FAR-INFRARED IRRADIATION EFFECT ON PASTEURIZATION OF BACTERIA ON OR WITHIN WET-SOLID MEDIUM

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The present purpose is to study the influence of far-infrared irradiation on pasteurization of *Escherichia coli* and *Staphylococcus aureus* on or within a model for wet-solid food. Agar medium was used as the food model. By determining the thermal resistances of the test bacteria, the pasteurization effect of far-infrared irradiation (radiative heating) was compared with that of hot-air heating (a conventional method) from the viewpoint of thermal death kinetics. It was found experimentally that far-infrared irradiation is more effective than hot-air heating for the test bacteria on the agar-plate. Moreover, it is suggested that the surface temperature of the pasteurization sample irradiated by far-infrared radiation is higher than that measured by the thermocouples.

Introduction

For infrared radiative heating, no heating medium is needed, and the air between an infrared heater and the irradiated matter is not heated. Application of infrared radiation to thermal operations can be expected to draw considerable interest for the following reasons: saving of energy, simplification of apparatus, clean working environments and easy thermal control. Furthermore, infrared radiative heating is superior to microwave heating and ultra-violet rays in terms of safety and economy.

Recently, the technology for producing infrared heaters with high emissivity in the region of far-infrared (FIR) radiation and with low emissivity in the region of near-infrared radiation has improved.⁸⁾ We classify infrared radiation in the wavelength range longer than 3 μm as FIR. In broad fields and in many important industries, applications of FIR hold considerable attraction. In the food process industry, FIR is already positively utilized.

Most processed foods are thermally treated, and are sufficiently pasteurized. However, the systems for processing from food materials to food products are complicated and spreading, and microorganisms adsorbed on processed food after thermal treatment shorten the shelf-life. Microbial contamination of foodstuffs is a matter of considerable concern. It is important, for prevention of microbial contamination of food products, to pasteurize the microorganisms

adsorbed after thermal treatment. New technology for preventing processed foods from microbial contamination is expected.

Organic materials and water, the main components of foodstuffs and microorganisms, considerably absorb FIR, and especially absorb radiative energy at wavelengths near 3 and 6 μm .^{1,5)} FIR is safe for foodstuffs and human beings. Fundamental studies of the application of FIR to pasteurization in food processing are thus desirable.

For *Escherichia coli* and *Staphylococcus aureus* suspended in phosphate-buffered saline (PBS, pH 7.0), the pasteurization effect of FIR irradiation was compared with that of thermal conductive heating (a conventional method), under the condition that the transient behavior of the bulk temperature of the suspension irradiated by FIR was the same as that heated by thermal conduction³⁾. The viable ratio of the test bacteria in the suspension irradiated by FIR was much less than that heated by thermal conduction. The superiority of FIR pasteurization to that by thermal conductive heating was confirmed. Moreover, it was detected that *E. coli* and *S. aureus* suspended in PBS kept below the lethal temperature were injured and killed by FIR irradiation²⁾.

However, there has been no fundamental study on pasteurization of solid food by FIR irradiation. Such fundamental study is needed for the application of FIR to direct pasteurization of wrapped foods after thermal treatment.

The present purpose is to study the influence of FIR irradiation on pasteurization of bacteria on or within a food model for wet solid food. Agar medium was

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used as the food model, as agar medium is nutritional enough for microorganisms and is analogous to many wet solid foods in its optical and thermal properties. By using the viable ratio calculated from the rate constant for thermal death, pasteurization by FIR irradiation (radiative heating) is compared with that by hot-air heating (a conventional method). The rate constant is obtained experimentally. Furthermore, by focusing our attention on the absorption of radiative energy in the region of FIR, the characteristics of FIR pasteurization of bacteria on or within the food model are studied.

1. Experimental Details

1.1 Preparation of sample

E. coli 745 and *S. aureus* 9779 stored at Tokyo Metropolitan Research Laboratory of Public Health were used. The test bacteria were cultured in Brain Heart Infusion broth (Difco) at 308 K for 24 h on a reciprocal shaker.

Agar plate medium was prepared in a petri dish (14.5 × 88.5 mmφ). Nutrient agar (Eiken Chemicals) was used for *E. coli*, and Standard Method agar (Eiken Chemicals) was used for *S. aureus*. The culture was diluted with sterile saline, and the dilution was overspread on the surface of the agar-plate to give a bacterial concentration of 200 ~ 300 CFU · plate⁻¹. The agar-plate was used as the pasteurization sample. By adding the agar medium at a thickness of 1 to 5 mm on the test bacteria, the other pasteurization sample was prepared (Fig. 1).

1.2 Pasteurization by far-infrared irradiation

The irradiation chamber (W 0.43 × D 0.32 × H 0.58 m) is made of aluminium plates, and a far-infrared heater (300 × 15 mmφ) with a reflector is placed at the top. The irradiation apparatus was that used in the previous studies.^{2,3)} The power irradiated on the agar-plate was from 2.46×10^3 to 7.01×10^3 W · m⁻², and irradiation time was from 1 to 8 min. Irradiation power was obtained experimentally from the evaporation rate⁶⁾.

After a specified electric power level was supplied to the heater, and after the surface temperature of the heater and the room temperature reached steady state, the pasteurization sample was placed under the heater and FIR irradiation was carried out. After irradiation for a specified time, the agar-plate in the petri dish was placed in an incubator at 308 K. After 24 h in the case of no addition of the agar medium, and after 48 h in the case of its addition, the colonies were enumerated.

CA-thermocouples sheathed by a stainless steel pipe (0.25 mmφ) were placed horizontally at four positions between the surface and the bottom. The agar-plate temperatures at four points were thereby recorded during irradiation.

Fig. 1. Pasteurization sample

Fig. 2. Photograph of sample for determining thermal resistances of the test bacteria

1.3 Pasteurization by hot-air heating

A constant-temperature and constant-humidity chamber (Tabai PR-3G) was used for pasteurization by hot-air heating. The heating chamber (W0.60 × D0.80 × H0.85 m) is made of stainless steel.

After the temperature and the humidity in the chamber reached steady state, the petri dish containing the agar-plate was placed at the center of the chamber and heating was carried out. Hot air temperature was from 323 to 363 K, the relative humidity was kept at 50%, and heating time was from 5 to 30 min.

The microbial counts and measurement of the agar-plate temperature were carried out by the same methods as those for pasteurization by FIR irradiation.

1.4 Thermal resistances

The dilution of the culture with sterile saline (5.0×10^{-4} dm³) and the agar-sol medium (4.5×10^{-3} dm³) kept below 323 K were dispensed into a sterile pouch made of polyvinyl chloride and were mixed thoroughly. The pouch was sealed at the nozzle (Fig. 2) in order to set a fixed shape and to prevent contamination during pasteurization and incubation. The mixture was gelatinized, and an agar-plate (1.0 × 85 mmφ) was prepared in the pouch.

To determine the thermal resistances for thermal conductive heating, a hot water bath with a stirrer was used. The pouch was exposed to the water bath and was vibrated during pasteurization. After heating for a specified time, the pouch was cooled by a water bath kept at 278 K. The agar-plate in the pouch was

placed in an incubator at 308 K. After 48 h, the colonies were enumerated. We call the method of determining the thermal resistances the “Agar-Pouch method”.

The CA-thermocouples sheathed by a stainless steel pipe (0.25 mm ϕ) were placed at the center of the agar-plate to record the temperature during heating.

2. Experimental Results and Discussion

2.1 Pasteurization by far-infrared irradiation

Figure 3 shows the transient behavior of the temperature profile of the agar-plate irradiated by FIR at an irradiation power (q_{ir}) of $4.36 \times 10^3 \text{ W} \cdot \text{m}^{-2}$, and parameter d is the distance between the surface and the test bacteria (Fig. 1). Figure 4 shows the survival curves of *E. coli* irradiated by FIR under the condition shown in Fig. 3, and N/N_0 is the viable ratio. N/N_0 on the agar-plate ($d=0 \text{ mm}$) is markedly lower than N/N_0 for $d \geq 1 \text{ mm}$. With decrease in d , N/N_0 decreases. For 6 min irradiation, all of the test bacteria at $d=0 \text{ mm}$ are killed, though N/N_0 at $d=1 \text{ mm}$ is 2.0×10^{-1} and N/N_0 at $d=2 \text{ mm}$ is 2.2×10^{-1} .

Under this experimental condition, N/N_0 does not start to decrease until the agar-plate temperature surrounding the test bacteria, which is measured by the thermocouples, reaches the lethal temperature (Figs. 3 and 4). The pasteurization results by FIR irradiation for *S. aureus* show a similar trend to those for *E. coli*, suggesting that the agar-plate temperature surrounding the test bacteria is one of the main factors in the pasteurization effect of FIR irradiation.

2.2 Pasteurization by hot-air heating

Figure 5 shows the transient temperature profile at the surface of the agar-plate heated by hot air. The steady temperature at the surface of the agar-plate approximately equals the wet-bulb temperature in the hot air. Figure 6 shows the survival curves of *E. coli* on the agar-plate pasteurized by hot air under the condition shown in Fig. 5. N/N_0 on the agar-plate decreases with increase in the hot air temperature and in the heating time.

It is impossible to set up experimental conditions such that the transient temperature profile of the agar-plate heated by hot air becomes equal to that heated by FIR. So, by using N/N_0 calculated from the thermal resistances of the test bacteria, the pasteurization effect of FIR irradiation is compared with that of hot-air heating. To determine the thermal resistances, pasteurization of the test bacteria by the Agar-Pouch method was performed.

2.3 Thermal resistances of test bacteria

Figure 7 shows the pasteurization results by the Agar-Pouch method for *E. coli*. The time required for the agar-plate to reach constant temperature, which differs no more than 0.1 K from the bath temperature, is less than several seconds. And, as shown in Fig. 7,

Fig. 3. Transient behavior of temperature of agar-plate irradiated by FIR

Fig. 4. Survival curves of *E. coli* from exposure to far-infrared radiation

Fig. 5. Transient behavior of temperature of agar-plate heated by hot air

it may be considered that the death of *E. coli* is logarithmic. For *S. aureus*, the same trends as for *E. coli* are found. The death rate of the test bacteria is thus assumed to be expressed mathematically by the following equation⁹⁾.

$$-\frac{dN}{dt_h} = k_{ag} N \quad (1)$$

Fig. 6. Survival curves of *E. coli* from exposure to hot air

Fig. 7. Survival curves of *E. coli* in pouched agar-plate from exposure to water bath temperature

N is the bacterial concentration in the agar-plate, and t_h is the heating time. The rate constant for thermal death, k_{ag} , is the proportionality factor and is obtained from the slope of the pasteurization curves shown in Fig. 7.

Figure 8 shows the experimental values of k_{ag} for *E. coli* and *S. aureus*. The frequency factor A and the activation energy E_a for thermal death are obtained from the results shown in Fig. 8 by using the following equation (Arrhenius' plot)⁹⁾.

$$k_{ag} = A \cdot \exp(-E_a/RT_{ag}) \quad (2)$$

A and E_a are shown in Table 1.

2.4 Comparison of pasteurization effect

The calculated values of N/N_0 for *E. coli* based on Fig. 5 and Table 1 are obtained by using Eq. (3), and are shown in Fig. 6.

$$N/N_0 = \exp\left(-\int_0^{t_h} k_{ag} dt_h\right) \quad (3)$$

The calculated results agree consistently with the experimental results in hot-air heating pasteurization for *E. coli* except for $T_a = 343$ K. However, in that case the calculated survival curve is similar to the

Fig. 8. Relationship between k_{ag} and $1/T_{ag}$

Table 1. Thermal resistances of the test bacteria

Bacteria	E_a [J·mol ⁻¹]	A [s ⁻¹]
<i>E. coli</i>	3.18×10^5	2.32×10^{47}
<i>S. aureus</i>	2.53×10^5	2.85×10^{37}

Fig. 9. Effect of d on N/N_0 for *E. coli*

experimental survival curve. For *S. aureus*, the same trends as for *E. coli* are obtained, suggesting that the calculated results may evaluate the experimental results for hot-air heating pasteurization.

In Figs 9 and 10 the results of FIR pasteurization are compared with the calculated results based on Fig. 3 and Table 1. In the region of $d \geq 1$ mm, the calculated results for *E. coli* agree with the experimental results approximately, and the calculated results for *S. aureus* show the same trend as the experimental results. For both *E. coli* and *S. aureus* the experimental results are smaller than the calculated results for the test bacteria on the agar-plate ($d = 0$ mm). Then, for the test bacteria on the agar-plate it is conceivable that N/N_0 in the case of FIR irradiation is lower than that

Fig. 10. Effect of d on N/N_0 for *S. aureus*

in the case of hot-air heating.

Comparison of the pasteurization effects described above indicates that FIR irradiation is more effective than hot-air heating for the test bacteria on the agar-plate. Thus it may be possible to apply FIR to the pasteurization of microorganisms adsorbed on processed food after the thermal treatment.

2.5 Pasteurization effect of far-infrared irradiation

The differences in pasteurization effects between FIR irradiation and hot-air heating are discussed by focusing our attention on the surface temperature of the agar-plate irradiated by FIR. A model of the survival curve (Fig. 4) is calculated from Eq. (3) by using Table 1 and the model of the transient surface temperature of the agar-plate indicated in Fig. 3. The model of the transient surface temperature is determined by trial and error in order to fit the model of the survival curve to the pasteurization results, especially in the region of $N/N_0 \leq 1/5$.

The model of the survival curve agrees consistently with the pasteurization results for *E. coli* on the agar-plate ($d=0$ mm) by far-infrared irradiation. The model of the transient surface temperature is higher than the transient temperature at $d=0$ mm measured by the thermocouples. From Figs. 3 and 4 it is thinkable that the surface temperature of the agar-plate irradiated by FIR is higher than the measured temperature at $d=0$ mm, if the death of the test bacteria irradiated by FIR may be due to thermal death.

So, consider the absorption of radiative energy by the pasteurization sample. As the agar medium consists mainly water (>90 wt%), the absorption coefficient of the agar-plate may be assumed to be approximately equal to that of water^{1,5)}. A model of the test bacteria composed of the freeze-dried material of the test bacteria and of water is considered, and

Fig. 11. Monochromatic absorption coefficients

the apparent absorption coefficient is estimated by using the methods described in the previous paper⁴⁾. Figure 11 shows the apparent absorption coefficient, which may be assumed to be equal approximately to that of water⁶⁾ from the viewpoint of radiative heat transfer. Therefore, it is thinkable that absorption of FIR at the surface of the pasteurization sample is almost the same as that for water. At a 773 K surface temperature of the heater, the power irradiated on the pasteurization sample is reduced to 80% of the initial value within a layer of 3 μm thickness, which is almost the size of the test bacteria, and is reduced to 10% of the initial value within a layer of 40 μm thickness³⁾.

The above suggests that as the temperature of the test bacteria on the agar-plate is higher than the temperature measured by the thermocouples at $d=0$ mm, N/N_0 values for FIR pasteurization are smaller than the calculated results. By scanning electron micrographs for gelatinization in a very thin domain near the surface of a noodle irradiated by FIR, Nakano⁷⁾ suggested that the surface temperature of the noodle was higher than the temperature measured by the thermocouples. Besides, as the penetration of radiative energy into the agar-plate is very small, N/N_0 in the region of $d \geq 1$ mm is estimated by calculated results due to pasteurization by thermal conductive heating.

3. Concluding Remarks

Agar medium was used as a food model, and pasteurization of *E. coli* and *S. aureus* on or within the agar-plate was performed by FIR irradiation and by hot-air heating. And, by determining the thermal resistances of the test bacteria, the pasteurization effect of FIR irradiation was compared with that by hot-air heating from the viewpoint of thermal death kinetics. The following results and the conclusions are obtained.

1) The viable ratio for FIR pasteurization can be evaluated for the test bacteria within an agar-plate

deeper than 1 mm. But, for the test bacteria on the agar-plate, the experimental results of the viable ratio for FIR pasteurization are smaller than the calculated results.

2) It is found experimentally that FIR irradiation is a more effective means of pasteurization than hot-air heating for the test bacteria on the agar-plate. And it is suggested that the surface temperature of the pasteurization sample irradiated by FIR is higher than that measured by the thermocouples.

Nomenclature

A	= frequency factor defined by Eq. (2)	$[s^{-1}]$
d	= distance from surface of agar-plate	$[mm]$
E_a	= activation energy obtained from Eq. (2)	$[J \cdot mol^{-1}]$
k_{ag}	= thermal death rate constant for pasteurization obtained from Eq. (2)	$[s^{-1}]$
N	= colony-forming unit per agar-plate	$[CFU \cdot plate^{-1}]$
R	= gas constant (=8.314)	$[J \cdot K^{-1} \cdot mol^{-1}]$
T	= temperature	$[K]$
t_h	= heating time	$[s]$
α	= absorption coefficient	$[m^{-1}]$
λ	= wave length	$[\mu m]$

<Subscripts>

a	= air
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ag	= agar-plate
b	= water bath
h	= infrared heater
λ	= monochromatic
0	= initial state

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