Comparison of ATP Values on Meat and Fish Cutting Boards before and after Alcohol Disinfection

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I. INTRODUCTION

Sanitary control of cutting boards in the kitchen is important to prevent food poisoning. In the past, we reported the results of hygiene management by repeatedly cleaning the cutting board with detergent and running water for 30 seconds or more. Currently, the COVID-19 epidemic requires stricter hygiene control. To control invisible microorganisms, it is necessary to take measures to avoid the risk of food poisoning due to familiarity with cooking work. As O-JT education, it is necessary to create a hygiene management manual and protect it with all the cooks. However, if the procedure is complicated and difficult, it will not last long. We need easy and reliable procedures and methods that anyone can do. The ATP tests and HACCP-based microbiological tests are useful in hygiene management to transform invisible bacteria into visible forms and educate them. Therefore, in this study, and the cutting board cleaning method we performed last time, a step of spraying 70% alcohol added. The effects of this alcohol disinfection compared by adding a stamping test (General bacteria, Staphylococcus aureus, Escherichia coli, Salmonella, Vibrio parahaemolyticus) used. The stamp test conducted by the inspector at the same time as the ATP. The stamp test was colony-counted after culturing in an incubator at 38 degrees for three days. The stamp test made by NISSUI.

II. MATERIALS AND METHODS

a) Kitchen cutting board

The six kitchen meat or fish thick cutting board (cutting board 1) and the six kitchen meat for the fish thin cutting board (cutting board 2) prepared in the kitchen were stored in the sterilization storage the day before cooking.

b) ATP inspection procedure

Each of the 12 cooks carried a kitchen cutting board for meat or fish at the start of their work and brought it to the cooking table. The work start time depends on the working conditions of the cooks. Still, the inspector always performed an ATP inspection before using meat or fish with a kitchen cutting board. Then, each cook finished the work, washes the cutting board firmly with detergent and sponge, rinse with running water for 30 seconds or more. Then, each cook repeated this process twice (as same as the last report). The inspector performed an ATP inspection after using meat or fish with a kitchen cutting board, again. Then, each cook sprayed 70% alcohol on the cutting board after washing. At last, the inspector performed an ATP inspection after using meat or fish with a kitchen cutting board. The ATP test kit used manufactured by KIKKOMAN.

c) Stamp test inspection procedure

Five types of stamp test (General bacteria, Staphylococcus aureus, Escherichia coli, Salmonella, Vibrio parahaemolyticus) used. The stamp test conducted by the inspector at the same time as the ATP. The stamp test was colony-counted after culturing in an incubator at 38 degrees for three days. The stamp test made by NISSUI.

d) Statistical processing

The results obtained compared using statistical methods. The data were statistically processed, was subjected to an F test to determine whether to use a parametric test or nonparametric test. When there is no difference in the F test, the presence or absence of a significant difference was confirmed using the student t-test with or without a correspondence. If there was a difference in the F test, the presence or absence of a significant difference was confirmed using the Wilcoxon
test with a pair or the Mann-Whitney test without correlation.

III. Results

a) ATP value results before and after alcohol disinfection

The table 1 and 2 shows the results of ATP wiping tests on cutting board before and after alcohol disinfection. It can see that the average value of the ATP values measured after washing before and after cleaning, after cleaning, the ATP value is low. The ATP value after 70% alcohol spraying was statistically significantly lower than that before alcohol spraying. The ATP value dropped below 100 for both cutting boards.

| No alcohol treatment   | Alcohol treatment |
|------------------------|-------------------|
| 1 | 844 | 501 | 13 |
| 2 | 210 | 56  | 31 |
| 3 | 132205 | 103 | 20 |
| 4 | 59141 | 62  | 31 |
| 5 | 30814 | 272 | 18 |
| 6 | 76010 | 70  | 72 |
| Average value           | 51132.3 | 8447.3 | 30.8 |
| SD                      | 49166.2 | 20416.0 | 21.4 |

b) Stamp test results before and after alcohol disinfection

Tables 3, 4, 5, 6, 7, 8, 9, 10, 11 and 12 show the results of ATP wiping tests on cutting board before and after 70% alcohol disinfection. Results of general bacteria show in Tables 3 and 4. Results of E. coli show in Tables 5 and 6. Results of Staphylococcus aureus shown in Tables 7 and 8. Result of Salmonella show in Tables 9 and 10. Result of Vibrio parahaemolyticus show in Tables 11 and 12. The number of all microbial bacteria was lower after washing than after cooking and after spraying 70% alcohol. However, there was no statistically significant difference in the number of microbial bacteria.

| No alcohol treatment   | Alcohol treatment |
|------------------------|-------------------|
| 1 | 58449.7 | 784.5 | 39.2 |
| 2 | 64022.1 | 1500.9 | 18.9 |

Table 3 Number of general bacteria on cutting board 1. and statistical processing result

| No alcohol treatment   | Alcohol treatment |
|------------------------|-------------------|
| 1 | 93.2 | 15.8 | 2.3 |
| 2 | 8  | 15.8 | 2.3 |
| 3 | 4  | 15.8 | 2.3 |
| 4 | 2  | 15.8 | 2.3 |
| 5 | 200 | 9   | 14 |
| 6 | 60  | 9   | 14 |
| Average value           | 93.2 | 15.8 | 2.3 |
| SD                      | 87.7 | 21.4 | 5.7 |

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Table 4  Number of general bacteria on cutting board 2. and statistical processing result

|     | No alcohol treatment | Alcohol treatment |
|-----|----------------------|-------------------|
|     | Before washing       | After washing     | After washing | After alcohol |
| 1   | 270                  | 7                 | 7             | 0             |
| 2   | 1                    | 18                | 18            | 8             |
| 3   | 200                  | 8                 | 8             | 0             |
| 4   | 2                    | 7                 | 7             | 0             |
| 5   | 200                  | 23                | 23            | 0             |
| 6   | 61                   | 20                | 20            | 0             |
|     | Average value        | 122.3             | 13.8          | 13.8          | 1.3           |
|     | SD                   | 115.6             | 7.3           | 7.3           | 3.3           |

F test  
Student-t*  
Wilcoxon  
P=0.0001**  
P=0.035*  
P=0.116  
P=0.028*  
P=0.075

*Paired Student-t test  
P<0.05, ** P<0.01

Table 5  Number of E. coli on cutting board 1. and statistical processing result

|     | No alcohol treatment | Alcohol treatment |
|-----|----------------------|-------------------|
|     | Before washing       | After washing     | After washing | After alcohol |
| 1   | 0                    | 0                 | 0             | 0             |
| 2   | 2                    | 1                 | 1             | 0             |
| 3   | 200                  | 8                 | 8             | 0             |
| 4   | 5                    | 0                 | 0             | 7             |
| 5   | 200                  | 0                 | 0             | 0             |
| 6   | 6                    | 0                 | 0             | 16            |
|     | Average value        | 68.8              | 1.5           | 1.5           | 3.8           |
|     | SD                   | 101.6             | 3.2           | 3.2           | 6.6           |

F test  
Student-t*  
Wilcoxon  
P=0.0001**  
P=0.052  
P=0.518  
P=0.043*  
P=0.418

*Paired Student-t test  
P<0.05, ** P<0.01

Table 6  Number of E. coli on cutting board 2. and statistical processing result

|     | No alcohol treatment | Alcohol treatment |
|-----|----------------------|-------------------|
|     | Before washing       | After washing     | After washing | After alcohol |
| 1   | 3                    | 2                 | 2             | 0             |
| 2   | 0                    | 0                 | 0             | 1             |
| 3   | 200                  | 23                | 23            | 0             |
| 4   | 0                    | 0                 | 0             | 0             |
| 5   | 15                   | 0                 | 0             | 0             |
| 6   | 35                   | 0                 | 0             | 0             |
|     | Average value        | 42.2              | 4.2           | 4.2           | 0.2           |
|     | SD                   | 78.5              | 9.3           | 9.3           | 0.4           |

F test  
Student-t*  
Wilcoxon  
P=0.0001**  
P=0.0001**  
P=0.068  
P=0.285  
P=0.080

*Paired Student-t test  
P<0.05, ** P<0.01

Table 7  Number of Staphylococcus aureus on cutting board 1. and statistical processing result

|     | No alcohol treatment | Alcohol treatment |
|-----|----------------------|-------------------|
|     | Before washing       | After washing     | After washing | After alcohol |
| 1   | 0                    | 0                 | 0             | 0             |
| 2   | 5                    | 1                 | 1             | 0             |
| 3   | 0                    | 0                 | 0             | 7             |
| 4   | 0                    | 0                 | 0             | 3             |
| 5   | 152                  | 2                 | 2             | 0             |
| 6   | 212                  | 0                 | 0             | 0             |
|     | Average value        | 61.5              | 0.5           | 0.5           | 1.7           |
|     | SD                   | 95.3              | 0.8           | 0.8           | 2.9           |

F test  
Student-t*  
Wilcoxon  
P=0.0001**  
P=0.004**  
P=0.109  
P=0.465  
P=0.345

*Paired Student-t test  
P<0.05, ** P<0.01
### Table 8  Number of Staphylococcus aureus on cutting board 2. and statistical processing result

|        | No alcohol treatment | Alcohol treatment |
|--------|----------------------|-------------------|
| For meat | Before washing | After washing | After washing | After washing | After alcohol |
| 1      | 0                   | 0                 | 0             | 0             |
| 2      | 0                   | 1                 | 1             | 16            |
| 3      | 24                  | 0                 | 0             | 0             |
| 4      | 0                   | 1                 | 1             | 0             |
| 5      | 200                 | 0                 | 0             | 0             |
| 6      | 432                 | 0                 | 0             | 0             |
| Average value | 109.3   | 0.3              | 0.3           | 2.7           |
| SD     | 176.3               | 0.5              | 0.5           | 6.5           |

**F test**

- Student-t*: P=0.0001**
- Wilcoxon: P=0.225

**F test**

- Student-t*: P=0.0001**
- Wilcoxon: P=0.655

*Paired Student-t test  * P<0.05, ** P<0.01

### Table 9  Number of Salmonella on cutting board 1. and statistical processing result

|        | No alcohol treatment | Alcohol treatment |
|--------|----------------------|-------------------|
| For meat | Before washing | After washing | After washing | After washing | After alcohol |
| 1      | 508                 | 0                 | 0             | 0             |
| 2      | 80                  | 0                 | 0             | 0             |
| 3      | 1                   | 1                 | 0             | 2             |
| 4      | 168                 | 0                 | 0             | 5             |
| 5      | 1                   | 9                 | 9             | 0             |
| 6      | 26                  | 15                | 15            | 5             |
| Average value | 130.7   | 4.0              | 4.0           | 2.0           |
| SD     | 195.4               | 6.5              | 6.5           | 2.4           |

**F test**

- Student-t*: P=0.0001**
- Wilcoxon: P=0.975

**F test**

- Student-t*: P=0.0001**
- Wilcoxon: P=0.465

*Paired Student-t test  * P<0.05, ** P<0.01

### Table 10  Number of Salmonella on cutting board 2. and statistical processing result

|        | No alcohol treatment | Alcohol treatment |
|--------|----------------------|-------------------|
| For meat | Before washing | After washing | After washing | After washing | After alcohol |
| 1      | 21                  | 0                 | 0             | 0             |
| 2      | 6                   | 0                 | 0             | 0             |
| 3      | 1                   | 7                 | 7             | 0             |
| 4      | 119                 | 0                 | 0             | 0             |
| 5      | 0                   | 38                | 38            | 2             |
| 6      | 18                  | 0                 | 0             | 0             |
| Average value | 27.5    | 7.5              | 7.5           | 0.3           |
| SD     | 45.7                | 15.2             | 15.2          | 0.8           |

**F test**

- Student-t*: P=0.0001**
- Wilcoxon: P=0.402

**F test**

- Student-t*: P=0.0001**
- Wilcoxon: P=0.138

*Paired Student-t test  * P<0.05, ** P<0.01

### Table 11  Number of Vibrio parahaemolyticus on cutting board 1. and statistical processing result

|        | No alcohol treatment | Alcohol treatment |
|--------|----------------------|-------------------|
| For meat | Before washing | After washing | After washing | After washing | After alcohol |
| 1      | 21                  | 0                 | 0             | 0             |
| 2      | 6                   | 0                 | 0             | 0             |
| 3      | 1                   | 7                 | 7             | 0             |
| 4      | 119                 | 0                 | 0             | 0             |
| 5      | 0                   | 38                | 38            | 2             |
| 6      | 18                  | 0                 | 0             | 0             |
| Average value | 27.5    | 7.5              | 7.5           | 0.3           |
| SD     | 45.7                | 15.2             | 15.2          | 0.8           |

**F test**

- Student-t*: P=0.0001**
- Wilcoxon: P=0.402

**F test**

- Student-t*: P=0.0001**
- Wilcoxon: P=0.138

*Paired Student-t test  * P<0.05, ** P<0.01
IV. Discussion

To manage the hygiene of meat and fish cutting board that has a high risk of causing secondary contamination in cooking. We tried to verify using the ATP test and microbial stamp test by spraying 70% alcohol after cleaning instead of controlling only by the cleaning method. The ATP value decreased after washing then after cooking and after spraying 70% alcohol than after washing. The ATP value was a statistically significant decrease, which was less than 100 after 70% alcohol spraying. However, the microbial stamp test results were not statistically significant reductions in bacterial counts. The cutting board inspected by spraying 70% alcohol after cleaning. But if 70% of alcohol not sprayed after sufficiently wiping off the water, the alcohol may be dilute, and the bactericidal effect may weakened. In the future, we would like to verify the sterilization of microorganisms by spraying 70% alcohol on the cutting board by thoroughly wiping off the water after cleaning and then spraying 70% alcohol. Not all microorganisms are killed even after spraying 70% alcohol, so when using a cutting board left at room temperature (with moist), it is better to wash repeatedly and cook after spraying 70% alcohol.

V. Conclusions

The effects of 70% alcohol spraying investigated using cutting boards for meat and fish. Both cutting boards had high ATP and microbiological test values after cooking. However, although the value of the cutting board decreased after cleaning, the ATP value did not fall below 100. Microbial test values were also high in many cases. After spraying with 70% alcohol, the ATP value was 100 or less, and the value decreased statistically significantly. Microbial test values were decreasing with or without statistically significant reductions. Providing safe and secure meals by further spraying 70% alcohol after cleaning the cooking utensils helps prevent food poisoning. However, since the microorganisms are present even after spraying with 70% alcohol, the bacteria may grow again if the cooking utensils left for a long time. It is advisable to clean and spray 70% alcohol before using the equipment.

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