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A green procedure using ozone for Cleaning-in-Place in the beverage industry

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HIGHLIGHTS

- Efficiency of typical cleanings was improved by changing the gasket material.
- Ozone dramatically improved cleaning efficiency comparing to typical cleaning.
- Combination of silicone gasket and ozone resulted in the most effective cleaning.

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ABSTRACT

Cleaning-in-Place (CIP) in the beverage industry is typically carried out in production lines with alkaline and acidic solutions with detergents. This cleaning not only produces alkaline and acidic wastewater with detergents but also takes significant time. One of the important targets for CIP is adsorbed odorous compounds on gaskets, hence, we have tried to establish a rapid and green CIP process to remove traces of such compounds, especially *d*-limonene, an odorous component of orange juice, using two approaches; an ozone cleaning method and a change of gasket material from ethylene propylene diene monomer (EPDM) rubber to silicone rubber. By changing the gasket material from EPDM rubber to silicone rubber, the removability of *d*-limonene by typical alkaline and acidic cleanings with detergents was improved. However, complete removal of 4 mg g^{-1} of *d*-limonene on both EPDM and silicone gaskets could not be achieved even using a series of conventional cleaning procedures that included alkaline and acidic cleaning for 220 min. Ozone treatment dramatically improved the removability of *d*-limonene, removing 87% from the EPDM gasket at 60 min and 100% from the silicone gasket at 30 min. The combination of the silicone gasket and ozone treatment resulted in the most effective cleaning. The main removal mechanism for ozone treatment was confirmed to be oxidation by molecular ozone. Effectiveness of changing the gasket material from EPDM rubber to silicone rubber in reducing residual amounts of odorous compounds adsorbed on the gaskets was also confirmed for furfural and 4-vinylguaiacol.

1. Introduction

In recent years the beverage industry has seen a steady increase in non-mass and jobbing production as opposed to mass production. In this production style, typically one production line is used to produce multiple products. Water and time are required for the cleaning and disinfection of the production line and filling equipment to prevent cross-contamination before changing to a different product. Frequent automatic cleaning, termed Cleaning-in-Place (CIP), is, therefore, applied. CIP normally consists of cold or hot water flushing, alkaline cleaning with detergents, acidic cleaning with detergents and disinfection by chemical disinfectants such as sodium hypochlorite (Eide et al., 2003). Water flushing is conducted to remove residues in pipes. However, many deposits cannot be removed by water alone or removal takes too long. Therefore, cleaning chemicals based on acidic or alkaline solutions are used to modify the deposit into a removable form (Fryer and Asteriadou, 2009). Alkaline solutions can dissolve mainly fats and proteins whereas acidic solutions can remove minerals. The detergents in both alkaline and acidic solutions enhance the solubility of adsorbed organic compounds and facilitate their desorption.

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Minimization of the environmental impact of CIP is important. Cleaning with alkaline or acidic solution can generate large volumes of wastewater that may be of very high or low pH and have high COD due to both deposits and cleaning chemicals. However, use of cleaning chemicals is necessary to shorten the time for CIP.

Appropriate management of industrial wastewater involves not only in-plant control alternatives such as waste minimization, reuse, water use reduction but the application of end-of-pipe treatment (Oktay et al., 2007). Although a management strategy sometimes skips the first stage for the financial outlay, in-plant control applications have a potential to reduce total wastewater management cost by the reduction of pollutant loading to be treated (Erdogan et al., 2004). Innovative in-plant control technologies should be developed to minimize water use and chemicals for CIP.

One of the important targets and significant challenges for CIP is odor. Contamination by trace odours from previous products would degrade production quality. For example, lipophilic aroma compounds such as *d*-limonene and methyl butanoate from orange juice (Arena et al., 2006; Qiao et al., 2007), 1-hexanol from apple juice (Elss et al., 2006) and 4-vinylguaiacol from coffee (Mayer et al., 2000) can be adsorbed on pipe surfaces and especially on rubber of gaskets, and be resistant to cleaning.

The challenge for innovative and efficient CIP is to use ozone rather than cleaning chemicals and not to use alkaline and acidic solutions. Ozone is a powerful oxidant and is frequently used for the decomposition and removal of organic compounds, especially odorous compounds, in air and water (Lalezary et al., 1986; Glaze et al., 1990). Moreover, ozonized water has been used in the semiconductor process as a cleaning solution (Uemura et al., 2007). Therefore, ozone would be expected to have the ability to remove organic deposits such as trace odorous compounds remaining in pipes. Ozone has a great advantage compared to alkaline and acidic solutions with cleaning chemicals because portions of deposits can be degraded by ozone and ozone itself is also degraded to oxygen during treatment and does not remain in the wastewater. In contrast, with conventional treatment, degradation of deposits does not occur and thus the alkaline and acidic wastewaters with deposits and cleaning chemicals require further wastewater treatment.

Gaskets in the production pipe lines must also be considered, since deposits can form on rubber surfaces. Rubber compounds used for pipe lines in the beverage industry are mainly ethylene-propylene-diene monomer (EPDM) rubbers, which have no unsaturated bonds in the main chain and are, therefore, resistant to ozone, thermal aging and weather (Rutherford et al., 2005). Moreover such rubbers have an economical advantage over other rubber compounds such as those based on chloroprene, acrylonitrile-butadiene/polyvinyl chloride, nitrile and silicone. Greene et al. (1994) found that among gasket materials used in fluid food processing, ozone treatment affected the tensile strength of EPDM and Viton, whereas the tensile strengths of polyethylene, silicone rubber and PTFE were not significantly affected; the authors concluded that the effects of ozone treatment on the physical properties of the gasket materials studied were minor even though slight effects were observed on EPDM and Viton. Polyethylene, silicone rubber and PTFE, are, therefore, considered to be strong candidates for gasket materials that would be amenable to ozone cleaning. However, to date, there have been no evaluations of gasket materials from the standpoint of cleaning.

In this study we sought to establish a rapid and green CIP process to remove trace odorous compounds from beverage products adsorbed on gasket rubber. Two removal approaches were tested; the application of an ozone cleaning method and changing of the gasket material from EPDM rubber (EPDM gasket) to silicone rubber (silicone gasket).

2. Materials and methods

2.1. Materials

The flavor of an orange consists of more than 200 compounds (Shaw, 1991) and limonene is the most abundant (Gomez-Ariza et al., 2004). Major constituents of apple juices and aromas were reported to be 1-hexanol, 1-butanol, *E*-2-hexenol, *E*-2-hexenal and butyl acetate, and 1-hexanl showed the highest concentration both in the juice and aroma (Elss et al., 2006). The major component of an apple juice concentrates was also reported to be furfural (Elss et al., 2006). 2-Furfurylthiol, 4-vinylguaiacol, several alkylpyrazines, furanones, acetaldehyde, propanal, methylpropanal and 2- and 3-methylbutanal had the greatest impact on the flavor of ground coffee (Czerny et al., 1999). Four odorous compounds, *d*-limonene from an orange, 1-hexanol and furfural from an apple and 4-vinylguaiacol from a coffee were used in this study and main physicochemical properties of these compounds are summarized in Table 1; they were purchased from Sigma Chemicals.

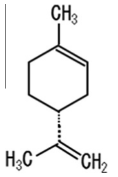
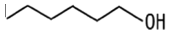
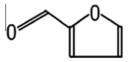
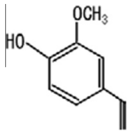
Prior to this research we investigated a beverage company to ascertain the CIP procedures and solutions in use. The alkaline cleaning solution for a conventional cleaning process contained 3% NaOH with detergents (Adeka Mate AC from Adeka Clean Aid) and the acidic cleaning solution contained 3% HCl with phosphate detergents (Adeka 3 from Adeka Clean Aid) and descaling agents (mixture of Pankereto from Hirata Kagaku and Alfa-RP from Futaba Kosan).

Gaskets of EPDM and silicone rubbers were purchased from Air Water Mach. The gasket diameters were 10.8 mm (inside) and 15.6 mm (outside) and the weight was 0.24 g.

2.2. Adsorption of odorous compound on gasket

A 300 mL-beaker with 250 mL of distilled water produced using a RFD240RA (Advantec) was prepared and a glass column (20 mm diameter) was inserted into the beaker. The top of the column was over the surface of the water and the bottom was set at the middle of the beaker. Then an odorous compound was added on the surface of the water inside the glass column. The odorous compound formed an oil layer on the surface of the water because of limiting solubility in water. To make the odorous compound saturated water, the water was stirred using a magnetic stirrer for 3 h. Then

Table 1
Physicochemical properties of odorous compounds used in this study.

Compound	Molecular structure	Molecular Mass (g mol^{-1})	Water solubility (g L^{-1})	Log P
d–limonene		136.2	0.014[a]	4.20
1–hexanol		102.2	5.9[b]	2.03
Furfural		96.1	83[b]	0.41
4–vinylguaiacol		150.2	0.93[a]	2.57[c]

[a] Water solubility at 25 °C.
[b] Water solubility at 20 °C.
[c] Predicted by using the ACD/Labs' ACD/PhysChem Suite.

10 EPDM or silicone gaskets were set up on hooks to prevent contact with each other (which otherwise might have contributed to some non-uniformity of adsorption) and then submerged in the water outside the glass column. The EPDM or silicone gaskets were kept in the saturated water at 80 °C for 12 and 9 h, respectively, until about 4–6 mg g^{-1}-gasket of *d*-limonene was adsorbed on the gaskets. The amounts of 1-hexanol, furfural, 2-furfurylthiol and 4-vinylguaiacol adsorbed on the gaskets were 0.085–0.4 mg g^{-1}-gasket, which were much lower than that of *d*-limonene because of low adsorbability.

2.3. Cleaning

The plant we investigated in Hiroshima prefecture, Japan, used the common CIP procedures typically reported; water flushing, alkaline cleaning with detergents, acidic cleaning with detergents and disinfection by chemical disinfectants such as sodium hypochlorite. Therefore, we adopted the CIP procedure of this company as the conventional cleaning procedure; specifically, rinsing by water at room temperature for 5 min, alkaline solution cleaning with detergents at 80 °C for 35 min, rinsing by water at room temperature for 20 min, acidic solution cleaning with detergents at 70 °C for 50 min, hot water cleaning at 70 °C for 50 min, rinsing by water at room temperature for 40 min and rinsing by hot water at 70 °C for 20 min. The chemical disinfection process was not performed in this experiment because the disinfection process is not connected with the removal of odorous compound. The total time to complete the conventional cleaning process was 220 min.

The 10 gaskets, which adsorbed about the odorous compound, were set up on hooks and submerged into the glass cylinder of 250 mL capacity with acidic or alkaline solution. The solution was weakly agitated by a magnetic stirrer for 60 min.

For ozone treatment, 100 mg O_3 L^{-1} of ozone gas produced by an ozonaizer (POX-10, Fuji Electric) was introduced into the glass cylinder through a diffuser at a flow rate of 50 mL min^{-1}. Ozone treatment was carried out at a water temperature of 20 or 70 °C for 60 min. In order to clarify the main cleaning mechanism for gaskets, ozone treatment at 70 °C was also carried out with 10 mM of *t*-butanol to scavenge hydroxyl radicals (°OH). Cleaning by hydroxyl radicals, produced by addition of 32 mM hydrogen peroxide during ozone treatment, was also carried out at 70 °C.

2.4. Analysis

Adsorbed *d*-limonene on the gasket before and after cleaning was extracted in acetone. Ten gaskets were submerged in 50 mL of acetone for 24 h and then the amount of *d*-limonene extracted into acetone was determined by GC/MS (Shimadzu QP 5050) with an Agilent Technologies DB-VRX capillary column (30 m × 0.25 mm id). The injector temperature was 230 °C. The oven temperature started at 40 °C and was programmed to rise at the rate of 6 °C min^{-1} to 180 °C which was held for 2 min. The MS transfer temperature was set at 240 °C.

Furfural and 1-Hexanol were extracted in pentane-diethyl ether (1:1) and were determined by GC/MS (Shimadzu QP 5050) with an Agilent Technologies DB-WAX capillary column (30 m × 0.25 mm id). The injector temperature was 220 °C. The oven temperature started at 40 °C. After 5 min, the temperature was risen at the rate of 10 °C min^{-1} to 90 °C, and then risen at the rate of 10 °C min^{-1} to 120 °C. The MS transfer temperature was set at 230 °C.

Extraction of 4-vinylguaiacol was done in dichloromethane and was determined by GC/MS (Shimadzu QP 5050) with an Agilent Technologies DB-FFAP capillary column (30 m × 0.25 mm id). The injector temperature and MS interface temperature were the same as the temperatures for 1-hexanol and furfural analysis. The oven temperature started at 35 °C. After 2 min, the temperature was risen at the rate of 40 °C min^{-1} to 60 °C and was held for 2 min, and then was raised at the rate of 6 °C min^{-1} to 250 °C which was held for 10 min.

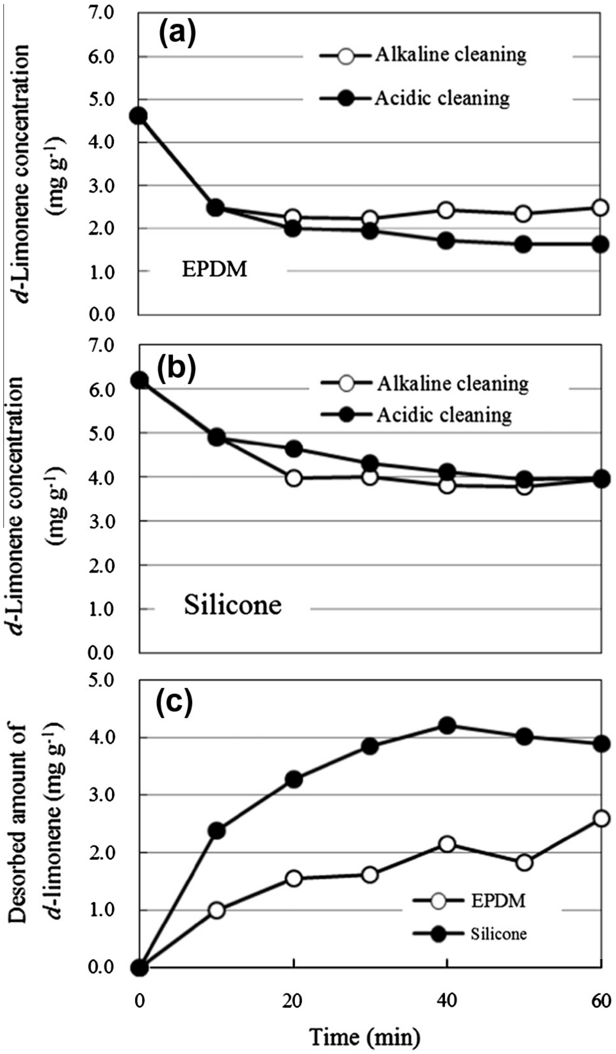


Fig. 1. Residual amounts of *d*-limonene on the EPDM (a) and silicone (b) gaskets during alkaline and acidic cleanings and desorption behavior of *d*-limonene from the EPDM and silicone gaskets (c).

The appearance of gasket treated with ozone was observed by the scanning electron microscope (SEM) (JCM-5700, JEOL) that was operated at an accelerated voltage of 20 kV.

3. Results and discussion

3.1. Effect of gasket materials on conventional cleaning method

To evaluate the effect of gasket materials on conventional cleaning method *d*-limonene was used. Both acidic and alkaline cleaning are effective at reducing the amount of *d*-limonene adsorbed on both types of gaskets (Fig. 1a and b). However, the removal efficiencies for *d*-limonene adsorbed on the EPDM gaskets were less than 40% in 60 min for both alkaline and acidic cleanings, whereas those for *d*-limonene adsorbed on the silicone gaskets were 50% and 65% for 60 min of alkaline and acidic cleanings, respectively, suggesting that the silicone gasket was better than the EPDM gasket from the view point of conventional cleaning efficiency.

To confirm which material is more readily cleaned, the desorbability of *d*-limonene from gaskets was also evaluated by using acetone which is a strong extraction solvent. On both gaskets, 4 mg g^{-1} of pre-adsorbed *d*-limonene was completely extracted by acetone in 24 h (Fig. 1c). However, 4 mg g^{-1} of pre-adsorbed *d*-limonene was not completely removed from the EPDM gasket in 60 min, whereas removal from the silicone gasket was complete in 30 min. This result further indicates that *d*-limonene was adsorbed more strongly on the EPDM gasket than the silicone gasket.

Silicone polymers are more hydrophobic than EPDM polymers because of the abundance on silicone of low surface energy (CH_3 groups) (Kim and Kim, 2001) and the adsorption capacity of silicone for *d*-limonene would be higher than that for EPDM. In fact, the water contact angle, which is an index for the surface hydrophobicity, is 105° for the silicone gasket (Khorasani et al., 2006), whereas it is 84° for the EPDM gasket (Sarathi et al., 2002). The amount of *d*-limonene adsorbed on the silicone gasket within a specified time was observed to be higher than EPDM in these experiments (data not shown). Although the silicone gasket with higher adsorbability to *d*-limonene was expected to show lower desorbability than the EPDM gasket, the desorbability of *d*-limonene from the silicone gasket was higher than that from the EPDM gasket. Davies et al. (1996) reported the similar result, that is, a silicone gasket contaminated by albumin was more effectively cleaned by cleaning solution (NaOH/Triton X-100) than was an EPDM gasket.

Finally, the superiority of the silicone gasket with regard to cleaning was confirmed by conventional cleaning. The total removal efficiencies for *d*-limonene were 41% and 75% for gaskets made of EPDM and silicone, respectively (Fig. 2a). Adsorbed *d*-limonene on the silicone gasket could be removed more quickly and thoroughly than that adsorbed on the EPDM gasket. However, even 220 min of conventional cleaning did not completely remove 4 mg g^{-1} of *d*-limonene adsorbed on either the EPDM or silicone gaskets.

3.2. Cleaning of *d*-limonene by ozone

Ozone treatment of EPDM and silicone gaskets at 20 °C was clearly effective in reducing residual amounts of adsorbed *d*-limonene (Fig. 2a); the removal efficiencies of *d*-limonene were 86 and 100% for EPDM and silicone gaskets, respectively, which were much higher than those by acidic or alkaline solution with detergents (see Fig. 1a and b). However, ozonation after 30 min did not further reduce *d*-limonene adsorbed on the EPDM gasket. The combination of the silicone gasket and ozone treatment resulted in the most effective cleaning. The cohesion of *d*-limonene on the surface of the silicone gasket was weaker than that on the EPDM gasket (see Fig. 1c). These different desorption properties may affect the cleaning efficiencies.

For the silicone gasket, increasing the water temperature from 20 and 70 °C did not enhance the cleaning efficiency (Fig. 2b). It is well established that ozone reactions may follow two pathways: direct reaction of molecular ozone (dipolar addition, electrophilic substitution), and indirect reaction involving free radicals (mainly °OH) formed following ozone decomposition in water (Staehelin et al., 1984; Tomiyasu et al., 1985). We compared the cleaning efficiencies by ozone in the presence or absence of *t*-butanol (known radical scavenger) at 70 °C; addition of *t*-butanol had no effect on the cleaning efficiency, suggesting that the high removal efficiency of *d*-limonene adsorbed on the silicone gasket by ozone was mainly caused by molecular ozone.

Limonene is known to be one of the biogenic volatile organic compounds that is emitted into the atmosphere from vegetation; the reactivity of limonene in the atmosphere has been studied (Atkinson and Arey, 2003). Atkinson and Arey (2003) reported that limonene was highly reactive with ozone with calculated lifetime of 2.0 h in the troposhpere. The reaction pathway of ozone with limonene has been described by others (Leungsakul et al., 2005a,b). Ozone reaction with *d*-limonene takes place at the internal cyclo double bonded carbon or at the external isopropylene bond.

Ozonation can change non-polar/hydrophobic organic carbons such as limonene into polar/hydrophilic organic carbons. Limononic acid, hydroxylimononic acid and ketolimononic acid, ketolimononaldehyde, hydroxylimononaldehyde and limononaldehyde were reported as byproducts in the ozonation of limonene (Pun et al., 2006). These aldehydes and organic acids are polar/hydrophilic organic carbons and can be easily detached from the hydrophobic surface. Therefore, we suggest that ozone attacked *d*-limonene adsorbed on the gasket surface and decomposed it into polar/hydrophilic organic carbons, which would then desorb from the surface during ozonation.

The appearance of both gaskets treated with ozone was observed by the naked eyes and the SEM and was found to remain unchanged after 60 min of treatment. However, Miwa et al. (2011) reported the surface degradation of EPDM by ozone in water. The experimental condition in their study was much more severe than our condition, that is, 5.5 ± 0.5 mg L^{-1} of ozone concentration and 24 h of exposure, whereas 2.0 mg L^{-1} in average and 1 h in our experiment. The mechanism of the degradation in EPDM was estimated that the C=C double bonds in the 5-ethylidene-2-norbornene units were oxidized and the propagation reaction led to changes of the C—H bonds into ketone, carboxyl, and hydroxyl groups. The authors also mentioned that the manner of surface oxidation was unique for EPDM. The silicone will not be degraded by the same mechanism because it does not have C=C double bonds in the structure.

3.3. Cleaning of other odorous compounds

Effectiveness of changing the gasket material from EPDM rubber to silicone rubber and ozone treatment in reducing residual amounts of odorous compounds adsorbed on the gaskets were also

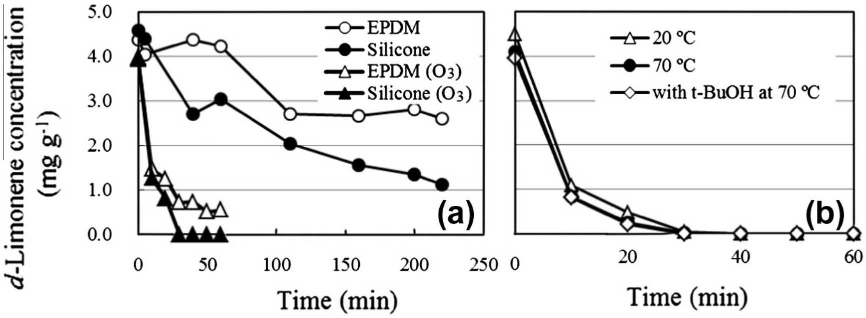


Fig. 2. Residual amounts of *d*-limonene on the gaskets during the conventional cleaning and ozone treatment (a) and during ozone treatment at different temperatures with and without *t*-butanol (b).

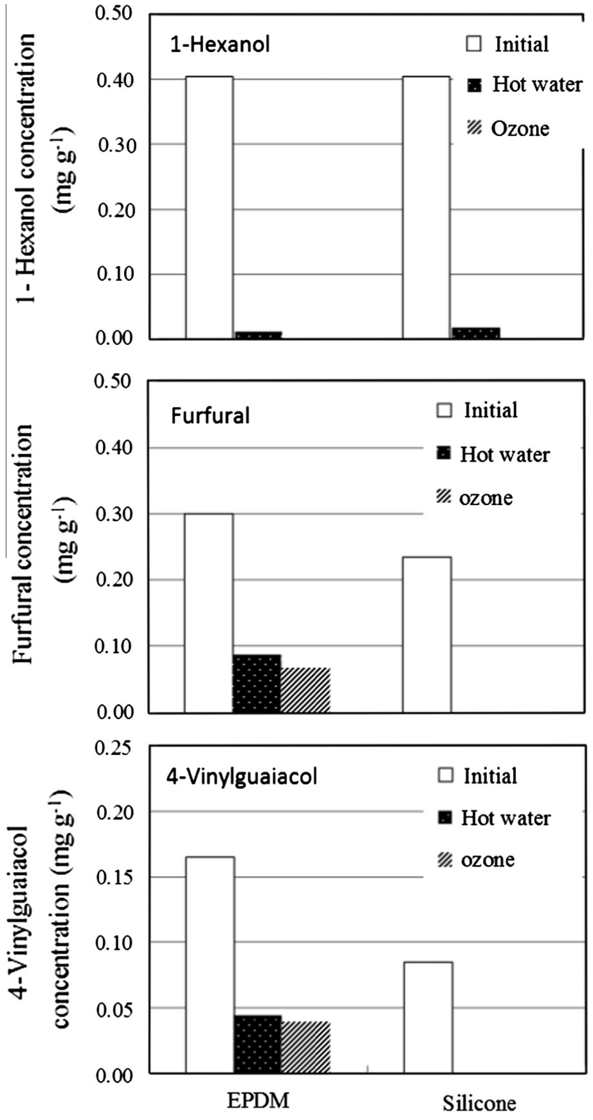


Fig. 3. Residual amounts of 1-hexanol, furfural and 4-vinylguaiacol on the EPDM and silicone gaskets after 60 min of hot water and ozone treatments.

evaluated for 1-hexanol and furfural from an apple and 4-vinylguaiacol from a coffee. Hot water cleaning at 70 °C for 60 min was effective at reducing the amount of 1-hexanol, furfural and 4-vinylguaiacol adsorbed on both types of gasket (Fig. 3). However, the removal efficiencies for furfural and 4-vinylguaiacol adsorbed on the EPDM gasket were about 70%, whereas those for furfural and 4-vinylguaiacol adsorbed on the silicone gasket were 100%, suggesting that the silicone gasket was better than the EPDM gasket from the view point of hot water cleaning efficiency. The higher desorbabilities of these three chemicals from the silicone gasket than the EPDM gasket were in agreement with the result for *d*-limonene.

The complete removal of 1-hexanol adsorbed on both types of gasket was achieved by ozone treatment. Ozone treatment showed a little improvement of the hot water cleaning efficiency for furfural and 4-vinylguaiacol adsorbed on the EPDM gasket.

Furfural was known to have much lower reactivity toward ozone (Borghei and Hosseini, 2008). Guaiacol was one of the intermediate products during lignin degradation by ozone-based processes and was reported to be oxidized by ozone (Ko et al., 2011). However, there is no report about the reactivity of 4-vinylguaiacol to ozone. The reactivity of guaiacol to ozone may decline to be combined with vinyl group. On the other hand, primary alcohols including 1-hexanol can be oxidized into ketones and carboxylic acids by ozone (Yu et al., 2007). The reactivity of each chemical would influence the improvement of cleaning efficiency by ozone.

4. Conclusions

In this study we have sought to establish a rapid and green CIP procedure to remove the trace odorous compound adsorbed on rubber gaskets. We investigated the application of an ozone cleaning method and a change of gasket material from EPDM to silicone. By changing the gasket material from EPDM rubber to silicone rubber the removability of *d*-limonene by typical alkaline and acidic cleaning solutions with detergents was improved. However, complete removal of 4 mg g^{-1} of *d*-limonene on both EPDM and silicone gaskets could not be achieved even if a series of conventional cleaning processes including alternative alkaline and acidic cleaning were used for 220 min. Ozone treatment resulted in a substantial improvement in the removability of *d*-limonene from both EPDM and silicone gaskets; the removal efficiencies were 87% for the EPDM gasket in 60 min and 100% for the silicone gasket in 30 min. The combination of the silicone gasket and ozone treatment resulted in the most effective cleaning. The main removal mechanism by ozone treatment was confirmed to be oxidation by molecular ozone. Effectiveness of changing the gasket material from EPDM rubber to silicone rubber in reducing residual amounts of odorous compounds adsorbed on the gaskets were also confirmed for furfural and 4-vinylguaiacol.

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