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Total Bacterial Count and Presence of Escherichia coli in Raw Ground Beef Samples Obtained from Three Major Supermarkets on the Island of Bonaire

Bacteria are everywhere. Some of them are good, some are indifferent and some are really bad. Foods can harvest all kinds of bacteria, but bacterial growth depends on plenty of factors. Grounded meat, for example, is more perishable than whole muscle cuts, because more of the meat surface is exposed to the harmful bacteria and the grinding process mixes them throughout the product.

Since our senses (smell, taste, touch, vision and the others) can neither detect bacteria nor distinguish between good, bad and indifferent, more sophisticated methods have to be used to evaluate the danger of bacteria in foods. Both the presence of specific bacteria and total bacterial counts help to evaluate the general hygiene, quality and the safety of foods. In meat products commonly tested are APC (Aerobic Plate Counts), nasty E. coli 0157:H7 and its “Big Six” Shiga toxin-producing cousins that are common causes of outbreaks.

To avoid false positive and false negative results, it is important to follow scientific standards and procedures designed for each step – from the sample collection to the interpretation and the presentation of the results. And SJSM students know that. Now, let’s see how they evaluated the hygiene, food safety and the needs for the improvements of safe meat purchase and handling on Bonaire.

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Total Bacterial Count and Presence of *Escherichia coli* in Raw Ground Beef Samples Obtained From Three Major Supermarkets on the Island of Bonaire, Dutch Caribbean

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Abstract

The quality of raw ground beef from three supermarkets in Kralendijk, Bonaire, Dutch Caribbean was compared by assessing colony forming units per gram (CFU/g) of total bacteria and to a lesser degree, presence of *Escherichia coli* (*E. coli*). Over 4 weeks, 12 samples of raw ground beef were obtained from three supermarkets, resulting in 36 total samples. Dilutions of meat were inoculated on plates of nutrient agar and allowed to incubate for 48 hours. Total bacteria counts were obtained for each sample. The three mean CFU/g were calculated and analyzed. It was found that the sample mean from Store 1 was significantly higher than those from Store 2 and Store 3. There was no significant difference between the latter two. Additionally, 5 samples were tested from each supermarket for *E. coli*. *E. coli* was detected in samples from Store 1 and to a lesser degree in samples from Store 2. *E. coli* was not found in samples from Store 3. Finally, Gram staining procedure revealed gram-negative bacteria in samples from each store.

Introduction

Several investigators have reported on total bacterial counts and occurrence of E. coli in retail ground beef and beef products The aim of the present study was to determine the total bacterial counts of raw ground beef sold at three of the major supermarkets in Kralendijk, Bonaire, Dutch Caribbean. Particularly the presence of *E. coli* in the meat sold at the same three supermarkets, will help us to determine if there is a need to suggest the improvements of safe meat purchase and handling amongst the producers, distributors and consumers. Presence of E. coli and total bacterial counts of meats are used to help determine general hygiene, quality and safety of meat products 4, 5 and 6. Bacteria such as *E. coli* that are often found in raw meats cause thousands of cases of illness, some of which result in hospitalizations and mortality each year[1,2]. Some of these bacteria are also resistant to antibiotics[2]. High total bacterial counts, also called Aerobic Plate Counts (APC), of raw meat are also associated with decreased shelf life[3].

Methods

Samples of raw ground beef were obtained from the three largest supermarkets on the island of Bonaire, Dutch Caribbean from the regular consumer sections each Monday, Wednesday and Friday for four weeks, totaling 36 samples.

Dilutions of raw ground beef were prepared by agitating 0.1 gram and 0.01 gram of meat in 10 ml sterile distilled water for two minutes. An inoculating loop was used to deliver 0.01 ml of the dilution to petri dishes prepared with nutrient agar. The mixture was spread uniformly. Plates were inverted and allowed to incubate aerobically for 48 hours at ambient temperature. Total bacterial colony forming units were counted manually and then converted to CFU/g. After assumptions were met, the means were compared using One-way ANOVA and unpaired t-tests.

Five samples were randomly selected to also test for the presence of *E. coli*. Petri dishes containing MacConkey agar were streaked with 0.01 ml of the diluted samples extracted from 0.1 grams of meat. Dishes were inverted and allowed to incubate at ambient temperature for 48 hours. Pink *E. coli* forming units were counted. Means and proportions were compared using unpaired t-tests and Fisher's exact test.

Two colony forming units from each supermarket were also prepared by Gram stain. Crystal violet was the primary dye and safranin the counterstain. Slides were examined for the presence of gram-negative bacteria.

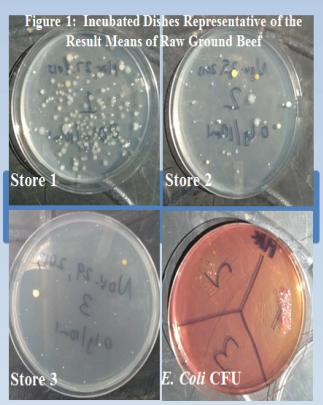

Figure 1. Incubated dishes representative of the result means of raw ground beef from three supermarkets in Bonaire, Dutch Caribbean. Store 1: Plated Nov. 27, 0.01 g meat diluted, 220,000 CFU/g. Store 2: Plated Nov. 25, 0.1 g meat diluted, 3700 CFU/g. Store 3: Plated Nov. 29, 0.1 g meat diluted, 4000 CFU/g. *E. coli* CFU: Plated Nov. 27, 0.1 g meat diluted for each store.

Results

The means of Aerobic plate count (APC) CFU/g of the sampled raw ground beef were different (p=1.488E-16). Specifically, the raw ground beef from Store 1 has a mean APC CFU/g that is higher than both Store 2 and Store 3 at a Bonferroni-adjusted p=0.025 significance level, while the means of APC CFU/g from Store 2 and Store 3 are not statistically different from each other (p=0.609).

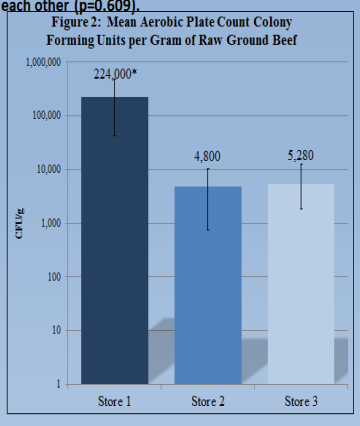

Figure 2. Graph of mean APC CFU/g of raw ground beef from three supermarkets in Bonaire, Dutch Caribbean with log scale. n=12 for each supermarket. Error bars represent 95% confidence intervals. *Denotes significance (p=1.488E-16) in this mean varying from the others.

Additionally, when comparing the *E. coli* data at a significance level of p=0.025, the only difference was between Store 1 and Store 3, with Store 1 having a higher proportion of presence and mean colony count of *E. coli*. Finally, gram-negative bacteria was seen in each of the six Gram stained slides.

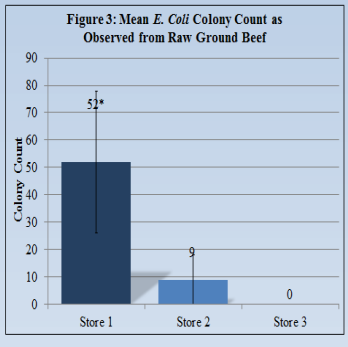

Figure 3. Graph of mean *E. coli* colony forming units of raw ground beef from three supermarkets in Bonaire, Dutch Caribbean. 0.1 grams raw meat diluted in 10 ml sterile distilled water. 0.01 ml dilution plated. n=5 for each supermarket. Error bars represent 95% confidence intervals. *Denotes significance (p=0.022) in this mean varying from the mean of Store 3.

Discussion

Our results pertaining to total bacterial counts fall within standards of health (maximum CFU/g = 10^7 colonies per gram) suggested by the Institute of Food Science and Technology[7]. The means of APC CFU/g from Store 2 and Store 3 are similar to numbers reported in nationwide US studies of raw ground beef, while the mean APC CFU/g from Store 1 is considerably larger[5, 6]. This suggests that although the Store 1 data is within limits due to health, its overall bacterial levels are greater than normal. This can negatively impact the shelf life of the meat[3] and poses possible health concerns for Store 1 consumers who undercook their beef or allow cross contamination.

While *E. coli* was detected in several of our samples, there are various different kinds of *E. coli*, many of which are opportunistic[2]. Therefore, we cannot make direct conclusions as to the health risk involved. Further research needs to be done. The presence of gram-negative bacteria in the Gram stained slides, along with the presence of *E. coli* in samples from two stores, suggest that other gram-negative pathogenic bacteria (generally more resistant to antibiotics[8]) such as *Salmonella* or *Shigella* may be present in the beef.

Limitations and Future Research

The major limitations of this study were the relatively smaller sample sizes and narrow scope. Further research could add immensely to our findings.

Suggestions include:

- Typing of specific bacteria
- Assessing level of antibiotic resistance
- Address Store 1 and why their beef differs in total bacteria count and *E. coli* proportions and counts than the other supermarkets.
- Assess other local stores at various times of the year

Conclusion

This study serves as a good foundation in assessing the overall quality of raw ground beef in Bonaire. Statistical comparisons allow us to see that there are differences in the means of total bacteria CFU/g and *E. coli* presence between different supermarkets. This information allows consumers to recognize where the highest quality of beef can be obtained as it relates to contamination, and to a lesser degree, prevalence of *E. coli*. Specific meat producers can see that improvements can be made in their products. All should be advised to handle and cook meat safely. Areas for further research are suggested as a means of building upon the groundwork laid here.

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