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Treatment characteristics of amino acid and sterilization mechanism Using RF oxygen plasma

高周波酸素プラズマによるアミノ酸の処理特性と滅菌機序の解明

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Spore of microorganism is quite resistant to heat, high pressure and chemical agent, and therefore its treatment is important. In order to investigate sterilization factors and mechanism of the plasma treatment for medical equipment using oxygen plasma, interaction between oxygen plasma and *Bacillus subtilis var. Natto* or metionine that is major composition of spore coat is investigated.

1. Introduction

On medical equipment sterilization, treatment of bacillus and virus attached surface of medical equipment is significantly important. Since, spore coat of Gram-positive bacteria contains amino acid abundantly, it is necessary to investigate the decomposition characteristics of amino acid in spore bacillus for sterilization. It is difficult to sterilize spore coat, therefore conventional sterilization uses heat, strong acid and radial ray, although those methods cannot be applied to non heat-proof and non chemical-proof, materials [1]. Recent years, methods of sterilization of medical equipment and foods are developed using plasma [2]. However, inactivation mechanism of bacillus and virus using plasmas is not clarified yet. In this study, metionine that is sulfur containing proteinogenic amino acids located spore coat and *Bacillus subtilis var.natto* are exposed to oxygen plasma. The treatment characteristics of amino acid and the sterilization mechanism are determined using RF oxygen plasma.

2. Experimental apparatus

The chamber made of stainless is evacuated and oxygen gas is introduced at several Pa. The oxygen plasma is produced using the RF CCP antenna. The RF power is 60W and the treatment time is 2 h. The samples to be treated are metionine and *Bacillus*

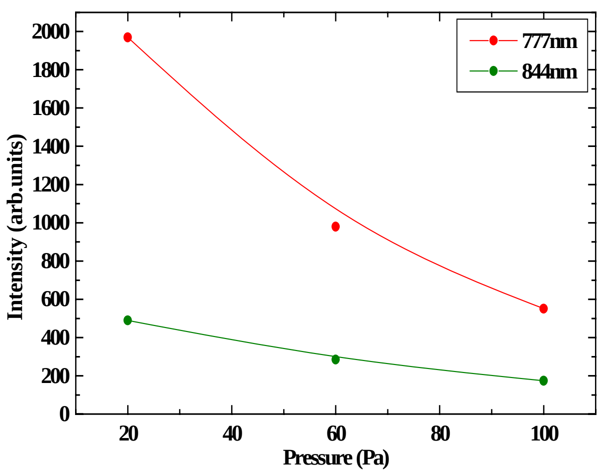

Fig.1 Dependency of light emission intensity from oxygen radicals on the gas pressure.

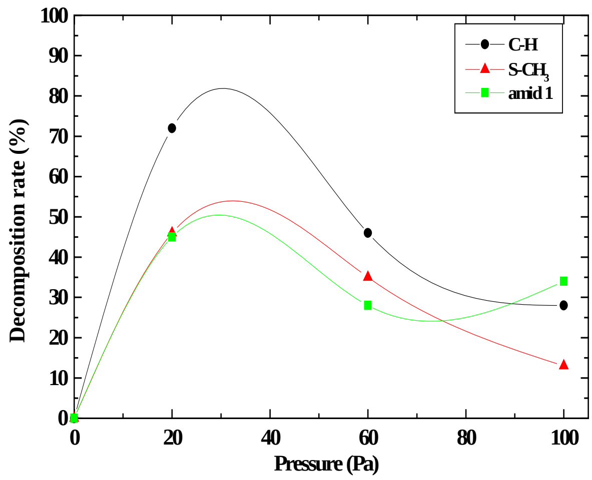

Fig.2 Decomposition rate of major bonds of metionine varying the pressure.

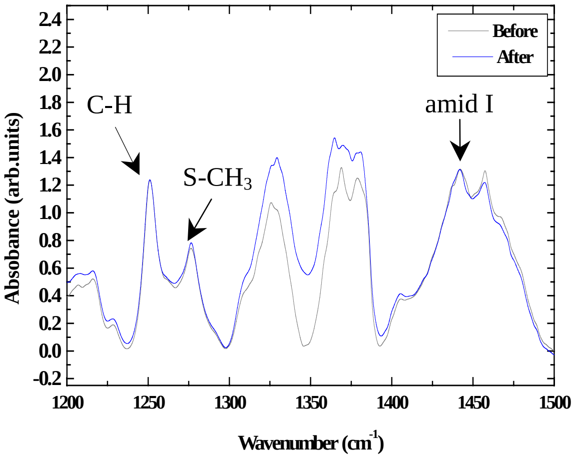

Fig.3 FTIR spectra of plasma treatment *Bacillus subtilis var. natto* at the pressure of 20Pa.

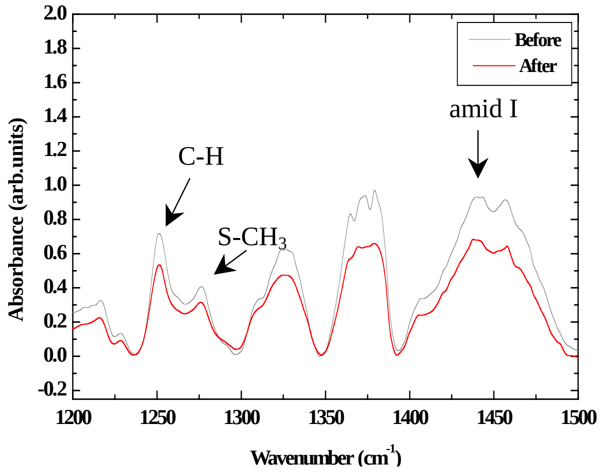

Fig.4 FTIR spectra of plasma treatment *Bacillus subtilis var. natto* at the pressure of 100Pa.

subtilis var. natto. These materials are put on CaF_2 plate and dried completely, and set on the center of chamber and exposed to oxygen plasma. The density of metionine and *Bacillus subtilis var. natto* is 19mg/cm^2 const and distance of sample to CCP antenna is 6cm [2]. After treatment, these samples are measured FTIR spectroscopy.

3. Result and discussion

3.1 *Treatment of metionine*

Figure2 shows decomposition rate of metionine estimated from major peak in FTIR spectra of sample before and after the plasma irradiation. At the pressure of 20Pa, C-H bond (at 951cm^{-1}) is decomposed 70% at maximum, and decomposition rates of S-CH_3 bond (at 1316cm^{-1}) and amid I (at 1617cm^{-1}) was 46%, 45% respectively.

While, at the pressure of 100Pa, the decomposition rates of C-H bonds and S-CH_3 bonds are lower than those of 20Pa, 60Pa, and decomposition rate of amid I is larger at 60Pa than 20Pa. Furthermore, it is obvious that oxygen radicals dissociate and combine to another bond and produces sulfur oxide (SOx), like a sulfo acid and cisteine acid at 1045cm^{-1}, 1149cm^{-1} and 1183cm^{-}1 a maximum at 100Pa. Moreover, the decomposition rate of amid I at the pressure of 60Pa is the lowest, 30% and of C-H bonds, S-CH_3 bonds are 47%, 34% respectively.

3.2 *Treatment of Bacillus subtilis var. natto*

Figure3 shows *Bacillus subtilis var. natto* exposed oxygen plasma at the pressure of 20Pa. Major peak C-H bonds (at 1250cm^{-1}), S-CH_3 bonds (at1275cm^{-1}) and amid I (at 1443cm^{-1}) cannot dissociate at the pressure of 20Pa. Figure4 shows the sample of *Bacillus subtilis var. natto* at the pressure of 100Pa. Major peak of C-H bonds, S-CH_3 bonds and NH bonds. Those decomposition rates are 41%, 31% and 28%, respectively.

On the treatment of *Bacillus subtilis var. natto*, surface on its spore coats would be laked by oxygen ions and radicals. Then spore coats lost the function of spore. Therefore, decomposition of sulfur-containing proteinogenic amino acids are one of the candidates of spore inactivation.

4. Conclusion

Decomposition characteristics of amino acid and bacillus spore are studied using the RF oxygen plasma, and sterilization mechanism of spore is investigated.

- Metionine is decomposed 70% (C-H bonds), 46% (S-CH_3 bonds) and 45% (amid I) at the pressure of 20Pa and generated sulfur oxide at 100Pa. At 100Pa decomposition rate of C-H bonds, S-CH_3 bons and amid I are 28%, 12% and 34%, respectively.
- *Bacillus subtilis var. natto* is decomposed 41% (C-H bonds), 30% (S-CH_3 bonds) and 28% (amid I), each at the pressure of 100Pa. Oxygen ions etch on the surface of bacillus on the CaF_2 plate.

References

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