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ORIGINAL PAPER

Biodegradability of tannin-containing wastewater from leather industry

Qiang He · Kai Yao · Danhong Sun · Bi Shi

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Abstract Tannins occur commonly in the wastewaters from forestry, plant medicine, paper and leather industries. The treatment of this kind of wastewaters, including settling and biodegradation, is usually difficult because tannins are highly soluble in water and would inhibit the growth of microorganisms in activated sludge. The objective of this study is to investigate biodegradability of tannin-containing wastewaters, so as to characterize the pollution properties of such wastewaters and provide a reference for their biological treatment in wastewater treatment plants. The research was typified by using the wastewater collected from vegetable tanning process in leather industry. A model was developed to describe the activated sludge process, and the biodegradation kinetics of vegetable tanning wastewater (VET wastewater) was studied. It was found that the biodegradability of tannin-containing wastewater varies heavily with the content of tannins in wastewater. The biodegradation of VET wastewater with tannin content around 4,900 mg/l occurred inefficiently due to the inhibition of tannins to the activated sludge process, and only 34.7% of biodegradation extent was reached in 14 days of incubation. The optimal biodegradability of VET wastewater was observed when its tannin content was diluted to 490 mg/l, where the COD and tannin removals reached 51.3% and 45.1% respectively in 6 days. Hence, it is suggested that a proper control of tannin content is necessary to achieve an effective biodegradation of tannin-containing wastewaters in wastewater treatment plants.

Keywords Tannins · Wastewater · Biodegradation · Leather industry

Q. He · D. Sun · B. Shi (✉)
The Key Laboratory of Leather Chemistry and Engineering of Ministry of Education, Sichuan University, Chengdu 610065, P.R. China
e-mail: Sibitannin@vip.163.com

K. Yao
The Department of Food Science and Engineering, Sichuan University, Chengdu 610065, P.R. China

Introduction

Vegetable tannins, the natural polyphenols with molecular weights varying from 500 to 3,000, occur commonly in the wastewaters generated from forestry, plant medicine, paper and leather industries. The presence of tannins in the wastewaters would cause many problems associated with environmental pollution and wastewater treatment. Owing to the presence of tannins, the wastewaters are usually highly colored, which is very difficulty to be eliminated by common methods like dilution and adsorption (Etiegni et al. 1999). In addition, tannins can inhibit growth of

microorganisms and therefore, are toxic to activated sludge. This negative effect can be observed when tannin-containing wastewaters are biologically treated in wastewater treatment plants (Bhat et al. 1998; Ren 2004).

Many methods have been explored to treat tannin-containing wastewaters, such as adsorption (Liao and Shi 2005; Marsal et al. 2003), membrane filtration (Cassano et al. 2003; Scholz and Lucas 2003), photocatalytic and sonochemical degradation (Arana et al. 2001; Svitelska et al. 2004). But, in practice, tannin-containing wastewaters are usually successively treated by chemical settling and biodegradation so as to meet the discharging specifications. Tannins are highly soluble in water. So, an extra high dose of metal coagulant is needed to obtain considerable effectiveness of settling, which is accompanied by introduction of metal pollutions (Rao et al. 2003). Meanwhile, the metals introduced into wastewater would negatively influence the subsequent biological treatment. Therefore, a direct biological treatment of tannin-containing wastewater, without pretreatment of settling, might be more reasonable in wastewater treatment plants. As mentioned above, tannins are able to inhibit growth of microorganisms in general. But it has been found that some microbes are resistant to tannins and can grow with tannins as carbon and energy source (Bhat et al. 1998) in proper conditions. This means it is possible to develop an acceptable degradation extent by optimizing biodegradation conditions.

The purpose of this research is to investigate the biodegradability of tannin-containing wastewaters in consideration of practical application in wastewater treatment plants. The research is typified by using a wastewater collected from vegetable tanning process in leather industry. In the process, vegetable tannin extracts are used as tanning agents to convert skins into leather in aqueous solution. To ensure full penetration and reaction of tannins with skin collagen, an excess of vegetable tannin extracts is usually used in tanneries (Madhan et al. 2005; Gao and Shu 2001). So, vegetable tanning wastewater (VET wastewater) is characterized by high contents of BOD, COD, total solids (TS) and tannins.

There are many reports concerning the biodegradation of phenol-containing wastewaters. Most of them aimed at simulated wastewater containing a few of simple phenol components (Huang et al. 2005). This kind of wastewaters cannot be employed to elucidate the environmental impact of tannin-containing wastewaters that included complex polyphenolic components, and might be unsuitable to be used as a reference of biodegradability of tannin-containing wastewaters in treating plants. Therefore, the biodegradation of VET wastewater, as a typical tannin-containing wastewater, was investigated in this paper.

Materials and methods

Materials and general methods

Five pickled pigskins were used for vegetable tanning. They were the in-process pelts undergoing common pretreatments of leather-making, such as soaking, degreasing, liming, deliming and pickling. The pickled skins were kept in pickle liquor (pH 2.5, NaCl content 8%). The vegetable tanning agents employed included bayberry, larch, valonea and black wattle tannin extracts, and they were all commercial products. Other chemicals were of commercial grade.

Tannin content in wastewater samples was measured by the method of Makkar et al. (1993). TS and suspended solids (SS) in wastewater were determined by using standard methods (Xi et al. 2001). Measurements of COD and BOD were carried out by using the determination system HANNA HI 99721 and HI 99724A-6 (Italy), respectively.

Vegetable tanning and collection of wastewater

To collect VET wastewater for biodegradation tests, a conventional vegetable tanning procedure was conducted on the basis of quality maintenance of leathers. Five pickled skins were weighed and the amounts of chemicals used in vegetable tanning were based on wet weight of pickled skins. The skins was put into a drum containing 20% pickle liquor and vegetable

tanning was carried out according to procedure listed in Table 1.

The residual solution and the water from washing operation were collected and mixed. The mixture, termed as VET wastewater, was used for biodegradation tests.

Biodegradation tests of VET wastewater

The BOD determination system was used for biodegradation tests. It can continuously measure oxygen consumption during the period of incubation based on pressure change in a hermetically closed bottle containing wastewater sample. The wastewater samples were all adjusted to pH 7 before test.

The activated sludge was collected from wastewater treatment laboratory of a local tannery (Chengdu, China). One liter of the activated sludge (SS: 2.6 g/l, pH 8.2) was mixed with 1 l water. This activated sludge solution was employed as inoculum and it was continuously aerated until use. In experiments, 20 ml of the activated sludge solution was transferred into the determining bottle of BOD measurement system. The determining bottles were incubated at 20°C with magnetic stirring. In a batch of oxygen consumption measurements, a blank test which contained only the inoculum and distilled water was run. The oxygen consumption measurement of wastewater was corrected by the value of black test that is endogenous respiration of activated sludge. The biodegradation extent of wastewater was expressed as oxygen consumption versus original COD.

The changes of COD and tannin content of wastewater during biodegradation process were measured by using the same method as above with the exception that the determining bottles were opened during incubating period. At the indicated incubating interval, 5 ml of wastewater was sampled for the determination of COD and tannin contents.

Results and discussion

The characteristics of VET wastewater

The contents of pollutants in VET wastewater are shown in Table 2. The SS content (325 mg/l) is much lower than the TS content (11,440 mg/l), which indicates that the solid substances in VET wastewater are mainly dissolved. This might be the reason for the low efficiency of a settling attempt of VET wastewater. In the attempt, aluminum salts were employed as coagulants to decrease the contents of pollutants in VET wastewater, but the settling occurred inefficiently due to the fact that the coagulation process is largely dependent on the SS concentration of wastewater (Newman et al. 1990).

The value of BOD_5/COD is commonly adopted to evaluate biodegrading property of wastewater, although the value is not always in agreement with the biodegradability in some special situations (Guo and Wu 1998; Hufschmid et al. 2003). In general, a higher BOD_5/COD value of a wastewater implies a higher degree of biodegradability. When the value is lower than 0.20, the wastewater is usually considered as hardly biodegradable one. From this point of view, it could be deduced that VET wastewater, whose BOD_5/COD value is 0.236 (as shown in Table 2), is not easy to be biodegraded. The difficulty of biodegradation of

Table 1 Vegetable tanning processes

Process	Chemicals used	Operation
Pretreatment	2% Na_2SO_3	Run drum for 0.5 h
Tanning	8% bayberry tannin extract	Run drum for 4 h
	8% larch tannin extract	Run drum for 10 h
	8% valonea tannin extract	Run drum for 10 h
	5% black wattle tannin extract and 180% H_2O	Run drum for 20 min each hour for a period of 8 h, and still for 12 h
Washing		Remove residual solution and wash leather with 200% H_2O

Table 2 The characteristics of VET wastewater

TS (mg/l)	11,440
SS (mg/l)	325
Tannins (mg/l)	4,900
pH	4.3
BOD_5 (mg/l)	2,100
COD (mg/l)	8,900
BOD_5/COD	0.236

VET wastewater should result from its high content of tannins (4,900 mg/l), due to the fact that tannins are inhibitor to microorganisms (Scalbert 1991).

The observations above illustrated typical characteristics of tannin-containing wastewaters which usually have poor settling and biodegrading properties in wastewater treatment plants. Hence, tannin-containing wastewaters have gained considerable attention and many technologies have been explored to develop efficient treating methods for the wastewaters. However, most of the explorations are at the stage of laboratory study, and activated sludge process has been so far playing an important role in wastewater treatment plants to remove the organic components of tannin-containing wastewaters.

Biodegradation of VET wastewater

Although vegetable tannins would inhibit the growth of microorganisms, some strains, such as fungi present in activated sludge, are resistant to tannins and are able to grow with tannins as sole carbon and energy source (Bhat et al. 1998; Huang et al. 2005). In addition, vegetable tannin extracts usually contain 20–30% of non-tannin components such as sugars. These components do not react with collagen during vegetable tanning process and therefore remain in VET wastewater. They can offer carbon and energy source for growth of microorganisms. Therefore, the biodegradation of VET wastewater by activated sludge could take place, and the results of degrading process are shown in Fig. 1.

Little oxygen consumption was observed during the first 2 days, implying that the biodegradation of the tannin-containing wastewater was retarded. In 2 days of incubation, the biodegradation extent only reached 4.7% due to the inhibition of tannins to the activated sludge process. Afterwards, the microorganisms present in activated sludge adapted to the tannin environment and the oxygen consumption of VET wastewater increased fast. Consequently, the fast degrading of VET wastewater was observed during the logarithmic growth phase of microorganisms. The biodegradation extent of VET wastewater reached 34.7% in 14 days of incubation, suggesting a possible treating extent of tannin-containing wastewaters when they were directly biodegraded in wastewater treatment plants without any pretreatment.

It was reported that tannins at levels of 300–3,000 mg/l can considerably inhibit the growth of many bacteria (Scalbert 1991). However, the activated sludge process achieved 34.7% of biodegradation of VET wastewater whose tannin content is 4,900 mg/l. This should be mainly attributed to the biodegradation of non-tannin components in the wastewater. That is, the non-tannin components existing in tannin-containing wastewaters could be first degraded during the activated sludge process even at a high content of tannins. This elucidation can be further confirmed in following observations.

Effects of diluting on biodegradability of VET wastewater

Although activated sludge process achieved 34.7% of degradation of VET wastewater in 14 days, the measures to increase degrading efficiency was further explored. An interesting phenomenon which had been observed is that vegetable tannins would inhibit growth of many microorganisms when they present at high concentration, but they could be used as carbon and energy source by the microorganisms at low concentration (Shi and Di 2000). That is, the biotreatability of tannin-containing wastewaters is closely related to their tannin content. In practice, tannin-containing wastewaters are often mixed with other wastewaters in wastewater treatment plants, and consequently the tannin concentration can be decreased to a large extent. To model this situation, VET wastewater was diluted by water and the effect of dilution treatment on biodegradability of VET wastewater

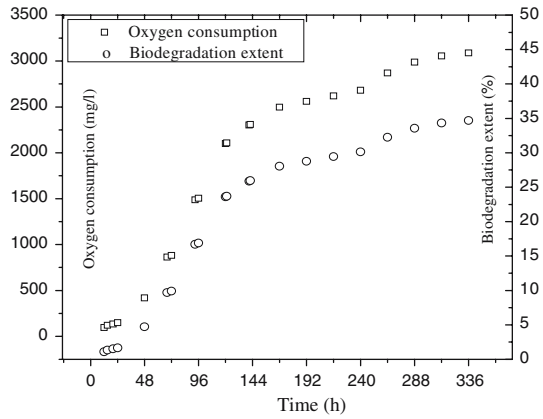

Fig. 1 Biodegradation of VET wastewater [endogenous respiration: Y (oxygen consumption, mg/l) = 0.7962 X(time, h) + 92.678, $R^2 = 0.9861$]

was investigated. The results could be employed to evaluate the practical biotreatability of tannin-containing wastewater in wastewater treatment plants.

Table 3 shows that the BOD_5/COD value of VET wastewater increases with increasing degree of dilution. This means a higher dilution ratio would lead to a better biodegradability based on the evaluation of BOD_5/COD value. But, as we know, the kinetic process of biodegradation is often more concerned in practice. Therefore, the kinetic behaviors of biodegradation of the wastewater were further investigated for comprehensive evaluation of its biodegradability. For this purpose, a model for describing the activated sludge process was developed to study the aerobic biodegradation of VET wastewater.

During activated sludge process, wastewater could be considered as a single substrate and the process as a monomolecular reaction (Mao and Smith 1995). Then, the disappearing rate of biochemical-oxygen-consuming substances in the wastewater can be derived as:

$$\frac{dL}{dt} = -kL \quad \text{or} \quad L = L_0 \exp[-k(t - t_0)]$$

where L_0 is the total amount of biochemical-oxygen-consuming substances in wastewater (equivalent to ultimate oxygen consumption), k is kinetic constant, t_0 is lag time, L is the biochemical-oxygen-consuming substances remaining in wastewater. Then, at time t, the amount of degraded biochemical-oxygen-consuming substances y_t (equivalent to exogenous oxygen consumption) can be expressed as:

$$y_t = L_0 - L_0 \exp[-k(t - t_0)]$$

The exogenous oxygen consumption of activated sludge can be calculated by subtracting endogenous oxygen consumption from the total oxygen consumptions. Then, the biodegradability of wastewater is described by three parameters, the ultimate oxygen consumption L_0, the kinetic constant k and the lag time t_0.

The exogenous oxygen consumptions of VET wastewater samples are shown in Fig. 2. There is a decrease of oxygen consumption with diluting treatment due to the decrease of content of biodegradable components in VET wastewater. Based on the observations, the data of exogenous oxygen consumption as a function of time were analyzed by the model above by using non-linear regression analysis method. The estimated first-order kinetic parameters of aerobic biodegradation of VET wastewater samples are presented in Table 4.

The lag time (t_0) of biodegradation of VET wastewater was shortened when dilution degree was increased, which indicates an improvement of adaptation of activated sludge to the wastewater. This is due to the decrease of tannin content in VET wastewater and consequently the inhibition of tannins to activated sludge is weakened. The lag time was 5.9 h when VET wastewater was diluted in 1:9, but no significant shortening of lag time was observed with further increase of

Table 3 Tannin contents and BOD_5/COD values of VET wastewater samples

Dilution (v:v)	1:0	1:0.3	1:3	1:6	1:9	1:14	1:19
Tannins (mg/l)	4,900	3,750	1,225	700	490	320	245
BOD_5 (mg/l)	2,100	1,712	678	475	407	291	237
COD (mg/l)	8,900	6,850	2,225	1,300	890	593	445
BOD_5/COD	0.236	0.25	0.305	0.365	0.457	0.491	0.533

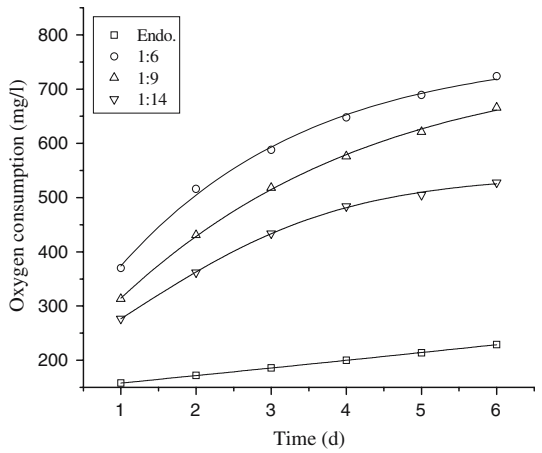

Fig. 2 The oxygen consumption curves of VET wastewater at different dilution ratio (v:v) [endogenous respiration: Y(oxygen consumption, mg/l) = 0.7962X(time, h) + 92.678, $R^2 = 0.9861$]

Table 4 The estimated biodegradation kinetic parameters of VET wastewater

Dilution (v:v)	1:6	1:9	1:14
t_0 (h)	9.6	5.9	5.4
k (day^{-1})	0.61	0.71	0.63
L_0 (mg/l)	496	441	296
L_0/COD (%)	38.15	49.55	51.92

dilution degree. Thus, it can be suggested that there is almost no inhibition of tannins to the biodegradation of VET wastewater when the tannin content in the wastewater is lower than 490 mg/l.

The ultimate oxygen consumption (L_0) is related to the concentration of biochemical-oxygen-consuming substances in wastewater. So a decrease of L_0 was observed with dilution treatment of VET wastewater. Meanwhile, with the decrease of tannin content, tannins in wastewater would gradually act as a carbon and energy source rather than as an inhibitor for the growth of microorganisms during the biodegradation process. This is demonstrated by the increase of ultimate biodegradation extent (L_0/COD) of VET wastewater when wastewater is diluted, as shown in Table 4.

The biodegrading kinetic constant (k) of tannin-containing wastewater is related to contents of tannins and other biodegradable components. The constant can be used to reflect the biologically treating efficiency of tannin-containing wastewater. It increased first and then decreased with the increase of dilution extent, and a maximum k value (0.71 day^{-1}) was observed when the wastewater was diluted in 1:9. This might be due to the fact that, at this dilution ratio, the inhibition of tannins to biodegradation of VET wastewater is very limited, meanwhile the content of biodegradable components in the wastewater remains at a level enough to keep a fast growth of microorganisms. Therefore, based on the results above, it could be concluded that a proper control of tannin content in wastewater is necessary for its biodegradation, and that the tannin content of 490 mg/l is suitable for achieving an efficient biological treatment of such wastewaters.

Removals of COD and tannins during biodegrading process of VET wastewater

Tannins could act as an inhibitor at high concentration, but they would be converted into biodegradable substrates at low concentration during the biodegrading processes of tannin-containing wastewaters. To confirm this conversion, the removals of tannins and COD during the biodegrading process of VET wastewater were investigated.

The removal extents of tannins and COD of VET wastewater samples increased fast in the first day of incubation, as shown in Fig. 3. The fast removal of tannins and COD might result from two different effects, namely physical absorption and biodegradation of organic compounds by activated sludge. Moreover, the physical absorption might play a major role in the first day (Gebara 1999; Guellil et al. 2001). Afterward, the removal extents of COD and tannins of the two wastewater samples increased gradually, but presented in different behaviors because of different tannin content.

For the wastewater sample diluted in 1:6, the removal extent of COD increased remarkably and reached 39.2% in 6 days of incubation. Meanwhile no substantial increase of tannin removal extent was observed and the removal extent of tannins was only 17.8% in 6 days.

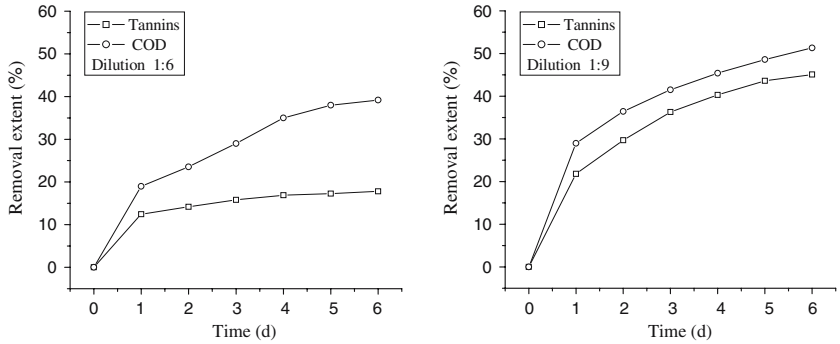

Fig. 3 Removals of COD and tannins during the biodegrading process of VET wastewater samples

Therefore, it could be deduced that the removal of COD should be less attributed to the removal of tannins but mainly to the biodegradation of non-tannin components in the wastewater. This is consistent with our previous discussion.

For the wastewater sample diluted in 1:9, the removal extents of COD and tannins increased analogously, and reached 51.3% and 45.1% respectively in 6 days of incubation. In comparison with the biodegradation of the wastewater sample diluted in 1:6, the biodegradation of the wastewater sample diluted in 1:9 achieved remarkably higher removal extents of COD and tannins in same period of incubation. Hence, it can be suggested that tannins would no longer act as an inhibitor but as a readily biodegradable substrate when diluted to a proper concentration. That is, tannin-containing wastewaters might exhibit improved biotreatability in wastewater treatment plants when the content of tannins is controlled to the range around 490 mg/l.

Biodegradation is actually the main treating method of tannin-containing wastewater in effluent treating yard of tannery. Our researches indicated that the presence of tannins could inhibit the biodegrading process or even show toxic effects on activated sludge when they appear at a high concentration. On the other hand, they could be employed as carbon and energy sources by activated sludge microorganisms at low concentration. Therefore, with the recognition that excessive dilution will largely increase the treating magnitude, a proper control of tannin content in effluent is significant for depolluting plants to dispose tannin-containing wastewater with high efficiency and low costs.

Conclusions

Tannin-containing wastewaters, such as VET wastewater, are characterized by high contents of COD, BOD, TS and tannins. This kind of wastewaters usually exhibits poor settling and biodegrading properties in wastewater treatment plants. The results of this research indicate that a dilution pretreatment is necessary to improve the biotreatability of tannin-containing wastewaters. For VET wastewater, whose tannin content is around 4,900 mg/l, the optimal biodegradability can be obtained when its tannin content is diluted to 490 mg/l. At this content, tannins can be biodegraded and the inhibition of tannins to activated sludge is negligible during the biodegrading processes.

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