

# Food hygiene and safety statuses of common commercial meals within the capital city of Yilo Krobo Municipal in Eastern Region of Ghana

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## Abstract

Food-borne intestinal and diarrheal diseases caused by enteropathogenic bacteria (EPB) appear to be an emerging rural health challenge across Ghana. Using *Shigella-Salmonella* agar, colony-forming units (CFUs) of EPB in some commercial meals within the capital city of Yilo Krobo Municipal in Ghana were examined. To determine food hygiene and safety statuses, counts of CFUs of EPB were compared to their acceptable limits (ALs) set by food regulatory authorities (FRAs) in European Union, United States of America, Ghana, Ireland, China, Canada and United Kingdom, using a non-parametric model. Colonies of *Proteus* species, *Escherichia coli*, *Shigella* species and *Salmonella* species were observed on all tested commercial meals, except 'kenkey without stew'. Numbers of CFUs of *E. coli* on all tested meals, except *waakye*, were not significantly ( $P > 0.05$ ) greater than ALs set by all international FRAs. However, counts of CFUs of *E. coli* on all the meals tested, including *waakye*, were not significantly ( $P > 0.05$ ) greater than ALs set by a national FRA only. In terms of *Proteus* species on all meals, low counts of CFU were largely not significantly ( $P > 0.05$ ) greater than ALs set by both national and international FRAs. Conceivably, all the tested meals, which had counts of CFUs of either *E. coli* or *Proteus* species to be similar to values of ALs, received satisfactory food hygiene treatments. Moreover, counts of CFUs of *Shigella* spp. on all the tested meals were not significantly ( $P > 0.05$ ) larger than the ALs set by both national and international FRAs, thereby suggesting that these meals might be safe. Notwithstanding, lack of incidence of *Salmonella* species in any ready-to-eat meals has always been the strictest irrefutable policy by all FRAs worldwide. Therefore, perspectives to achieve improvement and sustainability in the future have been discussed.

## Keywords

Food hygiene; Food safety; Food-borne bacterial contaminations; Public exposure; Perspectives

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## 1. Introduction

Intestinal and diarrheal diseases caused by diverse enteropathogenic bacteria appear to be an emerging endemic rural health challenge in Ghana (Krumkamp et al., 2015; Akuffo et al., 2017; Nyadanu et al., 2017; Fusheini and Gyawu, 2020; SVI, 2021; Apenteng et al., 2023). *Vibrio cholerae*, *Proteus* spp., *Shigella* spp. and *Salmonella* spp., among others, are the most common harmful enteric bacteria detected for causing infectious intestinal diseases among categories of people in Ghana (Krumkamp et al., 2015; Akuffo et al., 2017; Nyadanu et al., 2017; Fusheini and Gyawu, 2020; SVI, 2021; Apenteng et al., 2023). Consequently, a report by SVI (2021) had established that children, who are  $\leq 15$ -year-old, represent at least 56% of people who are often infected by dangerous enteropathogenic bacteria across districts, municipalities and metropolitans in Ghana.

Intestinal disease-causing bacteria are more associated with lack of good practices for environmental sanitation and/or food hygiene (Odonkor and Mahami, 2020; Parker et al., 2021; Popa and Papa, 2021; Jamil et al., 2023; WHO, 2023a; CDC, 2024). Thus, various age-groups of people will often acquire infectious microbial entities through consumption of contaminated meals, drinks and fruits in cities, towns, villages, communities, suburbs, among others, where waste management and sanitation are poor. Because of these pieces of information in literature, food hygiene and safety statuses of some common kinds of food, being sold by vendors to the public across suburbs of the capital city of the Yilo Krobo Municipal, were assessed in southeastern Ghana.

A highly selective growth medium, similar to those in recent reports of Omorodion and Wokoma (2021), Aryal (2022) and Berihu et al. (2024), was used to correctly examine and differentiate between bacterial colony-forming units of *Proteus* spp., *Escherichia coli*, *Shigella* spp. and *Salmonella* spp. in food samples, which were randomly bought from vendors across some major suburbs of the capital city of the Yilo Krobo Municipal. Relative to the acceptable limits set by numerous national and international food regulatory authorities for ready-to-eat meals in reports, counts of colony-forming units of *Proteus* species and *E. coli* on food samples were used to assess food hygiene status of commercially cooked food samples from the major suburbs of the capital city of the municipal, similar to the way it was done in previous reports (Feglo and Sakyi, 2012; Darko et al., 2015; Kortei et al., 2020; Lente et al., 2022; Osei-Owusu et al., 2023). Similarly, acceptable limits of *Shigella* spp. and *Salmonella* spp. on food samples were used to determine safety status of these commercial meals sold to the public across the

- assess incidence of some selected food-borne enteropathogenic bacteria in the most common commercial food sold within the capital city of Yilo Municipal;
- compare levels of detectable enteric infectious bacteria to their acceptable limits, set by both national and some selected international food regulatory authorities (FRAs);
- confirm whether the most common commercially cooked food (or meals) within the municipal's capital city do likely receive the acceptable treatments for food hygiene and safety standards, set by national and some selected international food regulatory bodies for consumption by the public;
- Suggest some viewpoints for any possible improvement and sustainability.

## 2. Materials and Methods

### 2.1 Preparation of a differential growth medium for enteropathogenic bacterial colonies in food samples

*Shigella* – *Salmonella* agar (SSA) (Oxoid<sup>TM</sup>) was used as a selective and/or differential growth medium, which allows the growth of differentiating colonies of *Salmonella*, *Proteus*, *Escherichia coli* and *Shigella* (Aryal, 2022; Ruiz Gomez et al., 1998). The SSA was prepared according to the manufacturer's instructions under aseptic conditions. Each of the Petri-dishes containing about 20 mL of solidified prepared agar was securely stored in an incubator aseptically at 25 – 27 °C until being used for the tests within three (3) days, similar to the procedures used in recent reports (Odonkor and Mahami, 2020; Osei-Owusu et al., 2023).

### 2.2 Food samples from selected suburbs of Somanya town in the Yilo Krobo Municipal

The Yilo Krobo Municipal occupies a surface area of 514.7 km<sup>2</sup> in the Eastern Region of Ghana (Brinkhoff, 2024). By 2021, this municipal had a population of 122,705 inhabitants (Brinkhoff, 2024; GSS, 2024). Its administrative and commercial capital town is Somanya, which is located at the geographical coordinate (6°06'14" N, 0°00'54" W). Somanya township has many highly populated suburbs, but some of the most popular ones include Trome-Somanya, Round-about-Somanya, Ogome-Somanya and the University of Environment and Sustainable Development (UESD).

Even though there had not been any previous reports on food contaminations by virulent enteric bacteria across Krobo municipalities, reports of Feglo and Sakyi (2012),

Darko et al. (2015) and Kortei et al. (2020) suggest that similar evidences on enteropathogenic bacterial contaminations of commercial meals in Accra and Kumasi metropolitans can be widespread in the streets across suburbs of other towns and cities in Ghana. Therefore, food samples were taken separately from the four (4) aforementioned suburbs within Somanya township in the Yilo Krobo municipality of Eastern Region in Ghana. The choice of these suburbs as sources of food samples for this study was guided by the fact that the selected suburbs are densely populated locations, where people's activities are higher in greater parts of a week. The targeted food samples were those that were commonly sold (i) in open-space, where shade was not available, (ii) in a shade of a huge tree, or (iii) in poorly constructed mini-kiosks. Commercially prepared meals considered for this study included (1) porridge, (2) sugar, which is usually added to porridge, (3) *kenkey*, (4) *kenkey* stew, (5) *waakye* (or cooked rice and beans) and (6) *waakye* stew. Thus, samples of each of these food materials were randomly bought from the sellers in each selected suburb in the Somanya township for testing to isolate, identify and quantify colony-forming units of enteropathogenic bacteria, following plating food samples on the differential SSA growth medium in Petri-dishes (diameter = 9 cm).

**2.2.1 Testing food samples for isolation and identification of enteropathogenic bacteria on solidified *Shigella-Salmonella* agar in Petri-dishes**

The food samples were brought to a laboratory and then immediately prepared for testing. According to suggestions by food regulatory authorities (FRAs in Table 1) for United States of America, Canada, Ghana, European Union, Ireland, United Kingdom and China, about 25 g of a food sample is apposite for analysis of food-borne illnesses. However, in this study, our preliminary tests showed that five (5) g of each homogenized food sample was satisfactorily revealing detectable and countable levels of colony-forming units of enteropathogenic bacteria in SSA. Therefore, five (5) g of each food sample was prepared into suspension under aseptic conditions, using 10 mL of an autoclaved distilled water. About one (1) mL of the prepared 10 mL stock suspension was diluted by either 10 or 100 times, depending on the cloudiness of the initially-prepared stock suspension from a food sample. About 500  $\mu$ L of the final diluted suspension of a food sample was plated on a solidified SSA in a Petri-dish, using a micropipette.

A control was made by pipetting 500  $\mu$ L of autoclaved distilled water only onto SSA in Petri-dishes. Including the set-ups for control, twelve (12) replicates were made for each food sample taken across suburbs. After that, treatments on SSA in Petri-dishes were incubated at 25 – 27 °C until inspected after 24 hours (hrs) for identification of differentiating colours of bacterial colonies, similar to the way it was done by Omorodion and Wokoma (2021).

**Table 1.** Acceptable limits of enteropathogenic bacteria used to examine food hygiene and safety statuses of cooked food or ready-to-eat meal.

Food regulatory body	Acceptable limit of the enteropathogenic bacteria in cooked food or ready-to-eat meal				Source of information
	<i>Proteus</i> spp. (CFU/g)	<i>E. coli</i> (CFU/g)	<i>Shigella</i> spp. (CFU/g)	<i>Salmonella</i> spp. (CFU (25/g))	
EU	10,000	700	-	0	WBG (2017); FSAI (2018); UKHSA (2024).
USA	-	1000	-	0	IMNRC (2003); NLM (2024)
GSB	-	100,000	100,000	0	Feglo and Sakyi (2012); Kortei et al. (2020)
UKHSA	-	-	-	0	UKHSA (2024)
FSAI	10,000	1000	-	0	FSAI (2018); NLM (2024)
China	-	1000	-	0	WBG (2017)
CS	-	1000	-	0	NLM (2024)

EU = European Union; USA = United States of America; UKHSA = United Kingdom Health Security Agency; GSB = Ghana Standard Board; FSAI = Food Safety Authority of Ireland; CS = Canadian Standard.

However, incidence and growth of bacterial colonies on SSA in Petri-dishes were examined every 24 hrs until 72nd hr elapsed to confirm evidence of growth of bacterial colonies in SSA.

### 2.2.2 Quantitation of enteropathogenic bacterial colonies from plated food samples on *Shigella–Salmonella* agar in Petri-dishes

The identified colonies were counted in each SSA plate. After that, the total number ( $N_{CFU}$ ) of identified enteropathogenic bacterial colony-forming units (CFU) per g of food sample was estimated using the equation 1 below, similar to the quantification procedures used in previous reports (Omorodion and Wokoma, 2021; Osei-Owusu et al., 2023):

$$N_{CFU} = ([CFU_{plate}] - [CFU_{control}]) \left( \frac{V_{stock}}{V_{plated}} \right) \left( \frac{n}{5g} \right) \quad (1)$$

Where,  $N_{CFU}$  is the total estimated number of bacterial colonies per gramme (g) of food sample;  $CFU_{plate}$  is the number of CFUs counted on the SSA per Petri-dish;  $CFU_{control}$  is the mean value of numbers of CFUs observed in control set-ups;  $V_{stock}$  is the volume of the stock suspension made from five (5) g of a food sample;  $V_{plated}$  is 500  $\mu$ L of the diluted or undiluted suspension, which was plated on SSA in a Petri-dish, using a micropipette;  $n$  is the number of times one (1) mL of the stock suspension was diluted;  $g$  is the gramme of food sample taken for the test.

## 2.3 Data analyses

### 2.3.1 Data analyses

R software [R.v.4.4.1 RStudio.v.SHA-2562c3cf96a] for programming was used for all analyses (R Core Team, 2024). After using *Shapiro-Wilk* test to confirm that data were non-parametric (i.e., not normally distributed), *Dunn's* test at  $P \leq 0.05$  was used to compare 'mean  $\pm$  standard error (SE)' of variables between the food samples, with the control (Gotelli and Ellison, 2013; Dinno, 2015). After that, bar graphs were used to present the 'mean  $\pm$  SE' values of bacterial CFUs for food samples.

## 2.4 Determination of food hygiene and safety statuses of commercial meals

R software [R.v.4.4.1 RStudio.v.SHA-2562c3cf96a] developed by R Core Team (2024) was also used for analysis here. *Shapiro-Wilk* test was first used to confirm normality status of each set of data, which proved requiring a non-parametric statistic, according to several previous reports (Gotelli and Ellison, 2013; Dinno, 2015; Lente et al., 2022; Heve et al. 2023; Osei-Owusu et al., 2023; Larbi et al., 2024). Therefore, hygiene and safety statuses of cooked food or ready-to-eat commercial meals were examined by using *Wilcoxon rank-sum* test to compare 'mean  $\pm$  SE' value of CFUs of each enteropathogenic bacterium

to the acceptable limits, set by national and some selected international food safety regulatory organizations (Table 1). This approach used in this study was similar to the way it was done in previous reports (Lente et al., 2022; Osei-Owusu et al., 2023). However, counts of CFUs of *Salmonella* species were not compared to the acceptable limit, which is zero (0) per 25 g of food sample in Table 1. This was largely because 'mean  $\pm$  SE' value of CFUs of *Salmonella* species was detectable in five (5) g of each of many food samples.

## 3. Results

### 3.1 Colony-forming units of *Proteus* species on food samples in *Shigella–Salmonella* agar

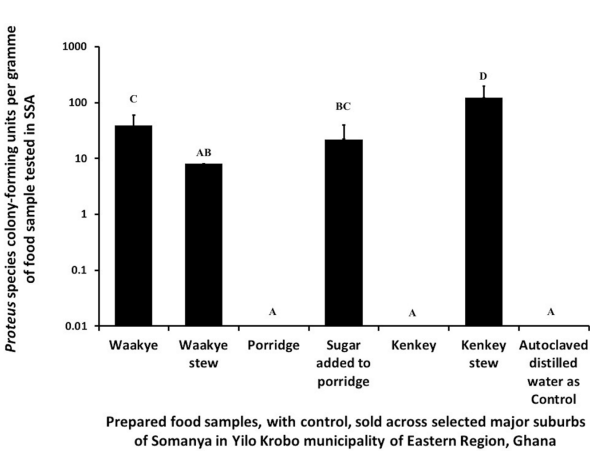
*Kenkey*, porridge and the 'control' had no colony-forming units (CFUs) of *Proteus* species (Figure 1). However, colonies of *Proteus* species were observed on *waakye*, *waakye* stew, sugar (normally added to porridge) and *kenkey* stew (Figure 1). Even though the numbers of CFUs of *Proteus* species on *waakye* stew, porridge, sugar and *kenkey* were not significantly ( $P \geq 0.05$ ) different from 'none' observed in the control, tens of CFUs were counted on *waakye* and sugar (Figure 1). Similarly, there was no significant difference ( $P \geq 0.05$ ) between mean $\pm$ SE-values of numbers of *Proteus* species CFUs on *waakye* and sugar (Figure 1). Notwithstanding, the numbers of *Proteus* species CFUs on *kenkey* stew alone were in hundreds, which were significantly ( $P \leq 0.05$ ) greater than those observed on *waakye*, *waakye* stew or sugar (Figure 1). The mean $\pm$ SE values of CFUs of *Proteus* spp. (in Figure 1) were compared to the acceptable limit (Table 1) set by the European Union (EU) and the Food Safety Authority of Ireland (FSAI). These mean $\pm$ SE values of CFUs of *Proteus* spp. on *waakye*, *waakye* stew, sugar and *kenkey* stew were similar to the acceptable limit (Table 2). However, the mean $\pm$ SE values of CFUs of *Proteus* spp. on porridge and *kenkey* were significantly ( $P \leq 0.05$ ) lower than the acceptable limit (Table 2).

**Table 2.** Wilcoxon rank-sum tests used to compare ‘mean±SE’ values of CFUs of *Proteus* spp. to the acceptable limit in order to examine food hygiene status of cooked food or ready-to-eat meal sold within the capital city (i.e., Somanya) of Yilo Krobo Municipal in Ghana.

Food regulatory bodies or countries	Acceptable limit for CFU of <i>Proteus</i> spp.	Mean $\pm$ SE-value of CFUs of <i>Proteus</i> spp. in each of the food samples	P-value of <i>Wilcoxon rank-sum</i> test for comparing the acceptable limit to the 'mean $\pm$ SE-value' of CFUs of <i>Proteus</i> spp. in each of the food samples.						Decision based on food safety analysis
			<i>Waakye</i>	<i>Waakye</i> stew	Porridge	Sugar added to porridge	<i>Kenkey</i>	<i>Kenkey</i> stew	
EU; FSAI	Table 1	Figure 1	0.25 ¶	0.5 ¶	0.005962 **	0.5 ¶	0.005962 **	0.09751 ¶	All food samples appeared to be acceptable

EU = European Union; FSAI = Food Safety Authority of Ireland.  
CFUs = Colony-forming units. SE = Standard error.

Wilcoxon rank-sum test: The symbol ‘¶’ indicates that the ‘mean ±SE’ value of CFUs of *Proteus* spp. in the food sample was significantly lower than the acceptable limit, set by EU and FSAI. According to Wilcoxon rank-sum test, \*\* denotes that the ‘mean ±SE’ value of CFUs of *Proteus* spp. in the food sample was significantly lower than the acceptable limit, set by EU and FSAI, at  $P \leq 0.01$ .  
CFUs = Colony-forming units, SE = Standard error.



**Figure 1.** Numbers of colony-forming units (CFUs) of *Proteus* species per gramme of each of the selected food samples plated in *Shigella–Salmonella* agar. According to Dunn’s test, different letters on top of bars indicate that values of ‘mean ± SE’ are significantly different at  $P \leq 0.05$ .

**3.2 Colony-forming units of *Escherichia coli* on food samples in *Shigella–Salmonella* agar**

Only kenkey had no CFU of *E. coli*, similar to ‘none’ in ‘control’ (Figure 2). However, colonies of *E. coli* were observed on waakye, waakye stew, porridge, sugar and kenkey stew (Figure 2). The number of CFUs of *E. coli* on porridge or sugar was significantly ( $P \leq 0.05$ ) different from ‘none’ observed on kenkey (Figure 2). Tens of CFUs of *E. coli* were enumerated on either porridge or sugar (Figure 2). However, the numbers of *E. coli* CFUs on kenkey stew, waakye stew and waakye were almost in thousands, ten thousands and hundred thousands (Figure 2), respectively. The latter was significantly ( $P \leq 0.05$ ) the highest (Figure 2). The mean±SE value of CFUs of *E. coli* on waakye was significantly ( $P \leq 0.05$ ) larger than the acceptable limits set by EU, USA, FSAI, China and the Canadian Standard (CS) (Figure 2; Tables 1 and 3), although it was similar to the acceptable limit set by the Ghana Standard Board (GSB) (Tables 1 and 3). Nonetheless, the numbers of CFUs of *E. coli* on waakye stew, porridge, sugar, kenkey or kenkey stew (in Figure 2) were within the ranges of all the acceptable limits set by EU, USA, FSAI, China, CS and GSB (Tables 1 and 3).

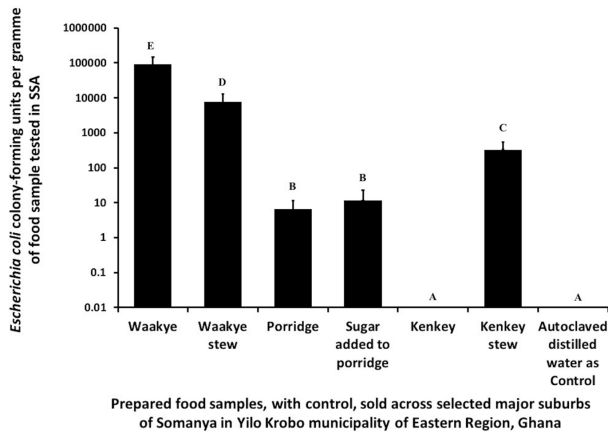


**Table 3.** *Wilcoxon rank-sum* tests used to compare ‘mean  $\pm$ SE’ values of CFUs of *E. coli* to the acceptable limit in order to examine food hygiene status of cooked food or ready-to-eat meal sold within the capital city (i.e., Somanya) of Yilo Krobo Municipal in Ghana.

Food regulatory bodies or countries	Acceptable limit for CFU of <i>E. coli</i>	Mean $\pm$ SE-value of CFUs of <i>E. coli</i> in each of the food samples	P-value of t- test for comparing the acceptable limit to the ‘mean $\pm$ SE-value’ of CFUs of <i>E. coli</i> in each of the food samples.						Decision based on food safety analysis	
			<i>Waakye</i>	<i>Waakye</i> stew	Porridge	Sugar added to porridge	<i>Kenkey</i>	<i>Kenkey</i> stew		
EU	Table 1	Figure 2	0.01953 *	0.05581 ¶	0.01038 *		0.01471 *	0.00063 ***	0.178 ¶	Only <i>Waakye</i> appeared ‘not acceptable’
USA; FSAI; China; CS	Table 1	Figure 2	0.02439 *	0.1971 ¶	0.01038 *		0.01471 *	0.00063 ***	0.03981 *	Only <i>Waakye</i> appeared not acceptable
GSB	Table 1	Figure 2	0.1055 ¶	0.00384 **	0.01038 *		0.01471 *	0.00063 ***	0.0131 *	All food samples appeared to be acceptable

EU = European Union; USA = United States of America; FSAI = Food Safety Authority of Ireland; GSB = Ghana Standard Board; CS = Canadian standard.  
CFUs = Colony-forming units. SE = Standard error.

According to *Wilcoxon rank-sum* test, \* and \*\*\* denotes that the ‘mean  $\pm$ SE’ value of CFUs of *E. coli* in the food sample was significantly different from the acceptable limit at  $P \leq 0.05$  and  $P \leq 0.001$ , respectively. However, the symbol ‘§’ indicates that ‘mean  $\pm$ SE’ value of CFUs of *E. coli* in *waakye* was significantly larger than the acceptable limit at  $P \leq 0.05$ . The symbol ‘¶’ indicates that the ‘mean  $\pm$ SE’ value of CFUs of *E. coli* in the food sample was similar to the acceptable limit.



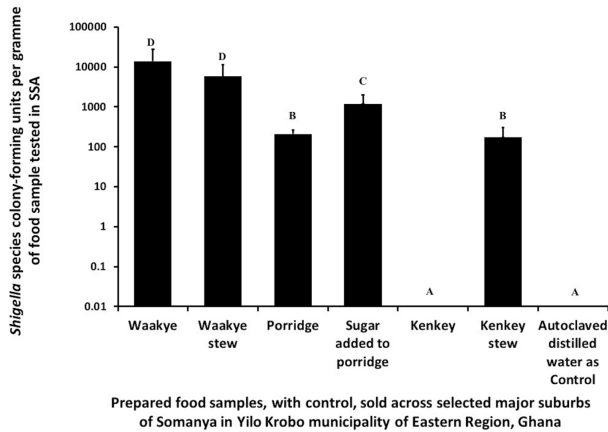
**Figure 2.** Numbers of colony-forming units (CFUs) of *E. coli* per gramme of each of the selected food samples plated in *Shigella–Salmonella* agar. *Dunn’s* test at  $P \leq 0.05$ : Different letters on top of bars indicate that the values of ‘mean  $\pm$  SE’ were significantly different.

### 3.3 Colony-forming units of *Shigella* species on food samples in *Shigella–Salmonella* agar

No CFUs of *Shigella* species were observed both on *kenkey* and in ‘control’ (Figure 3). However, the mean $\pm$ SE-values of CFUs of *Shigella* species on porridge and *kenkey* stew were in hundreds, whereas that of sugar was in thousands (Figure 3). Both *waakye* and *waakye* stew had the highest mean $\pm$ SE-values of CFUs of *Shigella* species in ten thousands (Figure 3). Thus, the mean $\pm$ SE-value of *Shigella* species CFUs observed on each food sample was significantly ( $P \leq 0.05$ ) different from the other, except that those of either ‘*waakye* and *waakye* stew’ or ‘porridge and *kenkey* stew’ were similar (Figure 3).

The mean $\pm$ SE values of CFUs of *Shigella* species on *waakye*, *waakye* stew, porridge, sugar and *kenkey* stew (in Figure 3) were similar to the acceptable limit set by GSB (Tables 1 and 4). However, the mean $\pm$ SE value of CFUs of *Shigella* species on *kenkey* (Figure 3) was significantly ( $P \leq 0.01$ ) less than the acceptable limit set by GSB

(Table 4).



**Figure 3.** Numbers of colony-forming units of *Shigella* species per gramme of each of the selected food samples plated in *Shigella–Salmonella* agar. *Dunn’s* test at  $P \leq 0.05$ : Different letters on top of bars denote that those values of ‘mean  $\pm$  SE’ were significantly different.

**Table 4.** Wilcoxon rank-sum tests used to compare ‘mean  $\pm$ SE’ values of CFUs of *Shigella* spp. to the acceptable limit in order to examine food safety status of cooked food or ready-to-eat meal sold within the capital city (i.e., Somanya) of Yilo Krobo Municipal in Ghana.

Food regulatory body or country	Acceptable limit for CFU of <i>Shigella</i> spp.	Mean $\pm$ SE-value of CFUs of <i>Shigella</i> spp. in each of the food samples	P-value of Wilcoxon rank-sum test for comparing the acceptable limit to the ‘mean $\pm$ SE-value’ of CFUs of <i>Shigella</i> spp. in each of the food samples.						Decision based on food safety analysis
			Waakye	Waakye stew	Porridge	Sugar added to porridge	Kenkey	Kenkey stew	
GSB	Table 1	Figure 3	0.5 ¶	0.125 ¶	0.25 ¶	0.5 ¶	0.006 **	0.09751 ¶	All food samples appeared to be acceptable

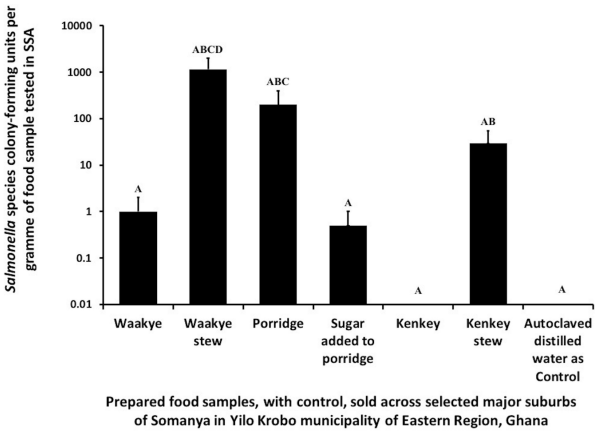
GSB = Ghana Standard Board.  
CFUs = Colony-forming units. SE = Standard error.

Wilcoxon rank-sum test: The symbol ‘¶’ indicates that the ‘mean  $\pm$ SE’ value of CFUs of *Shigella* spp. in the food sample was similar to the acceptable limit set by GSB. According to Wilcoxon rank-sum test, \*\* denotes that the ‘mean  $\pm$ SE’ value of CFUs of *Shigella* spp. in the food sample was significantly lower than the acceptable limit, set by GSB, at  $P \leq 0.01$ .

### 3.4 Colony-forming units of *Salmonella* species on food samples in *Shigella*–*Salmonella* agar

Similar to observations in Figures 1, 2 and 3, no CFUs of *Salmonella* species were observed both on *kenkey* and in ‘control’ (Figure 4). Moreover, the mean $\pm$ SE-value of CFUs of *Salmonella* species on each of the remaining food samples was not significantly ( $P > 0.05$ ) larger than that observed on *kenkey* (Figure 4).

These were largely so, because no CFUs of *Salmonella* species could be observed in the majority of the replicates for each of the food samples. However, the ‘mean  $\pm$  SE’ values of CFUs of *Salmonella* species on all the food samples, except *kenkey* in Figure 4, exceeded the acceptable limit (i.e., the ‘zero CFU policy’ in Table 1) set by all the food regulatory authorities in EU, USA, Ghana, Ireland, China, Canada and UK.



**Figure 4.** Numbers of colony-forming units of *Salmonella* species per gramme of each of the selected food samples plated in *Shigella*–*Salmonella* agar. Dunn’s test at  $P \leq 0.05$ : The same letters on top of bars denote that those values of ‘mean  $\pm$  SE’ were not significantly different.

### 4.1 *Shigella*–*Salmonella* agar for isolating enteropathogenic bacteria in samples

To large extent, bacterial colonies in the differential growth medium have been highly selected, because only specific genera of bacteria have often been targeted in a selective growth medium (Uyttendaele et al., 2001; Tosisa et al., 2020; Amare et al., 2024). Thus, selective and differential solid growth media are widely used to confirm contamination of clinical specimens, water, milk, chocolate, beverages, exportable food products, imported foodstuff and prepared food, among others, by harmful virulent enteric bacteria to humans and animals (Uyttendaele et al., 2001; Tosisa et al., 2020; Amare et al., 2024). Moreover, these solid bacterial growth media have been useful to grow, identify and quantify colonies of infectious enteric bacteria present in samples (Mulatu et al., 2014; Tosisa et al., 2020; Omorodion and Wokoma, 2021; Amare et al., 2024). In this study, appearance of enteropathogenic bacterial colonies was not blurring in *Shigella*–*Salmonella* agar (SSA). This could largely be due to the fact that the specific differentiating colours of different bacterial colonies in SSA were visibly distinguishable from one another, similar to observations in previous reports (Mulatu et al., 2014; Tosisa et al., 2020; Omorodion and Wokoma, 2021).

The SSA was user-friendly, thereby allowing isolation, identification and quantification of enteropathogenic bacterial colony-forming units in samples of selected food stocks from across the major suburbs of Somanya in the Eastern Region of Ghana. Although only four (4) enteropathogenic bacteria (i.e., *Proteus* spp., *E. coli*, *Shigella* spp. and *Salmonella* spp.) could clearly be assessed in plates of SSA, the details were informative in this study. Nevertheless, additional analytical methods for food-borne illnesses may be more useful to examine incidence of many more dangerous enteropathogenic bacteria in commercial food samples from different locations throughout Krobo Municipals in the future.

## 4. Discussion

#### 4.2 Incidence of enteropathogenic bacteria in selected commercial meals: hygiene and safety statuses

Prokaryotic entities commonly contaminate surfaces, air, water, materials, plants, animals and humans, among others, because they are highly ubiquitous in the biosphere (Parker et al., 2021). Consequently, the majority of the enteropathogenic bacteria had often been detected in drinks, fruits, salads, vegetables, foodstuffs, processed food in packages and many more ready-to-eat meals (Feglo and Sakyi, 2012; Darko et al., 2015; Kortei et al., 2020; Lente et al., 2022; Osei-Owusu et al., 2023). Thus, higher counts of CFUs of several enteropathogenic bacteria than their permissible limits had been observed in the majority of commercial meals sold by street-food-vendors and hotels across two major Ghanaian metropolises (Feglo and Sakyi, 2012; Darko et al., 2015; Kortei et al., 2020). Unfortunately, commercial meals sold in the streets (or in open spaces) across districts, municipals, and metropolises in Ghana appear to be readily available and affordable to the public (*authors' personal observations*). From practical standpoint, the public will be more exposed to these commercial meals, which are widely contaminated by numerous food-borne infectious enteropathogens.

Nonetheless, similar to all other food samples in this study, kenkey is normally prepared at high temperatures for at least two (2) hours (*authors' personal experience*). During selling, food vendors in the study area practically employ local techniques to significantly reduce loss of heat from bolls of kenkey and other meals in containers so that these ready-to-eat cooked food stocks are sold to the public when they are still hot over extended period (*authors' personal observations*). Fortunately, all the enteropathogenic bacteria observed in this study are not known to be tolerant of the boiling point of water (Parker et al., 2021). Possibly, high amount of heat during cooking and selling could kill and reduce any infectious enteropathogens in these cooked meals. Nevertheless, according to Oduro-Yeboah et al. (2018), a boll of *kenkey* is preferably often consumed with *kenkey* stew, from which we detected colonies of *Proteus* spp., *E. coli*, *Shigella* spp., and *Salmonella* spp., similar to the way almost all these bacteria were observed in *waakye*, *waakye* stew, porridge, *kenkey* stew and sugar (which is normally added to porridge). The presence of these enteropathogenic bacteria in these food samples, except commercial *kenkey* meal, should create panic concerns to consumers and food regulatory institutions.

Nonetheless, analytical assessment that compared the estimated CFUs of *Proteus* spp. to the acceptable limits in literature appeared to prove that all the selected commercially cooked food samples might have received satisfactory food hygiene treatments that are acceptable by food regulatory bodies in Ghana, UK, China, USA, Ireland, Canada and Europe. Also, when similar analytical assessment compared incidence of *E. coli* on food

samples to the acceptable limits in literature, the results showed that *waakye* stew, porridge, *kenkey*, *kenkey* stew and sugar might have received satisfactory food hygiene treatments that could possibly be acceptable by Ghana, UK, China, USA, Ireland, Canada and Europe. However, the acceptable limit set by the Ghana Standard Board (GSB), when compared to those of the food regulatory institutions in UK, China, USA, Ireland, Canada and Europe, for *E. coli* on ready-to-eat meals suggests that the commercially cooked 'beans-and-rice' known as *waakye* might have received satisfactory food hygiene treatments that are acceptable by Ghana only (IMNRC, 2003; WBG, 2017; FSAI, 2018; Feglo and Sakyi, 2012; Kortei et al., 2020; NLM, 2024; UKHSA, 2024). Perhaps, high amount of beans added to rice in preparing this meal (i.e., *waakye*) could support rapid growth of *E. coli* colonies. This is because recent observations have proved that beans of legumes are highly effective growth media for *E. coli* and several other bacteria (Purnawati et al., 2023; Condriillon et al., 2024). Moreover, beans of legumes contain high amount of proteins, which could be used by growing *E. coli* colonies within commercially cooked 'waakye environment'. Thus, a rapid growth of *E. coli* or similar enteric bacteria, belonging to the family Enterobacteriaceae, in commercially cooked 'beans-rich' meals may provide misleading information on food hygiene status.

Enteropathogenic bacterial species belonging to the genera *Shigella* and *Salmonella* have high health risks to humans, because they largely cause shigellosis and severe typhoid fever in humans, especially in younger ones (Fusheini and Gyawu, 2020; Kortei et al., 2020; Parker et al., 2021; SVI, 2021). Consequently, large numbers of CFUs of *Shigella* spp. and/or strains of *Salmonella* species on food, foodstuff, beverages and water, among others, become serious concerns to several food regulatory organizations worldwide (Kortei et al., 2020). Thus, food safety status of ready-to-eat meals is very vital in this case. Fortunately, in this study, comparison of the acceptable limits to numbers of CFUs of *Shigella* spp. proved that all the food samples from the commercially cooked meals within the capital city of Yilo Krobo Municipal appeared to be safe for consumption by livestock, pets and the public. However, the required acceptable limit for *Salmonella* species in food so that the commercial meals can be considered to be safe for consumption is zero (0) CFUs (25 g)<sup>-1</sup> (IMNRC, 2003; Feglo and Sakyi, 2012; WBG, 2017; WBG, 2017; FSAI, 2018; Kortei et al., 2020; NLM, 2024; UKHSA, 2024). Unfortunately, in this study, incidence of CFUs of *Salmonella* species per 5 g of the majority of the selected food samples appeared to support a high likelihood that the commercial meals may not be considered as totally safe for consumption. Moreover, our study did not include drinks, beverages, fruits and candies, among others, which are more a potential fomite to *Salmonella* spp. than the commercial meals used in



this study could be (Parker et al., 2021). Perhaps, regular studies to monitor cases of typhoid fever caused by *Salmonella* species in Yilo Krobo Municipal, similar to the report of Fusheini and Gyawu (2020) will be significant towards implementation of eradication strategies by stakeholders.

### 4.3 Perspectives for improvement and sustainability in the future

Permissible (or acceptable) limits for unwholesome chemical, physical and biological pollutants in water for drinking, water for irrigation, agricultural soils and ready-to-eat-meals largely vary from country to country across the world (WBG, 2017; Lente et al. 2022; Osei-Owusu et al., 2023). These variations are primarily due to differences in statutory laws or policies of the various countries that mandate their individual regulatory authorities (Osei-Owusu et al., 2023). In terms of incidence of heavy metals with microbes in water for drinking, the majority of the values of acceptable limits set by Ghanaian water resource regulatory authority are lower, when compared to those set by USA, European Union, China, Australia, UN-WHO (United Nations-World Health Organization) and Canada, among others (Lente et al. 2022; Osei-Owusu et al., 2023). In this case, high potable water quality standard across Ghana will be expected to effectively protect the public against undesirable levels of toxic soluble heavy metals with other contaminants in water for drinking. Notwithstanding, in terms of some enteropathogenic bacterial contaminations of commercial meals, the public will be less protected by the Ghana Standard Board (GSB), when compared to the way humans are effectively protected by all the international food regulatory authorities used in this study. This is primarily because values of acceptable limits set by GSB for incidence of enteropathogenic strains of either *E. coli* or *Shigella* species alone in commercial meals are about 99,000 CFU/g higher than those set by all the international food regulatory authorities, considered in the current study.

Thus, although the current study is limited to Somanya township, consideration to review and reduce values of the current acceptable limits set by GSB for some enteropathogenic bacteria in ready-to-eat meals will effectively protect the public across Ghana better. According to FDA (2022), Ghana has a powerful national food safety policy which aims at achieving (i) food production and safety, (ii) food value chain, (iii) imports and exports of safe food, (iv) training people in food safety, (v) public and consumer education, (vi) laboratories and surveillance and (vii) biosecurity issues. The main objective of this policy is to protect the public against unsafe and unwholesome food products. Moreover, public education and awareness have been effective, in terms of outbreak of epidemic and pandemic infectious diseases across communities in Ghana (Tutu et al., 2019; Tasiame et al., 2020;

Engmann et al., 2021; SVI, 2021). Perhaps, similar initiatives, which have been embedded in the Food Safety Policy (FSP) of the Government of Ghana, may be useful if they are implemented at district, municipal and metropolitan assembly levels to sustain and improve any correct acceptable food hygiene and safety practices among food vendors. Additionally, promoting food safety through community and municipal engagement approaches may efficiently protect the public against infectious food-borne diseases (WHO, 2023b; Lee et al., 2024). Generally, enteropathogenic bacteria are commonly associated with accumulation of wastes, dirt and dusts in the environments (Parker et al., 2021). According to suggestions by Larbi et al. (2024), effective waste segregation practices for environmental sanitation-driven circular economy can be adopted in districts, municipalities and metropolitans to reduce spread of these dangerous bacteria among humans.

## 5. Conclusion

*Proteus* species, *E. coli*, *Shigella* species and *Salmonella* species were isolated from all the commercial food samples, except 'kenkey without kenkey stew'. However, numbers of CFUs of *Proteus* species were within the acceptable limits set by food regulatory institutions in Ghana, UK, China, USA, Ireland, Canada and Europe, suggesting that all the commercially cooked food might receive satisfactory food hygiene treatments. High counts of CFUs of *E. coli* on *waakye* were not within the acceptable limits set by all the international food regulatory authorities. Nonetheless, numbers of CFUs of *E. coli* on *waakye* stew, porridge, sugar added to porridge, *kenkey* and *kenkey* stew were within the acceptable limit set by all the international food regulatory authorities, thereby suggesting that these tested food samples had satisfactory food hygiene requirements, similar to those meals in UK, China, USA, Ireland, Canada and Europe. Using the guidelines of Ghana standard board, numbers of CFUs of *E. coli* on all the tested commercial meals were within the acceptable limit set for satisfactory food hygiene requirements in Ghana only. Moreover, numbers of CFUs of *Shigella* spp. on all the food samples were within the acceptable limits set by regulatory authorities in Ghana, UK, China, USA, Ireland, Canada and Europe, suggesting that the commercial meals appeared to be safe for consumption. Nonetheless, lack of incidence of *Salmonella* in food samples has always been the strictest irrefutable policy by all food regulatory agencies worldwide. Therefore, perspectives to achieve improvement and sustainability in the future have been discussed.

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## 8. Conflict of interest

The authors have no conflicting interest to declare.

## 9. Data availability statement

Information collected during this study is included in this article. However, data may be available upon request.

## 10. Ethical standard and consideration

Authors did not use human or animal bodies as objects for this study.

## 11. Authors' contributions

**WKH** supervised the collection of data, analysed the data using R software, wrote the manuscript and carefully edited it for publication; **GKD** and **EKO-D** collected data according to bacteriological standard procedures, under the auspices of **WKH**, and then revised the manuscript; **JO** and **NNDA** revised the manuscript and then contributed valuable information.

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