------------------|------|---------|------|------|-----|
| Initial Values      | 717.6| 22      | 1.52 | 320  | 144 |

2.4. Experimental set-up

Chlorella Vulgaris has cultivated in 300 ml Erlenmeyer flasks containing a total of 250 mL poultry wastewater (after flotation) at a shaker (Biosan, Multi shaker PSU 20) at room temperature with 160 rpm shaking and a continuous illumination, fluorescent light (intensity is 3600 lux). Microalgae mixtures were added to the total volume of 250 mL with 0.8, 4, 8, 12 and 20 % different ratios. In the studies conducted with Chlorella Vulgaris in the literature, different cultivation times such as 7, 8, 12 and 14 days were selected [1–4]. Based on these studies, cultivation time was determined as 15. Chlorella Vulgaris, the initial pH of solutions was adjusted to be 7.0 ± 0.1 with pH meter (Hanna Ins. 301) by using 1.0 M HCl and 1.0 M NaOH solutions. No external CO2 source was used for the algal cultivation other than the atmosphere. Fig 1 shows the first day of cultivation flask after microalgae addition with the different ratios. All studies were repeated three times.

Fig 1. Poultry wastewater (after dissolved air flotation) samples with different ratio microalgae addition (0.8, 4, 8, 12 and 20%)
2.5. Parameters analysis

Water samples COD analyzes were carried out according to Standard Methods 5220-D-COD by using MERCK Spectroquant Pharo-300. Suspended organic matter analyses were carried out to obtain algal growth [6]. Suspended Solids filtered by using millipore 47 mm 0.22 µm glass fibre filters [23].

3. RESULTS AND DISCUSSION

3.1. Optimum concentration of microalgae

Biomass growth can be characterized as total suspended solids in algal systems [6, 24]. Glass fibre filter papers filtered the algal solutions, and after drying, the residual solid mass refers to algal growth. The growth of the algae culture in different algal additions of AFPSWW throughout the cultivation period is illustrated in Fig. 1. During 15 days, under the same experimental conditions, biomass growth has been examined in different algal addition rates.

As can be seen in Table 1, raw AFPS wastewater included only 22 mg L\(^{-1}\) total suspended solids. The initial suspended solids amount was determined after algal addition for all ratios separately. Algal growth calculated with the formula of

\[
\text{Algal Growth (\%)} = \frac{S - S_0}{S_0} \times 100 \quad (1)
\]

\(S\); Total suspended solid matter (mg L\(^{-1}\)) after incubation, \(S_0\); Initial total suspended solid matter (different for all algal addition ratios) (mg L\(^{-1}\))

After five days of incubation, the amount of TSS was started to increase rapidly for all algal addition levels except 20%. Although TSS rise was observed for all inoculum rates, the maximum biomass growth was investigated at 12%. Fig 2 shows the algal growth levels as TSS percentages.

![Fig 2. Algal growth (%)](image)

Except for 20%, all other four algal concentrations algal growth trends were observed as similar in the log and lag phase. The further biomass content of 20% addition caused a lack of organic matter and nutrients. According to the experiment’s results, biomass growth was directly affected by the nutrient and organic substances levels. C, N or P ingredients in wastewater can be a limiting factor for algal growth [18,25-26]. Similar research was conducted for raw slaughterhouse wastewaters diluted with distilled water by Azam et al., (2020) [27], Taşgan, E. (2016) [28], and with tap water Hernández et al., (2016) [29].

3.2. COD removal

CO\(_2\) is the first inorganic carbon source for microalgae. The Absence of CO\(_2\), bicarbonate (HCO\(_3^−\)) is the second source [30]. Microalgae are autotrophic organisms that can synthesize organic matter from inorganic substances. The well-known algal cell stoichiometric formula is \(\text{C}_{106\text{H}_{180\text{O}_{67\text{N}_{16\text{P}}}}\text{O}}\). That means the optimal reproduction media should be includes with these proportions [31]. In the other research, these proportions specified as 53:15:1 (C/N/P) [32].

Considering Table 1, AFPSWW provides a suitable environment for algae nutrition under the rates specified in the literature [14, 31–34].

It should be noted that *Chlorella Vulgaris* can function under heterotrophic conditions depending on the nature of carbon availability [35]. COD concentrations were observed during microalgae treatment for 15 days. All removal efficiencies values calculated with the formula;

\[
\text{Removal Efficiency (\%)} = \frac{C_0 - C}{C_0} \times 100 \quad (2)
\]

\(C_0\); Initial COD value after algal solution addition(mg L\(^{-1}\)), \(C\); COD value after incubation (mg L\(^{-1}\))

Fig 3 shows COD removal rates trends for AFPSWW for all different inoculum rates.

![Fig 3. Removal of COD (%)](image)

The more efficient inoculum rate was observed from the inoculum rates perspective as 12% with 76% COD removal efficiency. Within 15 days, COD values were decreased from 717 to 171 mg L\(^{-1}\) by microalgae. Therefore, the poultry wastewater treatment depends on the appropriate nutrient ratio to algae cells grow up rather than the algae density in the water.

As shown in Fig 2, the increase in algae quantity is not proportional to the removal efficiency. Hongyang et al. (2011) reported 70.3% COD removal using *Chlorella Pyrenoidosa* from soybean processing wastewater [36]. Hernández et al. (2016) observed 92 % COD removal for slaughterhouse wastewater in the high exchange.
rate algal ponds with Chlamydomonas Subacaudata, Anabaena sp. and Nitzschia sp. [29]

4. CONCLUSION

According to Turkey water quality and control directive table 5.8 [5], discharge standards of slaughterhouse wastewater to the receiving environment are determined as the maximum COD values 200 mg L−1 for 2 hours of composite samples. Within this scope, the treatment with microalgae is proper for discharge to the receiving environment. On the other hand, if the produced algal biomass is considered an energy source, it is an acceptable wastewater treatment method that can bring considerable gains.

This study observed that 3.58 mg biomass could be produced per 1 mg COD removal for one litre of wastewater in the 12% algal addition ratio. In other words, a slaughterhouse facility producing 1000 m³ wastewater can be grown theoretically 35.8 kg biomass per day.

Nowadays, microalgal biomass is used for many applications such as human consumption (food, cosmetics and pharmaceuticals), agricultural aims, etc. [38]. From this perspective, an economically valuable raw material will be produced from the biomass obtained from the wastewater treatment. In this study, even if it is not sufficient for energy production in the facility where it can purchase to a suitable market. Thus, both a very harmful wastewater to nature will be treated, and the biomass sale could reduce the treatment costs.

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