



MICROBIOLOGICAL QUALITY OF SELECTED READY-TO-EAT FOOD AT HULU LANGAT DISTRICT, MALAYSIA

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ABSTRACT

A study was conducted to evaluate the microbiological quality of ready-to-eat (RTE) meals in three different sites at Hulu Langat District in Malaysia between September to December 2007. Forty eight RTE samples were collected from cafeteria of Faculty of Science and Technology in the Universiti Kebangsaan Malaysia (UKM), restaurants within the Kajang Town and the street vended RTE foods in the night market at Bandar Baru Bangi and the quality of these foods sold and the hygienic conditions under which sites operated were compared. The study found that the RTE food vendors in the night market and the restaurants in this district were capable of preparing and selling relatively safe foods, with low bacterial counts, although there was still a need for proper hygiene conditions and supporting of basic sanitary facilities. The microbiological quality of the food samples was compared to the local microbiological guidelines for RTE foods formulated by the Ministry of Health Malaysia and other international guidelines. The total aerobic counts was exceeded the proposed guidelines. Neither *Salmonella* nor coagulase-positive Staphylococci were identified in the samples. However, low counts of *E. coli* were present which indicate the degree of ignorance from the food handlers causing a need for food safety regulations and vendors education. Therefore, the results of the study indicate that even though the bacterial levels detected in the food were below the set guideline limits except for the TPC, it still require the local authority in that area to emphasize on educating the RTE food handlers. This is to avoid problems which might develop, and to ensure that the standard of RTE meals is best achieved at the time of sale and consumption.

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ABSTRAK

Kajian ini dijalankan untuk mengenalpasti tahap kualiti mikrobiologi bagi makanan- sedia-dimakan, (MSD) dari tiga kawasan di Daerah Hulu Langat, Malaysia, antara bulan September sehingga Disember 2007. Empat puluh lapan jenis sampel MSD diambil dari Kafeteria Fakulti Sains dan Teknologi, Universiti Kebangsaan Malaysia, restoran di sekitar Pekan Kajang, serta dari peniaga makanan di pasar malam Bandar Baru Bangi, dan seterusnya kualiti makanan yang dijual serta tahap kebersihan proses penyediaan kesemua makanan tersebut dibandingkan. Kajian mendapati bahawa peniaga-peniaga makanan sedia dimakan di pasar malam, dan juga restoran di daerah ini, telah memenuhi ciri-ciri penyajian dan penyediaan makanan yang sepatutnya, dan kesemua makanan selamat dimakan berdasarkan keputusan kiraan koloni bakteria yang rendah. Namun begitu, keperluan untuk peningkatan tahap kebersihan serta penyediaan kemudahan sanitasi yang sempurna haruslah dititikberatkan. Tahap kualiti mikrobiologi bagi kesemua sampel makanan tersebut dibanding dengan garis panduan piawaian mikrobiologi, keselamatan makanan tempatan bagi MSD yang digariskan oleh Kementerian Kesihatan Malaysia serta institusi piawaian antarabangsa. Hanyai Kiraan Koloni Piring didapati melebihi tahap piawaian yang ditetapkan. Kesemua sampel kajian ini tidak menunjukkan kehadiran Salmonella dan Staphylococci koagulase-positif. Namun E. coli didapati hadir dalam bilangan yang rendah, dan ini sekaligus menunjukkan berlakunya kecuaiian oleh pengendali makanan itu sendiri. Maka, perhatian haruslah dititikberatkan aspek perundangan dan pendidikan kepada peniaga. Hasil kajian ini menunjukkan walaupun tahap pencemaran bakteria di dalam MSD adalah rendah dari tahap piawaian yang ditetapkan, namun pihak berkuasa tempatan seharusnya memberi pengetahuan asas kepada pengendali makanan. Ini bagi mengelakkan masalah yang sama berulang, dan juga untuk memastikan tahap piawaian bagi makanan sedia dimakan mencapai tahap kebersihan yang terbaik terutama ketika jualan dan juga ketika hidangan untuk santapan.

Katakunci: Kualiti mikrob, makanan sedia dimakan, Hulu Langat, Malaysia

INTRODUCTION

Ready-to-eat (RTE) food is the food that is ready for immediate consumption at the points of sale (Food and Environment 2001). It can be raw or cooked, hot or chilled and can be consumed without heat including reheating. In general, RTE foods are all food products including chilled

and frozen foods, fruits (either cut or minimally process), vegetables (that consumed as salad) and beverages (which prepared at the vending site including flavoured ice drink). These foods did not require further processing or heating before consume even though, some times it need to be reheated before serving (Gillbert et al. 2000). RTE included snacks may also be prepared and served separately, including dried meat, fish and cereal based ready-to-eat food (Gadaga et al. 2007). Several studies showed that different pathogens have been isolated from RTE foods including verocytotoxigenic *Escherichia coli* (VETC), *Salmonella* spp. and *Campylobacter* spp., confirming the risk posed by consuming these foods (Gibbons et al. 2006). Ingestion of foods containing these types of microorganism causes food poisoning which is a health problem that may significantly reduce economic growth (Mankeea et al. 2003). Many studies were done for microbiological quality and food safety evaluation on the RTE foods whether at street vending, restaurants or in specific locations such as hospitals, universities, school and others. Furthermore, similar studies were carried out in Malaysia to demonstrate the presence of specific microorganisms in certain foods such as prevalence of Salmonella in raw and cooked foods in Malaysia (Arumugaswamy et al. 1995) and incident of Klebsiella pneumonia in street foods sold in Malaysia (Haryani et al. 2007). However, some of reports in food poisoning cases from different sources in Malaysia have been reported by ministry of health (MOH 2002). The overall objective of this study were to compare the microbiological quality of ready-to-eat food (meat products) sold at three different markets namely Night market in Bandar Baru Bangi, Kajang restaurants and UKM's cafeteria.

MATERIALS AND METHODS

Sample Collection

Samples were collected from September to December 2007 in Hulu Langat District in Malaysia. A total of 48 ready-to-eat (RTE) meals were collected in two sampling batches representing four types of popular foods in Malaysia (12 chicken curry, 12 beef curry, 12 fried chicken and 12 chicken with chilly sauce). A total of 16 samples were collected from each of the three sites namely night market in the Bandar Baru Bangi, restaurants in Kajang and cafeteria of the Faculty of Science and Technology, Universiti Kebangsaan Malaysia (UKM). Samples were collected at the beginning and the end of the market supply between 5-9 pm from the Bandar Baru Bangi and Kajang while samples were purchased at 10 am to 3 pm from the cafeteria of the Faculty of Science and Technology. Eight samples were purchased from each site on one sampling batch (4 samples at the beginning of the daily supply and other 4 samples at the end of the day). Three

different locations were chosen to sample the street food in Bandar Baru Bangi night market namely, vendors at the roadside, vendors in the middle and at the end, whereas, 2 restaurants at different location were chosen in Kajang.

Sample Preparation

25g of food samples were homogenized and diluted in 225ml sterile maximum recovery diluent (MRD) in a stomacher bag for 60 sec to give a 10^{-1} homogenate dilution. Another 25g of food samples were weighed and diluted with buffer peptone water (BPW) specific for *Salmonella* spp. pre-enrichment test (AS 1766.2.5 1991).

Microbiological Analysis

Total Plate Count (Total Aerobic Count)

Samples for the total plate count were serially diluted in MRD to 10^{-6} . The pour plate method was applied and the calculation according to AS1766 2.1 1991 and AS1766 1.3 1991.

Total Coliforms

Coliforms is a lactose-positive members of the Enterobacteriaceae was determined by most probable number (MPN) method (AS 1766.2.3 1992). From each dilution 1 ml was transferred to each triple tubes containing lauryl sulphate tryptose (LST) broth containing Durham tubes. Coliform control test was prepared by inoculating with either one of the following reference culture *Enterobacter aerogenes* NCTC 1006 or *Escherichia coli* NCTC 9001.

Escherichia coli

In this study, *E. coli* were determined by combining the Australian and the British Standard procedures MPN method as similar process of coliform determination. The gas positive lauryl sulphate tryptose (LST) broth tubes were gently agitated and subcultured to *E. coli* (EC) medium which were incubated for 48 h at temperature 44°C and followed by indole test (AS 1766.2.3 1992). *E. coli* control test was carried out with the two previous reference cultures.

Salmonella spp.

Salmonella was detected by surface spread plate method as described by Australian Standard (AS 1766.2.5-1991). Two *Salmonella* reference cultures, *Salmonella Salford-IMVS* 1710 and *Citrobactor freundii*-NCTC 9750 were inoculated in control tubes of BPW. The suspect colonies were subcultured in order to obtain pure colonies and then incubated for 24 h at 37°C.

Biochemical tests were analyzed after the subculture incubation in Triple Sugar Iron (TSI) agar and Microgen test to confirm the *Salmonella* colonies.

Coagulase Positive Staphylococci

Coagulase positive staphylococci was isolated quantitatively by surface spread method according to Australian Standard (AS 1766.2.4.1994). Controls were prepared for the tests by streaking two reference cultures specific for Staphylococci, *Staphylococcus aureus* NCTC 6571- coagulase-positive and *Staphylococcus epidermidis* NCTC 6513- coagulase-negative. Colonies were subcultured into (5 -10 ml) volume of brain heart infusion (BHI) broth and incubated at 37°C for 18-24h. After the incubation, 0.2 ml of the BHI test tubes were dispensed 0.2 ml of prepared blood plasma in a sterile test tube. Control was added and incubated with the samples at 37°C for 4h. Tubes were examined for coagulation after 4h and incubated at room temperature for 10h if negative coagulation and then were examined again.

2.4 Data Analysis

Statistical analysis of the data was performed using T. Test; SPSS software to identify the variance. Significance was determined at the $P < 0.05$ level. The Windows Excel programme was used to draw all the discussion's graphs and to calculate the percentage of the samples that were not complied with guidelines.

RESULTS AND DISCUSSION

Microbiological Quality Of Ready-To-Eat Meals In Hulu Langat District

Total Plate Count

Based on average TPC, UKM's cafeteria rendered the best quality meals at the beginning of the selling day (5.0 log CFU/g), followed by the Bandar Baru Bangi night market (5.1 log CFU/g) (Figure 1A).

Changes of TPC in the samples after three to four hours after the beginning of the selling day are shown in the Figure 1B. From this figure, even though it was observed that TPC growth was not significantly increased in all the samples at the end of the vending day but still gave the poor microbiological quality as the count were higher (4.00 to 7.00 log cfu/g). This may be due to poor handling and keeping method by the vendors. TPCs for samples obtained in Bandar Baru Bangi

ranged from 5.9 to 7.1 log CFU/g while samples from UKM cafeteria ranged from 6.1 to 6.8 log CFU/g.

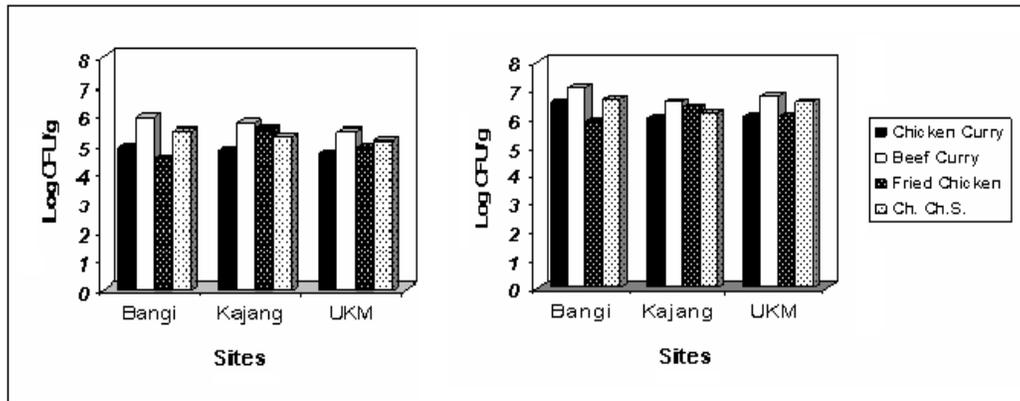


Figure 1 Average total plate count of RTE samples at the beginning (Left) and ending (Right) of the vending day. Ch. Ch. S. = chicken with chili sauce.

According to the results, fried chicken from the night market had the lowest TPC of 5.5 log CFU/g and an average of 5.9 log CFU/g whereas, beef curry from the same source had the highest count of 7.2 log CFU/g with an average of 7.1 log CFU/g. Among the different sites, beef curry had the highest microbial growth value of 6.6 log CFU/g in the restaurants of Kajang and 6.8 log CFU/g at the UKM cafeteria.

Total Coliforms

Similar to results of TPC, fried chicken sold in the night market had the lowest number of coliforms (4 MPN/g) with an average of 9.5 MPN/g at the beginning of the vending day, whereas beef curry from the same site had the largest number of coliforms: 1100 MNP/g with an average of 625 MPN/g (Figure 2A). Even so, coliform counts obtained from beef curry in this experiment was still lower compared to tsire-suya, a traditional RTE meat in Nigeria, that had coliform growth varying from 1×10^2 to 4.2×10^3 cfu/g (Uzeh et al. 2006).

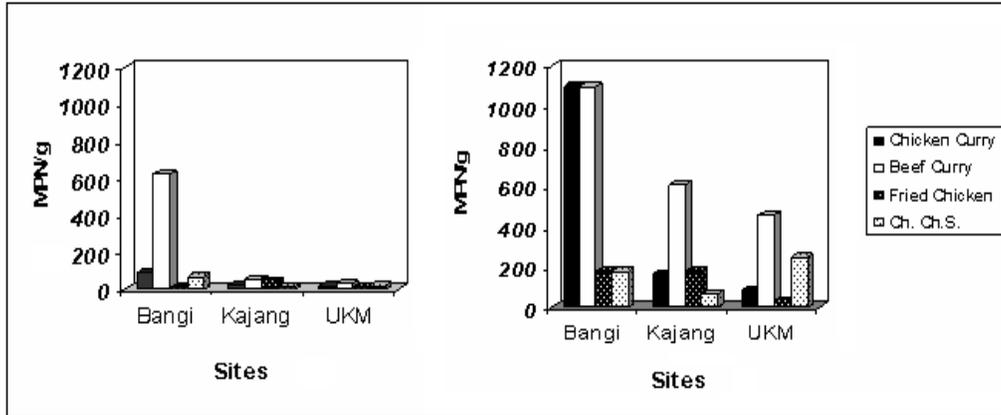


Figure. 2 Average total coliform counts of RTE samples at the beginning (Left) and ending (Right) of the vending day. Ch. Ch. S. = chicken with chili sauce.

At the end of marketing day, it was observed that coliforms growth has significantly increased in all the samples. Chicken and beef curry from the night market had the maximum coliform growth (1100 MPN/g) at the end of the marketing day while fried chicken from the UKM cafeteria had the least coliforms, (35.5 MPN/g), compared to all other RTE samples from all sampling sites (Figure 2B). It was expected that both chicken and beef curry would incur heavy growth of coliforms and other microorganisms, because these positions were open to different sources of contamination such as cars exhausts, rising dusts and garbage which provide a hiding place for insects and animal pests (Bryan et al. 1997).

Escherichia coli

E. coli was not found in the cafeteria of UKM at the beginning of the food sold (Figure 3A). Moreover, neither fried chicken nor chicken curry from all sampling sites contained *E. coli* at the beginning of vending day which is comparable to Meldrum et al. (2006), who found no *E. coli* in RTE burgers, pasty meat and pate meat in the UK. Chicken curry from the Bandar Baru Bangi night market and Kajang restaurants both had an average of 13.5 MPN/g whereas, beef curry had *E. coli* counts of 48.5 and 16 MPN/g in the night market and in Kajang, respectively.

Beef curry had the highest *E. coli* count (240 MPN/g), averaging 131.5 MPN/g for samples obtained at the end of the night market (Figure 3B).

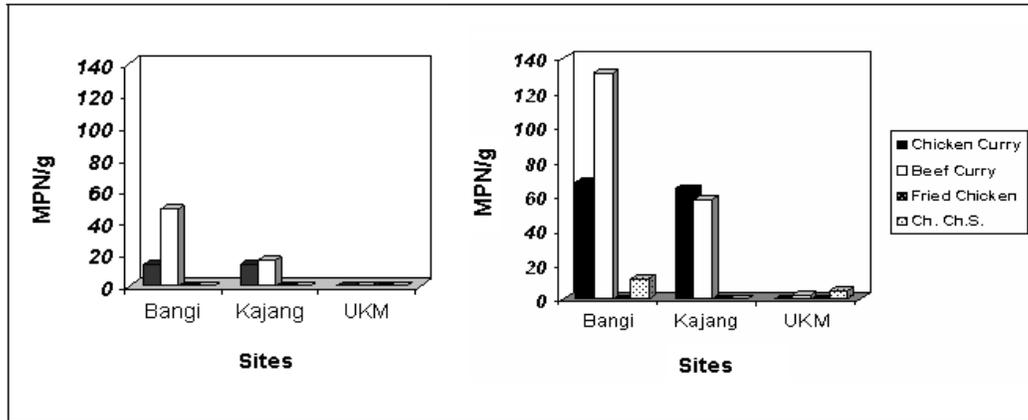


Figure 3 Average *Escherichia coli* counts at the beginning (Left) and ending (Right) of the vending day. Ch. Ch. S. = chicken with chili sauce

Results of the analysis showed the different between the beef curry and other samples collected from the three sites were significantly different ($P < 0.05$). This is in agreement with studies by Viswanathan and Kaur (2000) which suggested that food borne *E. coli* is found and transmitted mainly in food derived from cows such as raw milk, raw or rare ground beef and fruit or vegetables that are contaminated.

Salmonella spp.

This study is in agreement with the study reported by Soriano et al. (2001) who found no *Salmonella* in raw and RTE samples from 20 University restaurants in Valencia, Spain. Umoh and Odoaba (1999) also indicated that none of the RTE food samples from mobile food sellers were contaminated with *Salmonella*, similar with results of research done by Aycicek et al. (2004) in a military hospital in Ankara, Turkey. Meldrum et al. (2006) from the United Kingdom also revealed the absence of this pathogen during the investigation of the microbiological quality of RTE foods between 2003 and 2005 in Wales. The absence of *Salmonella* in these studies indicated that good handling practices during the cooking process and good storage facilities were available for both raw and RTE meat and poultry items used in the meals.

Coagulase-positive Staphylococci

Like *Salmonella*, coagulase-positive Staphylococci were not found in all the examined samples. Similarly, Hanashiro et al. (2005) stated that Staphylococci were not detected in home- and street-prepared RTE meal samples in Sao Paulo, Brazil.

Compliance Of Samples With Microbiological Criterion

Beginning of the Vending Day

The non-compliance percentages of various types of RTE foods in the Hulu Langat District sampled at the beginning of the marketing day in three defined sampling sites to acceptable microbiological standards are shown in Figure 4. Very few of the meal samples at the beginning of the marketing day exceeded the microbiological guideline/standards except the TPC count. In terms of TPC, the samples of beef curry, chicken with chili sauce, fried chicken and chicken curry gave 100 %, 83.3%, 33.3% and 16.7 % non-compliance with microbiological criterion respectively. However, the percentage of non-compliance with total coliforms limits was much lower - 16.7% which accounts for 1 beef curry sample. Overall, 95.8% (23 out of 24) samples complied to refer total coliforms guideline/standards. That means only 4.2% of the samples exceeded the acceptable limits for coliforms. This is much lower than results obtained by Fang et al. (2003) who observed that 80% of food samples containing meat failed to meet the standard for total coliforms.

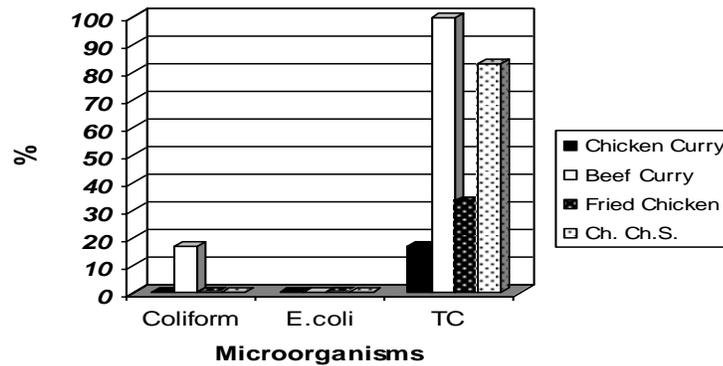


Figure 4 Percentage (%) non-compliance of RTE meals collected at the beginning of the marketing day with the microbiological guideline/standards imposed by the Ministry of Health of Malaysia (MOH) and other international standards. Ch. Ch. S. = chicken with chili sauce.

On other hand, none of the samples failed to meet the standards set for *E. coli*, *Salmonella* and coagulase-positive Staphylococci. A study by Fang et al. (2003), 10% and 12% of the RTE meat products were observed to exceed the Chinese government standard for *E. coli* and *S. aureus*, respectively. The percentages of non-compliant samples in this study suggest that the meals sampled in the Hulu Langat District were of high microbiological quality, consequently as properly prepared, stored and handled

End of the Vending Day

50% (3 out of 6) and 33.3% (2 out of 6) of beef and chicken curry samples, respectively, were unacceptable to the three referred guideline/standards; i.e. the MOH guideline, the Argentina (food code: article 442) and French (order 26/6/74) regulations for total coliforms ($10^3/g$). On the other hand, both fried chicken and chicken curry with chilly sauce were 100% acceptable according to *E. coli* and total coliforms reference guideline/standards at the end of the marketing day. However, 1 out of 6 samples of beef curry and chicken curry were non-compliant to acceptable *E. coli* guideline/standards ($> 10^2$ MPN/g) (Figure 5).

The graph of *Salmonella* and coagulase-positive Staphylococci were not showed as none of them were isolated in this study. The absence of these pathogens in all the samples indicate high microbiological quality of the food, as discussed above. The above mentioned results were consistent with those obtained from the study conducted by Aycicek et al. (2004) who evaluated total coliforms and *E. coli* according to the Turkish Standard Institution.

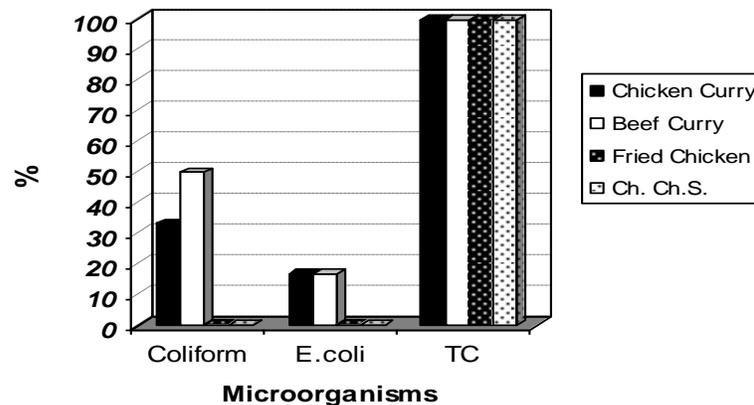


Figure 5. Percentage (%) non-compliance of RTE meals collected at the end of the marketing day with the microbiological standards imposed by the Ministry of Health of Malaysia (MOH) and other international standards. Ch. Ch. S. = chicken with chili sauce.

CONCLUSION

The change of bacterial growth at the beginning where RTE meals started to be sold and the end of all the sites has been studied. The growth of TPC for the sites were not significantly different ($P>0.05$) between the beginning and end whereas, total coliforms and *E. coli* growth were significant ($P>0.05$). Possibly, cross-contamination occurred to the foods between these intervals at some of the sites. However, based on the acceptable level of the microbiological guideline/standards, only the total plate count was exceeded in both samples collected at intervals

of marketing times. This study indicates several things such as good constitution accomplished by the Malaysian government to regulate vending safe foods, implementation of these regulations was acceptable during the inspection and other monitoring system and most of the RTE vendors, observed proper preparation and handling of the RTE foods and able to safeguard from different contaminations.

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