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Bacteriological and Physiochemical Evaluation of contamination with *E. coli* for food samples collected from some markets in Nyala town South Darfur -Sudan

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Abstract

E.col. has been an important issues in most countries overall the world since 1982. This study was conducted to examine the levels of foodborne microbial contamination in food samples displayed in Nyala markets –South Darfur State - Sudan . A total of 778 food samples were microbiologically examined for their contamination with *E. coli*, the microbiological examination was followed by a physical confirmatory test. The results showed that almost all the food samples collected from the study area are contaminated with several strains of bacteria, while 71(9.1%) of the food samples under study proved to contain *E. coli*. Thus unhygienic practice may considered the risk factor associated with food processing and contamination. Health professional in the area of the study are recommended to have strategic plans to combat and reduce the spread of the foodborne pathogens, with the aim of controlling their outbreak in the community.

Key words: **Bacteriological,Physio-chemical, *E coli*, contamination**, **Foodborne ,Nyala**

1- Introduction

Food is chemically complex matrix, and predicting whether, microorganisms will grow in any given food is difficult. Most foods contain sufficient nutrients to supports microbial growth. Several factors encourage present, or limit the growth of microorganisms in food

the most important are water availability PH ,and temperature (Saadia, 2010). *E. coli*, is a type of bacteria that normally lives in human intestine. It's also found in the gut of some animal. Most types of *E. coli* are harmless and even help to keep the digestive tract healthy, but some strains can cause diarrhea if you eat contaminated food or drink is consumed. The quality of food and treatment of food borne diseases are critical public health issue. Bacteria contaminant of different food sources is most common health risk (Aberaetal, 2011). The different strains of *E. coli* are an important foodborne pathogens which were first identified as human enteric pathogen in 1982 (You *etal*., 2006, Dontorouetal, 2003, Lee and Choi,2006). Continued to occur in large outbreaks and sporadic cases, although outbreaks were decreased after 1999 (Lim *etal*., 2010). It has been estimated that *E. coli* causes 73000 illnesses and 250 death annually in the United State (Mead *etal*.,1999). The natural reservoirs pathogens are many kind of animals especially cell, sheep, goats, and wild animals. Consumption of undercooks of *E. coli*. However, fecal contaminations of other infected animals have also been linked as routes of transmission of human illness (Mead *etal*., 1999, kiranmay *etal*., 2010). Contaminated raw or under cooked poultry and red meat are particularly important in transmitting these foodborne pathogens. Other sources of human infections include contaminated produce and contact with farm animals and pets. Person to person transmission has also been described. Dangerous microorganisms are found in soil, water, animals and people. These microorganisms are carried on hands, wiping cloths and utensils especially hooping boards. The slightest contact can transfer them to food and cause foodborne disease, examples of zoonotic pathogens that may be transmitted in this way include *Salmonella, Campylobacter*, *E. coli* and the eggs of the tap worn *Taenia solinm* (Uyttendatle, 1999).Traditional culture methods for detecting *E. coli* in food are based on the incorporation of food samples into medium in which the bacteria can multiply, this providing visual confirmation of their growth (Betts and Blackburn,2009).

In south Darfur over 70% of communicable diseases are due to poor environmental health condition arising from unsafe food (Madina, 2016).

The rotation of helicoidal flagella allows the motion of the *E. coli* at Reynolds number (Re) and its caused by the drag anisotropy, (Lauga and Powers,2008).

2- Material and methods

Collection of samples:

778 food samples were obtained from different markets in Nyala town. All the samples were randomly collected aseptically in sterile container and immediately transferred to the laboratory within 2hrs on the day of collection to isolate *E. coli* according to procedure proposed by food and Drag Administration (FAD,2001).

Primary test for *E .coli* isolation

The food samples were dissolved in sterile distilled water and cultured on blood agar and incubated aerobically at 37C°- 48C° and examined for bacterial growth (Luciano *et a.,l* 2003). Then all the bacterial cultures on blood agar were cultured on MacConkey agar medium and incubated at

37 C° for 24hrs (Nazik.2003).

Confirmatory test for isolation *E. coli*

The pink colonies were growth on macConkey agar medium sub cultured on EMB agar and incubated at 37C° for 24hrs , green metallic shiny colonies were test by 1MViC - test (Nada,2008).

Physical Analysis:

Physically, we consider that the accumulation of bacteria in a sample of food depends on the viscosity factor and the flow rate of the medium that contain the bacteria. If the flow velocity of the bacterial medium is very small, food contamination with bacteria becomes greater. Therefore, the contamination is inversely proportional to the velocity

$$C \propto \frac{1}{v} \text{----------1}$$

It can also be considered that the contamination of food with bacteria is linked to a factor of viscosity, or

$$C \propto \eta \text{ ----------2}$$

From Eq(1) and Eq(2), can obtain.

$$Co = \kappa \frac{\eta}{v} \text{ ---------- 3}$$

Where: Co is the contamination, k is a constant proportionality, η is the viscosity factor, v is the velocity.

To confirm the correlation between the number bacteria with viscosity factor, the Reynolds (Re) number can be used. Assuming that the movement of bacteria is very slow even if food is available, the amount of (Co) is equivalent one kilogram per square meter, and that due to

$$k = 1$$

therefore, Eq(3) becomes as follow

$$\eta = Co\ v \text{ ---------- } 4$$

Depending on this , and by using eq(4) and Reynolds formula which is governs cells bacteria motility, can obtain

$$Re = \frac{\rho d}{Co} \text{ ---------- } 5$$

Where ρ is a density , d is a distance

Thermal effect on the viscosity of the bacterial contaminated food

It is known that the viscosity coefficient in the case of liquidity is inversely proportional to the temperature, and therefore if the temperature decreases, the viscosity increases, when the material changes from the liquid state to the hardness state. This lead to

$$\eta \propto \frac{1}{T}$$

Or

$$\eta = \frac{\lambda}{T} \text{ ---------- } 6$$

Therefore, it can be considered there is a critical temperature that separating between flow medium and semi-flow medium . Thus the exponential formula can be used to express the relationship between viscosity coefficient (η) and temperature(T), as follow

$$\eta = \eta_o \exp(\frac{\lambda}{T}) \text{ ----------} 7$$

Where: η is coefficient of viscosity, which directly depends on the temperature, $\eta_o$is an initial viscosity , λ is constant.

By substituting Eq(4) in Eq(7), can obtain

$$C = C_o \exp(\frac{\lambda}{T}) \text{----------} 8$$

Where: C is contamination bacteria (kg per square meter), C_o is an initial contamination bacteria (Reynolds, 1883).

3- Results

The results showed that 579 out of 778 (74.4%) food samples contaminated with bacteria . Table(1), Fig.(1)

Table(1)Number of Bacterial samples isolated from different food growth on blood agar medium

		Frequency	Percent
Valid	Positive	579	74.4
	Negative	199	25.6
	Total	778	100.0

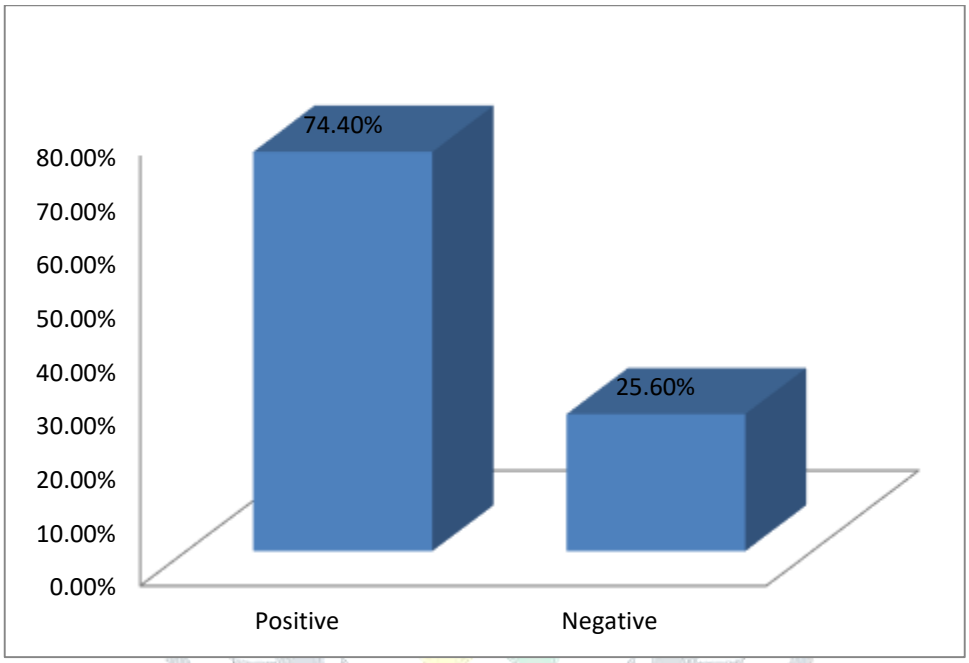

Fig.(1)The percentage of bacterial growth on blood agar mediun

323 out of 579 (56%) food samples contaminated with Entero bacteria.

Table(2)Fig.(2)

Table(2) Number of Enterobacteria growth on MacConkeyAgar medium isolated from different food.

		Frequency	Percent
Valid	Positive	323	56
	Negative	256	44
	Total	579	100.0

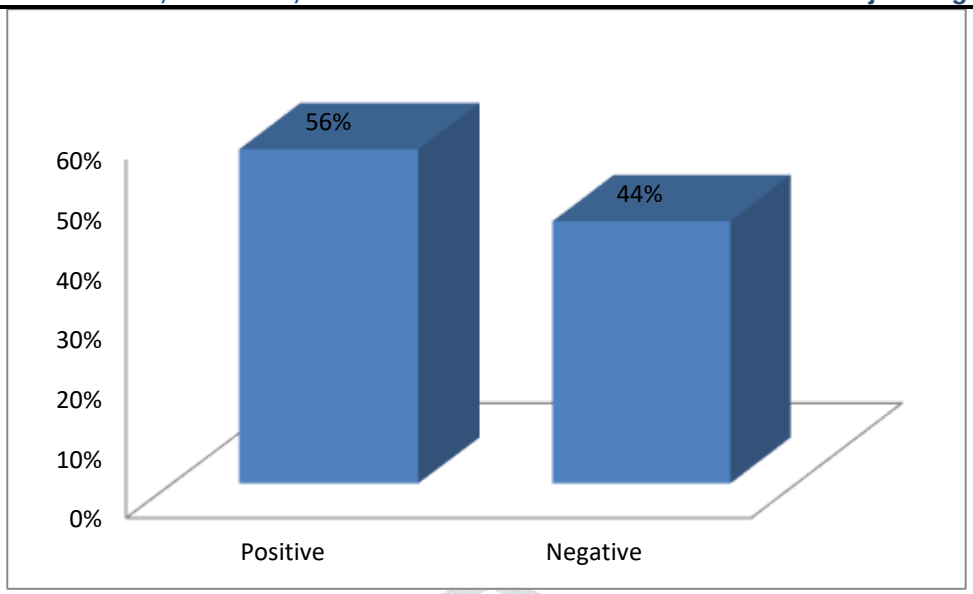

Fig.(2) The percentage of *Enterobacterial* growth on MacConkeyAgar medium

71 out of 323 (9.1%) samples were contaminated with *E. coli*. Table (3) Fig.(3)

Table (3)**Number of *E. coli* bacteria growth on EMB agar medium isolated from different food .**

		Frequency	Percent
Valid	Positive	71	9.1
	Negative	252	90.9
	Total	323	100.0

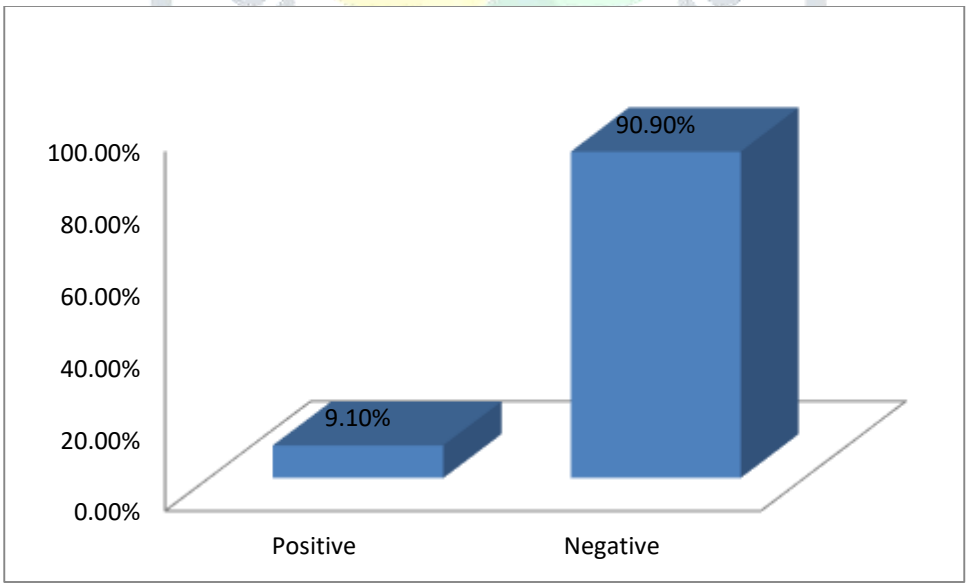

Fig.(3) **The percentage of *E. coli* bacteria growth on EMB agar medium isolated from different food**

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According to Table (4)& Fig.(4) The highest food contamination with bacteria was in summer (65%) followed by Winter(45,4%) and the lowest contamination was in Autum (8,64%)

Table (4) The number of contaminated food in different Seasons Isolated from Nyala markets

Seasons		Food samples	Percent
Valid	Summer	513	65.9
	Autumn	67	8.6
	Winter	198	25.4
	Total	778	100.0

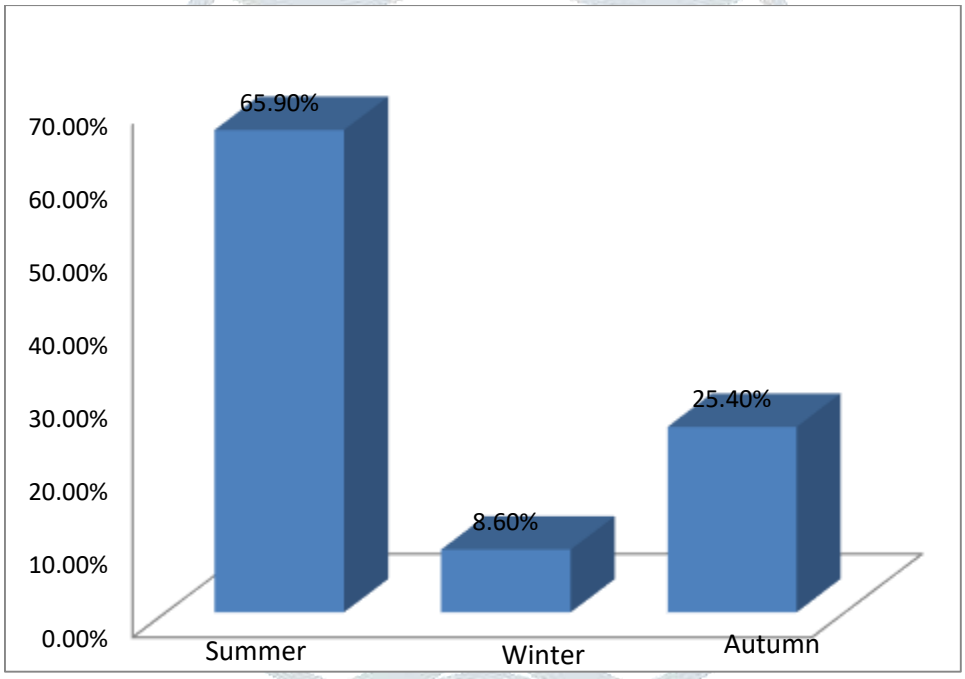


Fig.(4) The percentage of seasons showed high contamination o f food displayed in Nyala markets.

Table (5) The number of contaminated food with *E. coli* displayed in the study area

Samle Aeas (Nyala markets)	Samples of food contaminated with *E. coli*
Algabel market	5
Algenobi market	8
Algenena	17
Alshabi	14
The main market	12
Almalaja	15
Live stock market	0
Industrial area market	0
Total	71

Referred to Table & Fig (5) the highest contaminated food with *E. coli* was found Algenena market and the lowest food contamination was in Industrial area, Live stock markets respectivley

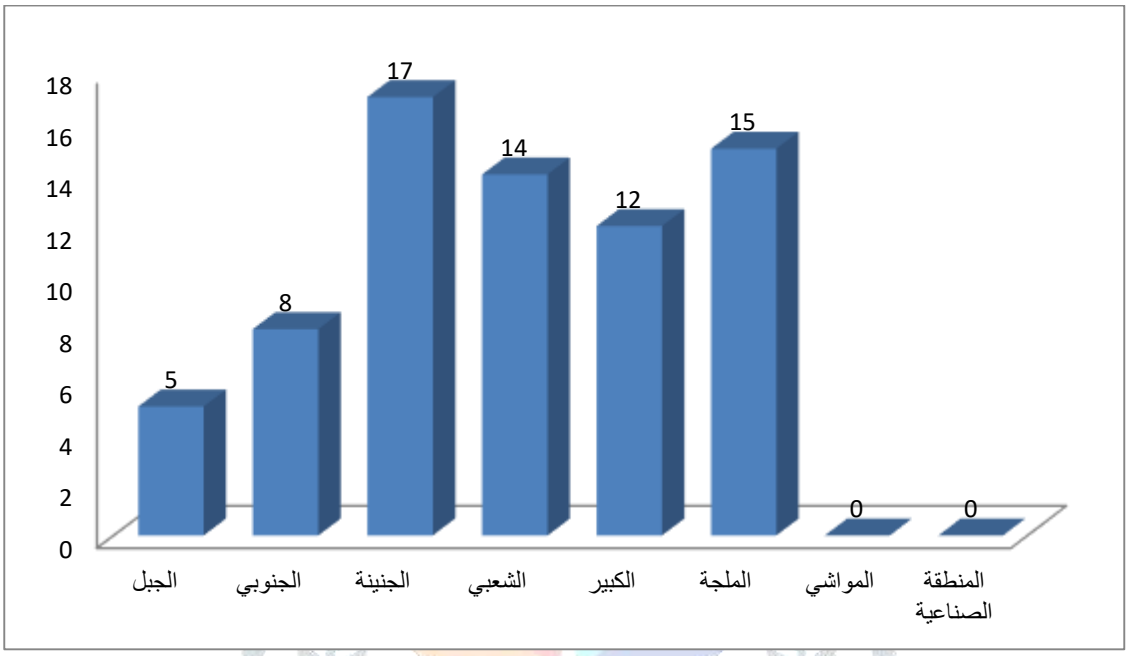
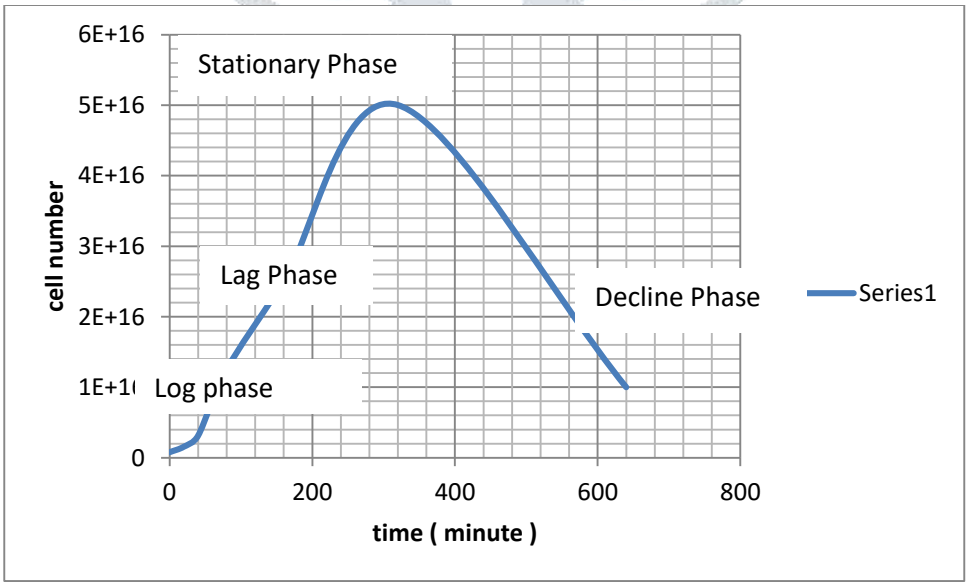


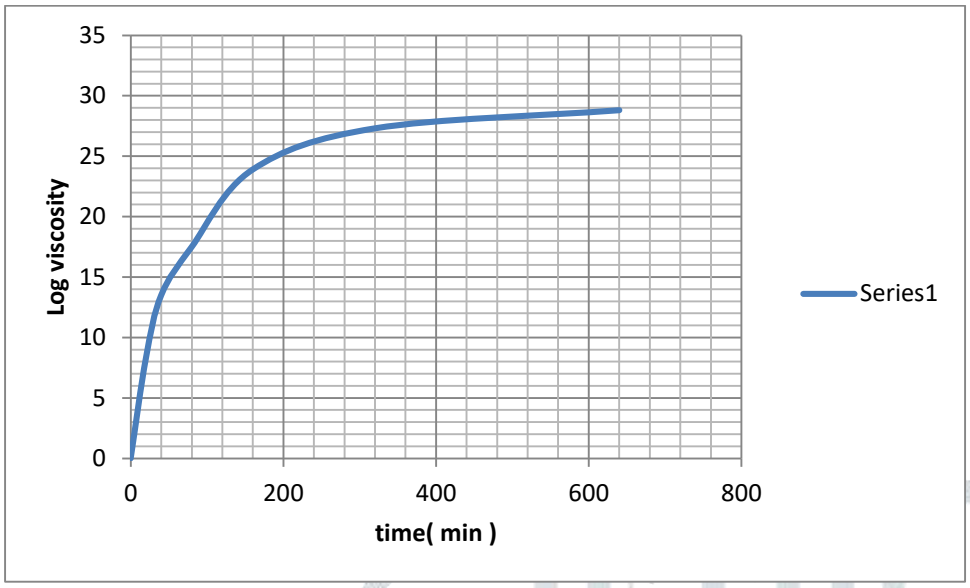
Fig.(5) The most highest of Nyala markets contain food contaminated with *E. coli*

The slow flow of bacteria in the food environment caused by increased viscosity, which leads to lower speed and to a lower value over time. We can deduce the food is highly contaminated ,Fig.(6).



Fig(6): illustrates the relationship between the number of bacterial cells and time .

In above figure , we note that within four hours , bacteria in food reach large number and then start to decline .



Fig(7) . show the relationship between *log* of the viscosity of contaminated food with time

Also, Fig(7) refers to bacterial overgrowth. Where we can see that slow movement of bacteria resulting by its high viscosity.

Discussion

As The effect of microorganisms on human health has been reported, the present study was performed to give information of the presence of pathogenic microorganisms in traditional foods , that important to human and to discuss their role in the food poising and also the causation of many human diseases. Studies on isolation of *E .coli* bacteria, from traditional foods in this investigation indicated that the food displayed in Nyala markets were extremely contaminated (Table1&Fig1).

The presence of *E. coli* in foods or drinks that inter the human body and cause symptoms such as diarrhea and various other gastrointestinal diseases (Kurniadi *et al.*, 2013).

The present study demonstrated that different traditional samples of food from markets in Nyala town were contaminated with *E. coli* spp. (9.1%) among 71 food samples Fig. (3). This very high level of contamination indicates to un favorable hygiene condition.

Our results are in agreement with several studies that reported by many researches, *(*Kay *et al*., 1994). Report that some pathogenic bacteria, fungi and yeasts were found in traditional fast foods.

Yunus *et al.*,2015 also mentioned that there was a significant relationship between personal hygiene and *E. coli* contamination on food with a P value of 0.002.

Tessi *et al.,* (2002). *E. coli* contamination (6.43 %) was observed a mong 101 cooked and prepared food in a university of center in Argentina. Bichai *et al.,* (2008) showed that, the presence of *E. coli* can be related to use of polluted irrigation waters during growth. Contamination through human handling, the use of contaminated containers, or washing after harvest with polluted water, could increase the incidence of enteric pathogens (Angelillo *et al.*, 2000*).*Nyala could be considered as an endemic zone for diarrhea, more than 5% of death of children below 5 years of ages is attributed to it.To confirm the contamination of food with bacteria we use in this study a physical parameter (Reynolds number equation) , the result revealed that there is correlation between bacterial number and time, it was found that the number of the bacterial cells increase till it reached the stationary phase in then decline due to diminishing of the nutrients from the food(Fig. 6). The physical parameter also showed the relationship between the viscosity of the food and motility of the bacteria, the more vicious the medium(food) the movement of the bacteria becomes slow. The slow flow of bacteria in the food environment caused by increased viscosity, which leads to lower speed and a higher value of contamination(Fig.7).

According to our knowledge it's the first time that " Reynolds's number " is used for checking food contamination .

Conclusion

Food borne diseases is an urgent public health problem and requires rapid intervention. This study was conducted to examine the levels of foodborne microbial contamination in food samples displayed in Nyala markets. The findings from this study suggested that most foods displayed in Nyala markets had a satisfactory level of contamination with *E. coli*. Unhygienic practice may considered the risk factor associated with processing and contamination of food. According to our knowledge its the first time that " Reynolds's number " is used for determination of the relationship between bacterial motility and number of bacterial cell with time . However further molecular study is necessary to detect the strains of *E. coli* in food.

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