Paper B36

Potential Usage of Tannic Acid in Soaking Process

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Abstract

Tannic acid (TA) is a polyphenolic compound in the structure of vegetable tanning agents and its structure is based mainly on glucose esters of gallic acid. It is well known that some plants and especially the phenolic compounds have an antimicrobial and a strong bactericide effect. Thus, tannic acid was used as a bactericide in soaking process for determining the effect on microorganisms, raw skins, and soaking effluents in this study. For this purpose, 4 different concentrations of tannic acid such as 0.1, 0.3, 0.5, and 1.0% were added to the soaking bath and total mezophilic bacteria in samples taken from the medium were counted on plate count agar by using Tube dilution plate method at the 8th and 24th hours of soaking bath. A bath with and without commercial bactericide were chosen as two blank samples and their results were compared. For determination the changes on raw skin and effluent samples, the shrinkage temperature, nitrogen and COD (Chemical Oxygen Demand) analysis were performed. The shrinkage temperature of raw skin samples were measured according to TS 4120 EN ISO 3380 method and the nitrogen content of raw skin samples were determined by Kjeldahl method. COD and total nitrogen contents of effluents were measured photometrically by using standard test kits and Merck SQ 300 device.

The total bacterial count results of 0.3% TA was found comparable with commercial biocide. But 0.5% TA showed better performance than commercial biocide on both microbial growth and effluent COD values. Consequently, tannic acid can be used as a bactericide for soaking process as an environmentally friendly alternative to the commercial biocides which most of them are harmful to environment and human health.

Keywords: tannic acid, bactericide, soaking, COD

Introduction

The attack of the microorganisms always can be considered as a threat, due to organic nature of leather, especially at soaking process considering its high microbial activity. Not only the raw hides and skins during curing, soaking, pickling processes but also the tanned leathers during wet-end processes must be protected against this attack. Also, it should be considered that the possible microbial defects could be higher depending on various factors such as preservation method, extended storage time of hides and skins, temperature and pH[1,2]. For providing limited microbial growth or preventing the hides/skins from microbial damages, biocides are used commonly in leather industry. However, most of the biocides have negative impact on environment and human health because of containing hazardous active compounds. Carbamates, benzisothiazolinon, organo bromine compounds, and oxidative agents are known as common active substances which their use in leather industry have become the focus because of the chemical safety and consumer health issues considering their toxic effect for aquatic organisms and potential heritable genetic damage. Besides, they may impair fertility, cause harm to the unborn child and long-term adverse effects in the aquatic environment[3]. Also Tisler et al. 2004 confirmed the toxicity of bactericides and fungicides in tannery effluents by determination of their extremely high acute and chronic toxic effects on bacteria, algae,

daphnids and fish[4,5]. Therefore, it shouldn't be surprising that recent studies have mainly focused on new environmentally friendly alternative biocides or methods[6,7,8,9].

Vegetable tannins, also known as plant polyphenols, are used in many areas such as leather industry and medical science due to their tanning and antimicrobial properties[6]. Tannic acid is an important gallotannin classified in hydrolysable vegetable tannins, which has a central carbohydrate (glucose) and 10 galloyl groups[10] and has antimicrobial, antimutagenic, anticarcinogenic and antioxidant activities like other polyphenols[10,11,12, 13, 14,15,16,17,18,19,20].

There is limited number of studies related to tannic acid usage in leather industry such as application of tannic acid in tanning process[21,22,23,24], the reduction of Cr (VI) formation by tannic acid[25] and use of tannic acid in leather dyeing process as a natural dye[26]. Furthermore, Çolak et al. 2010 studied the antimicrobial activity of tannic acid in pickling process against some microorganisms such as *B. cereus, A. niger, G. candidum*[27]. To our knowledge up to date there has been no reported research on the application of tannic acid as a bactericide in soaking process for leather industry.

In this study, the applicability of tannic acid as an alternative material in respect to common biocides used in soaking process were investigated to reduce the negative environmental impact of these protective chemicals.

Materials and methods

Materials

The bactericide effect of tannic acid was tested using 100 cm^2 pieces of raw metis sheepskin. The effect of tannic acid on bacterial growth was compared with commercial bactericide Gemacide LP (Gemsan, Istanbul, Turkey) according to recommended dosage levels of the manufacturer.

Methods

First, all leather samples were presoaked for 2 hours and washed for 5 minutes to remove the dirt and remained salts. Then the samples were put in 2L beakers and soaked with different concentrations of tannic acid such as 0.1, 0.3, 0.5, and 1.0 % and 0.1% of commercial bactericide Gemacide LP at 27°C in 1L baths. At 8th and 24th hours of process, samples were taken for determination of soaking bath properties and skins. A blank bath without any protective chemicals was also analyzed. In order to demonstrate the applicability of TA in soaking process, not only the effect of TA as biocide, but also its effect on soaking effluent and the composition of skin were determined.

Determination of Total Bacterial Count

The bacterial load of the soaking baths were determined with the samples taken at 8th and 24th hours of process and the total bacterial counts were obtained by use of the tube dilution plate method with Plate Count Agar (Merck). Colonies were counted after 24 hours of incubation at 37°C from appropriate dilutions.

Determination of Chemical Oxygen Demand (COD) and Total Nitrogen Content

COD values and total nitrogen (N) content of soaking effluents were measured with Merck SQ 300 Water and Wastewater Spectrophotometer by using standard kits (Merck).

Determination of Shrinkage Temperature and Total Kjeldahl Nitrogen

Shrinkage temperature of leather samples was determined in accordance with TS 4120 EN ISO 3380[28]. The total nitrogen content of skins was determined by Kjeldahl nitrogen method[29,30].

Results and discussion

Bacterial load of soaking baths were presented in Figure 1. Accordingly, the total bacterial count results of blank samples were determined as $8.5x10^4$cfu/ml and $11x10^7$cfu/ml at 8th and 24th hours of process respectively. The bacterial growth at 8th hours of soaking indicates that all the concentrations of TA wt% can prevent the microbial growth under ideal conditions. All applied TA concentrations were found significantly effective up to 35 times more than the commercial bactericide depending on the concentration of TA for 8h.

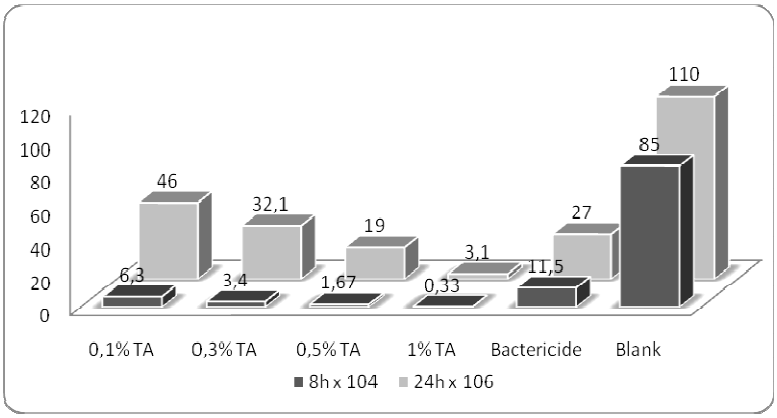

Figure 1 Total Bacterial Count at the end of 8th and 24th hours of soaking (cfu/ml)

When the results for 24h of soaking are evaluated, it can be seen that the usage of 0.3 wt% TA provided comparable results with commercial bactericide. For the same time of period, although 0.1 wt% TA gave the highest colony forming units, it has a limitation at microbial growth as much as half of the blank sample. It is obvious that the results of 0.5 and 1 wt% TA had apparently better results than $2.7x10^7$cfu/ml which was obtained by commercial bactericide.

The total nitrogen content of blank sample for 24h has the highest value among the other effluent samples. This might be due to the heavy infection in the soaking float. Owing to inhibition effect of TA and bactericide on microbial growth[31], lower N values are acquired in these effluents. Generally, all effluents contain N values in between 3-10 mg/l[32] which are the limit values specified for directly discharge into the surface water (Table 1).

The COD values of soaking effluents were determined below the previously reported values[4,33]. With the usage of 1 wt% TA instead of bactericide, the COD values of the soaking effluents were reduced nearly 20% (Table 1). On the other hand, the highest COD value reduction by 35% was determined with the use of 0.1, 0.3 and 0.5 wt% TA.

Table 1 Chemical Oxygen Demand (COD) and Total Nitrogen (N) Contents of Soaking Effluents ($mg.L^{-1}$) for 24h soaking process

	Blank	0.1% TA	0.3% TA	0.5% TA	1% TA	Bactericide
COD	1560	1457	1451	1450	1838	2265
N	3,8	3	2,9	2,7	2,5	2,9

The shrinkage temperature of the skins soaked with different concentrations of TA shows only slight differences in comparison to skin treated with bactericide and black sample (Table 2). All skin samples provided similar total Kjeldahl nitrogen values that varied between 14.5-14.9% of nitrogen.

Table 2 Shrinkage Temperature (°C) and Total Kjeldahl Nitrogen (TKN) values of skin samples (%) for 24h soaking process

	Blank	0.1% TA	0.3% TA	0.5% TA	1% TA	Bactericide
Shrinkage Temperature	53°C	55°C	55°C	56°C	57°C	55°C
TKN of skin	14.8	14.5	14.6	14.9	14.6	14.5

Thus, affirmative conclusions were obtained from the data given in Table 2 that tannic acid usage in soaking process did not cause any adverse effects in skin matrix.

Conclusions

In the study, tannic acid applicability at soaking process was investigated both from microbiological and environmental point of view. Experiments were carried out using different concentrations of tannic acid in comparison to commercial biocide at soaking process and following conclusions have been drawn.

Total bacterial count of 0.3 wt% was found comparable to commercial bactericide that indicates the growth of microorganisms could be kept under control to some extent. Besides, the 0.5 and 1 wt% treatments of TA were more effective when compared to commercial bactericide and other TA concentrations.

The reduction of COD and N content of effluents were significantly influenced by TA concentrations which actually is a good sign of its environmentally friendly structure. In addition, by showing no significant difference in between TKN values of skin, tannic acid demonstrates its potential usage as an alternative and environmentally friendly bactericide for leather industry.

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