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(54) Title: ENZYMATIC-ASSISTED CHLORINE DIOXIDE TREATMENT

(57) Abstract: The present invention provides methods and compositions for chlorine dioxide delignification and/or bleaching processes using exo-acting xylanases and/or exo-acting mannanases.



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## **Enzymatic-Assisted Chlorine Dioxide Treatment**

### **FIELD OF THE INVENTION**

**[0001]** The present invention relates to methods and compositions for improving chlorine dioxide treatment processes, such as, pulp delignification and bleaching processes, using an exo-acting xylanase and/or exo-acting mannanase.

### **BACKGROUND**

**[0002]** Chlorine dioxide is one of the most widely used delignification/bleaching agents in the pulp and paper industry, providing a high-quality, low-cost delignification and bleaching process. Chlorine dioxide treatment is superior to chlorine bleaching processes in that it virtually eliminates all dioxin discharges into the environment, and has accordingly, helped pulp and paper manufactures to employ environmentally friendly processes and to meet environmental requirements. Accordingly, the use of chlorine dioxide treatment is increasing and most pulp and paper mills now have at least one chlorine dioxide delignification or bleaching stage. Chlorine dioxide treatment has also been used to treat wastewater, sludge and other process streams.

**[0003]** During the chlorine dioxide treatment processes some of the chlorine dioxide is converted to chlorate and chlorite, which decreases the efficiency of the chlorine dioxide treatment. Methods have been proposed to improve the efficiency of the chlorine dioxide treatment process by reducing chlorate and chlorite formation. Seger et al., Chiang, Tappi J., 1992, 75(7):174-180, for example, discloses a two step high-pH and low-pH process, which is believed to reduce the formation of chlorate at the higher pH and chlorite becomes reactive in the low-pH step. Joncourt et al., International Symp. Wood Pulping Chemistry, Montreal, Jun. 9-12, 1997, discloses the use of iron to regenerate chlorine dioxide from chlorite. Jiang et al, U.S. Patent No. 6,235,154, discloses process for improving chlorine dioxide delignification or bleaching by using formaldehyde to regenerate chlorine dioxide from the chlorite.

**[0004]** In the context of disinfectant solutions, Roozdar et al., U.S. Patent No. 5,380,518, discloses a process to generate chlorine dioxide from chlorite by using an hydroxyl-free aldehyde, e.g., acetaldehyde, glutaraldehyde, cinnamic aldehyde. Davidson et al., U.S. Patent No. 4,986,990, discloses a process to generate chlorine dioxide for use in a disinfectant solution using the sugars, such as, mannose, glucose, glycerin and inositol.

**[0005]** In the pulp and paper industry, xylanases and other hemicellulases have been used to improve the bleachability of the pulp by degrading the xylans and other hemicullose materials. The aim of Kraft pulp bleaching is to remove the residual lignin that is left in pulp

after Kraft cooking. Xylanase treatment of Kraft pulp partially hydrolyses the xylan in Kraft pulp, which makes the pulp structure more porous and enables more efficient removal of lignin fragments in the subsequent bleaching and extraction stages. The enzymatic degradation of xylan requires the use of endo-xylanases, enzymes, which randomly split and solubilize the xylan polymer. Xylanase treatment has been reported to be useful in conjunction with chlorine dioxide bleaching and other bleaching processes. See Viikari, L. et al., FEMS Microbiol. Rev. 13: 335-350 (1994); Viikari, L. et al., in: Saddler, J. N., ed., Bioconversion of Forest and Agricultural Plant Residues, C-A-B International (1993), pp. 131-182; Grant, R., Pulp and Paper Int. (September 1993), pp.56-57; Senior & Hamilton, J. Pulp & Paper :111-114 (September 1992); Bajpai & Bajpai, Process Biochem. 27:319-325 (1992); Onysko, A., Biotech. Adv. 11:179-198 (1993); and Viikari, L. et al., J. Paper and Timber 73:384-389 (1991).

**[0006]** New compositions and methods are needed to improve the efficiency and effectiveness of chlorine dioxide treatment, including, chlorine dioxide delignification and bleaching processes.

#### **SUMMARY OF THE INVENTION**

**[0007]** The present invention provides methods and compositions for chlorine dioxide delignification and/or bleaching processes. In accordance with one aspect of the present invention, an exo-acting xylanase is added to a chlorine dioxide delignification and/or bleaching process. In accordance with another aspect of the present invention, an exo-acting mannanase is added to a chlorine dioxide delignification and/or bleaching process. In yet another aspect of the present invention, an exo-acting xylanase and an exo-acting mannanase may also be added in combination to improve a chlorine dioxide delignification and/or bleaching process.

**[0008]** The addition of an exo-acting xylanase and/or an exo-acting mannanase to a chlorine dioxide treating composition is believed to result in the regeneration of chlorine dioxide through the generation of xylose and mannose, respectively, or other short chain xylan or manno-oligomers from the pulp substrate, resulting in improved delignification and/or brightening during bleaching of pulp. In particular, the resulting short chain oligosaccharides promote the regeneration of chlorine dioxide from chlorite. Exo-acting xylanase and/or an exo-acting mannanase predominantly produce shorter chain oligosaccharides, such as, xylose or mannose, and thus differ from endo-acting xylanases and endo-acting mannanases, which hydrolyze xylans and mannans, respectively, and predominantly produce longer chain xylano-oligosaccharides and manno-oligosaccharides.

#### **DETAILED DESCRIPTION**

**[0009]** An "exo-acting xylanase" or "exoxylanase" is a xylanase which predominantly hydrolyzes 1,4-beta-D-xylans from the end of a xylan chain. Examples of exo-acting xylanase, are known in the art and include the exo-acting xylanase IV and the exo-acting xylanase V from *Aeromonas caviae* (as described by Kubata et al., "Xylanase IV, an Exoxylanase of *Aeromonas caviae* ME-1 Which Produces Xylotetraose as the Only Low-Molecular Weight Oligosaccharide From Xylan, App. Environ Microb., Apr. 1995, p. 1666-1668, Vol. 61, No. 4 and Kubata et al., "Purification and Characterization of *Aeromonas caviae* ME 1 Xylanase V, which produces exclusively xylobiose from xylan", App. Environ Microb., Feb. 1994, 60 (2): 531-535, and Usui et al., "XynX, A Possible Exo-xylanase of *Aeromonas caviae* ME-1 that Produces Exclusively Xylobiose and Xylotetraose From Xylan," Biosci. Biotech and Biochem, 63(8): 1346-1352, Aug. 1999); the exo-acting xylanase from *Trichoderma reesei* (as described by Tenkanen et al., "A Novel Exo-Acting Xylanase XYN IV From *Trichoderma reesei* RUT C30, Third European Symposium Sept. 25-27 2002).

**[00010]** An "exo-acting mannanase" or "exomannanase" is a mannanase which predominantly hydrolyzes terminal, beta-D-mannose residues from the end of a mannan chain. An examples of an exo-acting mannanase is the exo-beta-mannanase (1,4-beta-D-mannan mannobiohydrolase) isolated from the culture fluid of strain No. F-25 of *Aeromonas hydrophila* (Araki et al., Biochem, (1982), Apr. 91(4):1181-6) and the exo-acting mannanase from *Cyamopsis tetragonolobus* (McCleary, B.V., (1988), Methods in Enzymology, 160, 589-595.

**[00011]** A "chlorine dioxide treatment" means any chloride dioxide treatment process, such as, for example, chlorine dioxide treatment stages used in the pulp and paper mill. Typically chlorine dioxide treatment is applied in a pulp and paper mills in delignification and pulp bleaching processes. Any suitable pulp may be treated, although preferably, the pulp is a lignocellulosic pulp, such as, a Kraft or sulfite pulp. The pulp may be treated with other delignification and/or bleaching agents prior to the chlorine dioxide treatment or during or following the chlorine dioxide treatment, such as, e.g., oxygen delignification, peroxide treatment, and enzyme treatment processes.

**[00012]** The chlorine dioxide used in the treatment process may be generated by any suitable method. However, because chlorine dioxide is unstable as a gas and can only stored as a solution, it is usually generated on-site, e.g., at the pulp mill. Once in solution, however, chlorine dioxide is fairly stable.

**[00013]** Chlorine dioxide is generally added in amounts effective to treat the pulp or process waters containing lignocellulosic materials (e.g., waste water), as are known in the art. Typically, chlorine dioxide treatment of pulp is carried out at a temperature from about 40 to 80°C for a period of about 15 to 120 min. The effectiveness of the chlorine dioxide

depends in part on pH, and is maximized at a pH of about 2 to 4. Because the pH of pulp streams and other process waters are typically more basic, acid may be added to the treatment water to reduce the pH. In some processes, the pH of the process water may be controlled by applying excess amounts of chlorine dioxide.

**[00014]** The exo-acting mannanase and/or exo-acting xylanase treatment of the present invention may be added prior to the chlorine dioxide treatment, during the chlorine dioxide treatment, or after to the chlorine dioxide treatment, or a combination thereof. If the acting mannanase and/or exo-acting xylanase are added prior to the chlorine dioxide treatment, the resulting xylose or mannose can improve the subsequent chlorine dioxide treatment. If the acting mannanase and/or exo-acting are added during the chlorine dioxide treatment, they are preferably added when the chlorine dioxide is exhausted or near exhaustion so as to prevent deactivation of the xylanase or mannanase. If the exo acting mannanase and/or exo-acting xylanase are added after the chlorine dioxide treatment, they are preferably added before washing.

**[00015]** The exo-acting mannanase and/or exo-acting xylanase are added in amounts effective to improve the chlorine dioxide bleaching/delignification. Examples of suitable amounts of xylanase or mannanase include 0.05 to 10 kg/ton, preferably, 0.1 to 5 kg/ton, more preferably 0.2 to 3 kg/ton.

**[00016]** The xylanase and/or mannanase treatment temperature is preferably between 30°C to 100°C, more preferably, 40°C to 80°C, and even more preferably, 45°C to 75°C. The pH of the composition may be from pH 1.5 to 11, preferably from 2 to 10, more preferably, 3 to 9

**[00017]** In addition to the exo-acting xylanase and/or exo-acting mannanase, a xylosidase and/or mannosidase may also be added prior to, during or after the chlorine dioxide treatment, which may further assist in the production of xylose, mannose or sugar oligomers.

**[00018]** Although the invention herein has been described with reference to particular embodiments, it is to be understood that these embodiments are merely illustrative of the principles and applications of the present invention. It is therefore to be understood that numerous modifications may be made to the illustrative embodiments and that other arrangements may be devised without departing from the spirit and scope of the present invention as defined by the appended claims.

#### **Example 1:**

Dissolve 113 mg of NaClO<sub>2</sub> in 100 mL of DI water. Aliquot 10mL of the NaClO<sub>2</sub> solution to different test tubes. Add 1mL of 100mM of sugar solutions to each tube. Adjust the pH with

acetic acid to 3.5.  $\text{ClO}_2$  formation was detected by UV absorbency at 360 nm at various incubation time intervals. It is evident that all of the sugars can generate  $\text{ClO}_2$  to some extent under the conditions used in this experiment, but xylose is the most effective one at relative short reaction time.

Table 1. Chlorine Dioxide Generation by Sugars

| No. | Sample    | Absorbency at 360 nm<br>1 hr | Absorbency at 360 nm<br>20 hr<br>(5 times dilution) |
|-----|-----------|------------------------------|-----------------------------------------------------|
| 1   | Control   | 0.070                        | 0.064                                               |
| 2   | Xylose    | 1.487                        | 0.901                                               |
| 3   | Mannose   | 0.670                        | 0.904                                               |
| 4   | Galactose | 1.197                        | 0.917                                               |
| 5   | Arabinose | 1.303                        | 0.767                                               |
| 6   | Glucose   | 0.070                        | 0.476                                               |

**Example 2:**

Add 5 g (o.d. dry) of unbleached kraft pulp to each beaker and dilute it to about 5% consistency. Add 10 mL of 11.3 g/L of  $\text{NaClO}_2$  to each beaker. Adjust the pulp to pH 3.5 by 2N  $\text{H}_2\text{SO}_4$ . Add various amount of xylose to the solution and incubate the beaker at 60°C for 1hr. After bleaching, rinse the pulp with DI water and make handsheets for brightness testing. Brightness was tested according to Tappi standard (T452). As illustrated in Table 2, it is clear that the xylose generated chlorine dioxide significantly improved pulp brightness.

Table 2. Bleaching kraft Pulp by Xylose-Generated Chlorine Dioxide

| Sample No. | % Xylose on Pulp | Brightness |
|------------|------------------|------------|
| 1          | 0                | 50.60      |
| 2          | 0.06             | 52.70      |
| 3          | 0.15             | 53.77      |
| 4          | 0.6              | 54.11      |
| 5          | 1.5              | 57.71      |
| 6          | 3                | 58.16      |

**Example 3:**

Add 5 g (o.d. dry) of unbleached kraft pulp to each beaker and dilute it to about 5%

consistency. Add 10 mL of 11.3 g/L of  $\text{NaClO}_2$  to each beaker. Adjust the pulp to pH 3.5 by 2N  $\text{H}_2\text{SO}_4$ . Add 1kg/ton of xylanase to the solution and incubate the beaker at 60°C for 1hr. After bleaching, rinse the pulp with DI water and make handsheets for brightness testing. Brightness was tested according to Tappi standard (T452). It is clear the in situ generated chlorine dioxide by xylanase treatment significantly improved pulp brightness. Xylanase treatment and chlorine dioxide bleaching can be effectively carried out in either in a single stage or sequentially.

Table 3. Simultaneous Xylanase Treatment and Chlorine Dioxide Bleaching.

| Sample No. | % Xylose on Pulp | Brightness |
|------------|------------------|------------|
| 1          | Control          | 49.89      |
| 2          | Pulpzyme HC      | 50.50      |
| 3          | Shearzyme        | 51.21      |

**CLAIMS:**

1. A method for delignifying and/bleaching pulp comprising a chlorine dioxide treatment step, comprising adding an exo-acting xylanase to a pulp prior to, during or after the chlorine dioxide treatment step.
2. The method of claim 1, wherein the pulp is a Kraft pulp.
3. The method of claim 1, wherein the pulp is a sulfite pulp.
4. The method of claim 1, further comprising adding an exo-acting mannanase to the pulp prior to, during or after the chlorine dioxide treatment step.
5. The method of claim 1, comprising adding an exo-acting xylanase to a pulp prior to the chlorine dioxide treatment step.
6. The method of claim 1, comprising adding an exo-acting xylanase to a pulp during the chlorine dioxide treatment step.
7. The method of claim 1, comprising adding an exo-acting xylanase to a pulp after the chlorine dioxide treatment step.
8. The method of claim 1, further comprising adding a xylosidase and/or mannosidase prior to, during or after the chlorine dioxide treatment.
9. A method for delignifying and/bleaching pulp comprising a chlorine dioxide treatment step, comprising adding an exo-acting mannanase to a pulp prior to or during the chlorine dioxide treatment step.
10. The method of claim 9, wherein the pulp is a Kraft pulp.
11. The method of claim 9, wherein the pulp is a sulfite pulp.
12. The method of claim 9, further comprising adding an exo-acting xylanase to the pulp prior to or during the chlorine dioxide treatment step.

13. The method of claim 9, comprising adding an exo-acting mannanase to a pulp prior to the chlorine dioxide treatment step.
14. The method of claim 9, comprising adding an exo-acting mannanase to a pulp during the chlorine dioxide treatment step.
14. The method of claim 9, comprising adding an exo-acting mannanase to a pulp after the chlorine dioxide treatment step.
15. The method of claim 9, further comprising adding a xylosidase and/or mannosidase prior to, during or after the chlorine dioxide treatment.