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Prevalence and Detection of Beta Lactams and Tetracyclines in Raw Milk

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Abstract: Fifty-five raw cow's milk samples were collected from different places (35 samples from cooled tanks in supermarkets, 10 samples from sick individual animals and 10 samples from bulk farm milk). The samples were screened for Beta Lactams and tetracyclines by using Delvotest® SP NT and Betastar® combo HS kits. Comparison between both kits was done. Positive screened milk samples were analyzed by using High Performance Liquid Chromatography (HPLC) for detection of Ampicillin, Penicillin G, Amoxycillin, Tetracycline and oxytetracycline. Incidence of antibiotic residues in raw milk samples was 23.6% and 20% by using Delvotest® SP NT and Betastar® combo HS kits, respectively. The incidence of Beta Lactams and tetracyclines in raw milk samples was 12.7 and 10.1%, respectively by using both Betastar® combo HS kits and HPLC. Analysis by HPLC revealed that 53.8, 15.38, 46.1 and 15.38% from analyzed positive milk samples were contaminated with Ampicillin, Penicillin G, Amoxycillin, Tetracycline and Oxytetracycline, respectively. The mean value of Ampicillin, Penicillin G, Tetracycline and oxytetracycline residues were 902.72± 193.969 µg/L, 9.54± 0.005 µg/L, 1659.3± 685.81 µg/L and 2521.6± 1076.18 µg/L, respectively. Amoxycillin could not be detected in raw milk samples. All positive analyzed samples exceeded maximum residue limit established either by European Union (EU) or Food and Drug Administration (FDA) regulations.

Key words: Beta Lactams · Tetracyclines · Raw Milk

INTRODUCTION

From more than five decades, antimicrobial agents are used in veterinary practice [1]. They are used as a treatment for animals or as dietary supplements for disease prevention and growth promotion. Their improper use without veterinary control lead to presence of antibiotic residues in milk which give negative effects during the dairy productive sequences [2]. Antibiotic residues in milk can produce a great loss in fermented dairy products industry by inhibition of starter fermentation during cheese and yoghurt production [3]. Also, affect flavor and acid production associated with manufacture of butter [4]. In addition, the harmful effects of antibiotic residues on consumers, especially, the development of allergic problems, interference of intestinal microflora [5] and bacterial resistance which hinder antibiotic treatment [6].

Beta Lactams are the oldest family of antibiotics but, still one of the most important groups of antibiotics used in animal husbandry and it is responsible for 95% of milk contamination [7]. Tetracyclines are broad spectrum antibiotics widely used in food production animals [8]. In Africa, Tetracyclines are the most predominant antibiotics and represent 41.17% of antibiotic residues in animal derived foods, followed by Beta Lactams residues which represent 18% [9].

The aim of this study was to determine the prevalence of both Beta Lactams and tetracyclines in raw cow's milk. The residues were examined in milk samples which had been collected from different sources (cooled tanks in supermarkets, farm tanks and sick individual animals). Qualitative detection of both antibiotic groups in milk samples via two methods (Delvotest® SP NT and Betastar® combo HS) was carried out. Comparison between both kits was done. Beside that quantitative detection of residues to positive milk samples by using High Performance Liquid Chromatography (HPLC).

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MATERIALS AND METHODS

Qualitative Detection of Beta Lactams and Tetracyclines Antibiotic Residues in Raw Milk Samples: A total of 55 raw cow's milk samples including thirty five milk samples collected from supermarkets, ten milk samples from sick individual animals treated with beta lactams and tetracyclines and ten milk samples collected from farm tanks. The samples were screened for the presence of Beta Lactam and Tetracyclines antimicrobial residues using Delvotest® SP NT kits and Betastar® Combo HS kits.

Delvotest® SP NT kits (DSM, Netherlands): Delvotest is a broad spectrum screening test for the detection of different antibiotic residues in milk. The test is made of an agar gel containing a standard number of bacterial spores (Bacillus stearothermophilus) and a pH indicator bromocresol purple. The test is based on the diffusion of antimicrobial residues that may be present in milk in to agar. The residues decrease or prevent the growth of bacteria. So, delay or prevent colour changing from purple to yellow.

The test procedures were applied as test instructions. 0.1 ml of milk sample was injected in to the test ampoule and incubated for 3 hours at (64°C±2°C). The results were read visually, yellow color was interpreted as negative result and purple color was positive result. But, if the upper third of the agar gel was purple and the lower two third of the agar gel was yellow the test result was recorded as negative result.

Betastar® Combo HS Rapid Test (Chr. Hansen, Denmark): Betastar® combo HS is a high sensitive and rapid detection test for beta-lactam / cephalosporine antibiotics (e.g. penicillin, ampicillin, cephalonium etc.) and tetracycline antibiotics in cow, goat and sheep milk. The test performed as test instruction. 0.2ml of milk sample was added to a vial with a specific amount of binding reagents and incubated at 37.5 ± 1°C for 5 minutes, where binding reagents will react with any beta-lactams and/or tetracyclines present in milk sample. Immunochromatographic medium (Betastar® combo HS stick) is added and incubation is continued (3 minutes at 37.5 ± 1°C) where detection takes place on three detection bands on the stick. The lower line on this stick binds all the tetracycline binding reagents that have not interacted with tetracycline antibiotic during the first incubation. The middle line acts as a control line to ensure efficiency of the test itself. The upper line captures all the beta-lactam binding reagents that have not reacted to any beta-lactam antibiotic during the first incubation. The results were read visually. If the upper and the lower line showed red bands with higher color intensity than the control line, the test would be negative and the milk is free from beta-lactam or tetracycline antibiotics if no test band or a weaker red band was formed for each of the two antibiotics, the test would be positive. But, if the control line band is absent, the test is invalid.

Quantitative Detection of Beta-Lactams and Tetracyclines in Raw Milk Samples by High Performance Liquid Chromatography

Chemicals and Reagents: Amoxicillin, Penicillin, Ampicillin, Tetracycline HCl T3383 and Oxytetracycline HCl O5875 standards (Sigma, Aldrich), HPLC grade acetonitrile and methanol (Sigma), EDTA (Ethylene Diamine tetra acetic acid disodium salt), Citric acid monohydrate, Sodium hydrogen phosphate, Acetic acid (El-Nasr Pharmaceutical chemicals Co.,).

Preparation of Milk Samples:

Beta lactams detection according to Khaskheli *et al*. [10].

Five ml of milk sample was mixed with 400µl of 10% aqueous solution of acetic acid in sterilized plastic centrifuge tube. The solution was agitated for one minute using vortex. Then it was centrifuged at 3500 rpm/10 minutes in a cooling centrifuge at 4°C. The clear supernatant fluid was carefully separated and filtered by using 13mm diameter 0.45µm nylon filter. The filtrate was taken in to 2 ml plastic disposable vials and 5µl of filtrate was injected in to HPLC.

Tetracyclines detection according to Abbasi *et al*. [11].

Fifteen ml of milk sample was mixed in a 50 ml plastic centrifuge tube with 25ml Mcllvaine buffer solution (mixed citrate/phosphate with EDTA). The solution was agitated by using vortex for one minute then centrifuged at 10, 000 rpm/12min in a cooling centrifuge at 4°C. Avoid lipid layer and the disposed precipitates, the supernatant was used in Solid Phase Extraction (SPE). SPE cartridges were prepared by treating them with 3ml methanol at a flow rate no faster than 3ml/min, then rinsed by deionized water (2ml). All samples were extracted by loading 25ml of

supernatant fluid in SPE at a flow rate 5ml/min then the cartridge was treated with methanol solution 5%. Elution took place with 2ml of methanol HPLC grade at rate 4ml/min. The samples were dried on the rotary vacuum evaporator. The dried residues were diluted in to1ml of the mobile phase then filterated by using 0.2µm nylon filter then filtrate was injected in to HPLC system.

Preparation of Antibiotics Standard Solutions: Amoxicillin, Ampicillin and penicillin G stock standard solutions were prepared individually in deionized water at a concentration of 100µg/ml. Mixed standard working solutions were equipped from stock solutions at concentration from 500µg/ml to 5000µg/ml. Tetracycline HCl T3383 and oxytetracycline HCl O5875 standard were prepared with deionized water in concentration ranged from 500 to 5000ug/L. The working standard solutions were prepared from the stock solutions.

Quantification: The quantification was performed by injecting standard solutions, blank and spiked samples.

Condition of HPLC: The HPLC system was a constant liquid chromatography pump, Agilent 1200 series co., Germany, with variable wave length detector co., Germany in addition to a software chemistation, Germany. The mobile phase in Beta lactams quantitation was 0.1% trifluroacetic acid and acetonitrile (50:50). It was pumped at a flow rate of 1ml/ min. the Beta lactam was detected at wave length 254 nm. The injected volume was 50µl. HPLC column was C18 (250×5mm) Hypersil Eclipse XDP. But, the mobile phase in Tetracyclines quantitation was consisting of distilled water (H_2O 2.1 with H_2SO_4) acetonitrile 85:15 (v/v) was pumped at flow rate of 1.5ml /min. The analyte was detected at 360nm. The injected volume was 20ul and chromatography was performed at 24°C. HPLC column was Hypersil Eclipse XDP C18 (5µm, 250 × 4.6mm).

Statistical Analysis: Data were analyzed statistically by the software (SPSS) version 16. Chi-Square analysis at $P< 0.05$ was conducted to compare the incidence of antibiotic residues in raw milk by using Delvotest® SP NT and Betastar® combo HS kits, Kappa statistic was used to determine the agreement between the two kits and finally *t*-test was performed to compare the ampicillin, tetracycline and oxytetracycline concentrations in raw milk from cooled tanks of supermarkets and raw milk from sick individual animals.

RESULTS AND DISCUSSION

Incidence of Antibiotic Residues in Raw Cow's Milk Samples by Using Delvotest® SP NT kits, Betastar® Combo HS Kits and HPLC: The extensive uses of antibiotics in animal husbandry as well as the insufficient withdrawal time are the most important causes of antibiotic residues in milk. The incidence of antibiotic residues in raw milk by using Delvotest® SP NT kits in this study was 23.6% distributed to 5.7% of market raw milk, 100% of raw milk from sick individual animals and 10% of bulk tank milk. While by using Betastar® Combo HS kits and High Performance Liquid Chromatography (HPLC), the incidence of antibiotic residues in raw milk was 20% divided in to 5.7% of market raw milk, 90% of raw milk from sick animals. Antibiotic residues could not be detected in bulk tank milk as shown in (Table 1). There was a high significant difference (P value = 0.000) between positive raw milk samples of detectable antibiotic residues by using Delvotest® SP.NT kit and Betastar® Combo HS kit. When using the kappa statistics, the degree of agreement between the two kits was 89%.

In this study, the incidence of antibiotic residues in raw milk was nearly similar to that reported by Movassagh [12] who studied the prevalence of antibiotic residues in raw cow's milk and showed that 24% of all samples were positive for antibiotic residues. Several studies performed by Ceyhan and Bozkurt [13], Adesiyun and Webb [14], Movassagh and Karami [15] and Movassagh [16] showed lower incidence of antibiotic residues in raw milk than our study which were 5.5, 10.8, 5 and 14%, respectively. On the other hand, Salman *et al*. [17] recorded a higher incidence of antibiotic residues (33.1%).

There was a considerable difference in positive results. Delvotest SP NT kit gave a positive result while Betastar® Combo HS kits showed a negative one for the same sample. This difference is attributed to that Delvotest SP NT kit is a highly sensitive to wide range of antimicrobials used in the dairy practice but not specific to Beta Lactams and Tetracyclines antibiotic as Betastar® Combo HS kits. Ghidini *et al*. [7] reported that Delvotest SP® is very sensitive, but not very selective. Although, the most important advantage of qualitative screening tests is that they don't give false negative result but can give false positive responses, particularly in case of high somatic cell count [18] or after addition of some feed supplementation [19].

Table 1: Incidence of antibiotic residues in raw cow's milk samples by using Delvotest SP NT kits, Betastar Combo HS Kits and High Performance Liquid Chromatography (HPLC)

		Positive samples in Delvotest® SP.NT kits		Positive samples in Betastar® Combo HS Kits*		Positive samples in High Performance Liquid Chromatography (HPLC)*	
Examined raw milk samples	Number of samples	No.	%	No.	%	No.	%
Market raw milk	35	2	5.7	2	5.7	2	5.7
Raw milk from sick individual animals	10	10	100	9	90	9	90
Bulk farm milk	10	1	10	0	0	0	0
Total	55	13	23.6	11	20	11	20

Refer to detection of Beta lactams and Tetracyclines only.

A high significant difference was detected between positive raw milk samples of detectable antibiotic residues by using Delvotest® SP.NT kits and Betastar® Combo HS Kits. (Chi- Square value = 37.58, degree of freedom = 2, P value = 0.000)

Table 2: Incidence of Beta Lactams and Tetracycline residues in raw cow's milk samples by using Betastar® Combo HS Kits and High Performance Liquid Chromatography (HPLC)

		Positive samples in both Betastar® Combo HS Kits and High Performance Liquid Chromatography (HPLC).			
		Beta Lactam		Tetracyclines	
Examined samples	Number of samples	No.	%	No.	%
Market raw milk	35	2	5.7	2	5.7
Raw milk from sick individual animals	10	5	50	4	40
Bulk farm milk	10	0	0	0	0
Total	55	7	12.7	6	10.1

Betastar® Combo HS kit is a rapid highly sensitive assay for detection of both Beta Lactam and Tetracyclines in milk. This test is very easy to perform, incubation period is very short (3 minutes) and the results can be read visually. While, by using Delvotest SP NT the incubation period is longer (3 hours).

The most important feature of Betastar® Combo HS kits is having the ability to distinguish between Beta lactam and Tetracyclines antimicrobial residues. On the contrary, Delvotest SP NT indicates only presence or absence of antibiotic residues in milk samples but does not determine the type of antibiotic present.

Incidence of Beta Lactams and Tetracyclines Residues in Raw Cow's Milk Samples by Using Beta Star Combo HS Kits and HPLC: Beta Lactams and Tetracyclines are the most common used antibiotics for the treatment of mastitis in animals [20]. The incidence of Beta Lactam and Tetracyclines residues in raw milk samples by using Betastar® Combo HS kits was in accordance with their incidence by using High Performance Liquid Chromatography (HPLC).

Beta Lactam residues were found in 7(12.7%) of raw milk samples, distributed as 2(5.7%) and 5 (50%) in market raw milk and raw milk from sick individual animals, respectively. Tetracyclines residues were detected in 6 (10.1%) of raw milk samples, distributed as 2 (5.7%) and 4 (40%) in market raw milk and raw milk from sick individual animals, successively, however Beta lactam and Tetracyclines residues could not detected in raw farm milk samples as described in (Table 2).

In this study, the incidence of Beta Lactam in raw milk was 12.7% which show lower incidence than those given by Khaskheli *et al.* [10], Al Zuheir *et al.* [21] and Sulejmani *et al.* [22]. Movassagh and Karami [23] and Zheng *et al.* [24] reported a lower incidence of Beta Lactam than that given in this study.

Many research works applied for detection of tetracyclines in raw milk samples such as Abbasi *et al.* [11], Al Zuheir [21], Elizabeta *et al.* [25] and Chowdhury *et al*. [26]. The incidence of tetracyclines in raw milk samples was 48.9, 28.6, 22.2 and 23%, respectively which show higher prevalence than this study.

By using HPLC, only 7 samples were contaminated with Beta Lactam antibiotics which represented 53.8% from positive samples while, only 6 samples (46.1%) contained total tetracycline antibiotic residues from positive samples. These results were in the same line of screening Betastar® Combo HS kits.

Table 3: Ampicillin and Penicillin G concentrations (µg/L) in raw cow's milk samples determined by HPLC analysis

	Beta Lactam analyte											
	Ampicillin µg/L						Penicillin G µg/L					
					Positive samples exceed MRL						Positive samples exceed MRL	
					EU	FDA					EU	FDA
Milk sample	Positive samples (No.)	Min	Max	Mean±SE	No. (%).	No. (%)	Positive samples (No.)	Min	Max	Mean±SE	No. (%).	No. (%)
Market raw milk	2	692	844.2	768.1± 76.1[a]	2(100)	2(100)	0	---	---	---	---	---
Raw milk from sick individual animals	5	408.5	1986.2	956.6±275.5[a]	5(100)	5(100)	2	9.54	9.55	9.54± 0.005	2(100)	2(100)
Total	7	408.5	1986.2	902.7±193.9	7(100)	7(100)	2	9.54	9.55	9.54± 0.005	2(100)	2(100)

Maximum Residue Limit (MRL) recommended by European Union (EU) legislation for Ampicillin and Penicillin G residues in milk is set to 4 µg/L but, that regulated by Food and Drug Administration (FDA) regulation is 10, 5 µg/L, respectively.

[a] refer to no significant difference was detected between concentration of ampicillin residues in market raw milk and raw milk from sick individual animals.

It is clear from results obtained in this study that analysis of antibiotic residues by using High Performance Liquid Chromatography (HPLC) confirmed the results of screening Betastar® Combo HS kits. So, Betastar® Combo HS kit is more reliable than Delvotest® SP NT kit for detection of Beta lactam and Tetracyclines in milk samples. Despite this fact Betastar® Combo HS kit is still a qualitative screening method for detection only Beta lactam and Tetracyclines in milk. The test cannot distinguish between members of antibiotics in both families.

Incidence of Ampicillin, Penicillin G and Amoxycillin Residues in Raw Cow's Milk Samples Determined by HPLC Analysis: Thirteen positive samples in screening tests either by Delvotest® SP NT or Betastar® Combo HS kit were confirmed for presence of Beta lactams and Tetracyclines antimicrobial residues through High Performance Liquid Chromatography (HPLC). The quantitative analysis was restricted to Ampicillin, Penicillin G, Amoxicillin, Tetracycline and oxytetracycline. The results revealed that over the 13 samples analyzed, 7 (53.8%) and 2 (15.38%) were contaminated with ampicillin and Penicillin G residues, respectively as described in (Table 3).

Penicillin G residues were found in only 2 samples (20%) of raw milk from sick individual animals. While, it could not be detected in market raw milk and bulk tank milk. Ampicillin residues were determined in 2 samples (100%) and 5 samples (50%) from market raw milk and raw milk collected from sick individual animals, respectively. In the other hand, all milk samples were free from amoxicillin residues. In some cases, more than one molecule of antibiotic was determined in one sample. All bulk tank milk samples were free from penicillin, ampicillin and amoxicillin residues as described in (Fig. 1).

The incidence of Penicillin G in this study was 15.38% which is remarkably lower than residues level of Penicillin G reported in several studies by Ghidini *et al*. [7], Khaskheli *et al*. [10] and Riediker *et al*. [27] where the incidence of Penicillin G residues was 44.44, 64 and 49.1%, respectively. On the other hand, the incidence of Penicillin G residues in this study was higher than that recorded by Ghanavi *et al*. [28] which was 11% of raw milk samples.

Ampicillin was the most detected Beta Lactam antibiotic residues in this study. The incidence of ampicillin residues in raw milk samples was 53.8% which is higher than that recorded by Khaskheli *et al*. [10] and Riediker *et al*. [27]. Amoxicillin residues could not be detected in any of tested samples. On contrary of other studies performed by Ghidini *et al*. [7], Khaskheli *et al*. [10] and Chowdhury *et al*. [26] amoxicillin residues could be detected in different percentage in raw milk samples.

Incidence of Tetracycline and Oxytetracycline residues in raw milk samples determined by HPLC analysis HPLC analysis revealed that over the 13 samples analyzed, 6 (46.1%) and 2 (15.38%) were contaminated with Tetracycline and oxytetracycline residues, respectively. Tetracycline residues were found in 2 samples (100%) and 4 samples (40%) of raw market milk and raw milk collected from sick individual animals, successively. On the other hand, oxytetracycline residues were determined in one sample (50%) and other sample (10%) of raw market milk and raw milk collected from sick individual animals, respectively. All raw farm milk samples were free from any tetracycline and oxytetracycline residues. As shown in (Fig. 1).

The incidence of tetracycline residues in milk was higher than that determined by Abbasi *et al*. [11] and Elizabeta *et al*. [25] which was 7.1% and 13.1%, respectively. Tetracycline residues could be detected by Navratilova *et al.* [29] and Tona and Olusola [30] in higher incidence (100%).

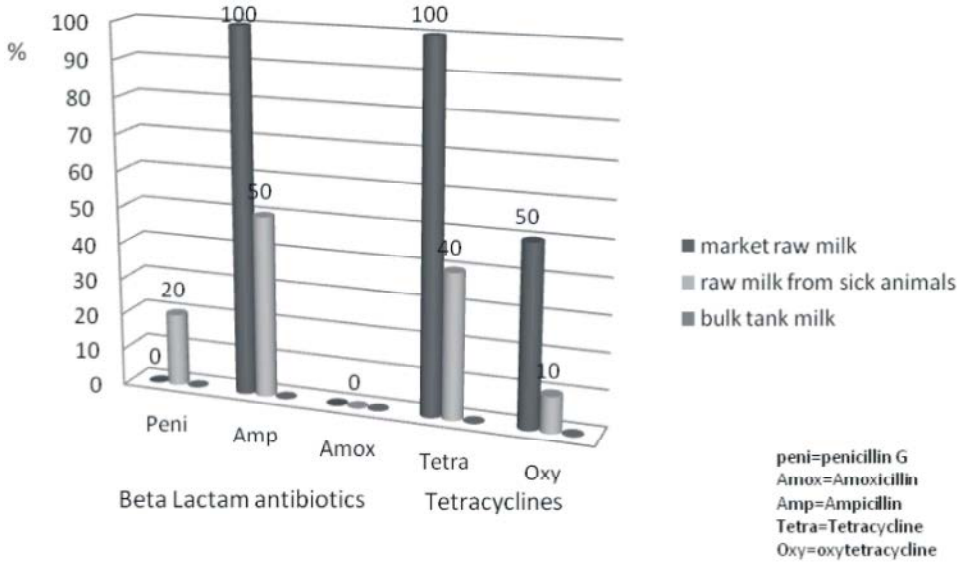

Fig. 1: Incidence of Ampicillin, Penicillin, Amoxycillin, Tetracycline and oxytetracycline in positive raw cow's milk samples determined by HPLC analysis.

Table 4: Tetracyclines and oxytetracycline concentrations in raw cow's milk samples determined by HPLC analysis

	Tetracycline analyte											
	Tetracycline µg/L						Oxytetracycline µg/L					
					Positive samples exceed MRL						Positive samples exceed MRL	
					EU	FDA					EU	FDA
Milk sample	Positive samples (No.)	Min	Max	Mean±SE	No. (%).	No. (%)	Positive samples (No.)	Min	Max	Mean±SE	No. (%).	No. (%)
Market raw milk	2	637.5	690.04	663.7±26.27[a]	2(100)	2(100)	1	1445.4	1445.4	---	1(100)	1(100)
Raw milk from sick individual animals	4	357.4	3809.8	1924.8±821.7[a]	4(100)	4(100)	1	3597.8	3597.8	---	1(100)	1(100)
Total	6	357.4	3809.8	1659.3±685.8	6(100)	6(100)	2	1445.4	3597.8	2521.6±1076.1	2(100)	2(100)

Maximum Residue Limit (MRL) recommended by European Union (EU) legislation for Tetracycline and oxytetracycline residues in milk is set to100 µg/L but, that regulated by Food and Drug Administration (FDA) regulation is 300 µg/L.

[a] refer to no significant difference was detected between concentration of tetracycline residues in market raw milk and raw milk from sick individual animals

In some reports by Abbasi *et al.* [11], Elizabeta *et al*. [25] and Navratilova *et al*. [29] detected oxytetracycline residues in a higher percentage than this study (50.6, 57.15 and 32.7%) from examined samples, respectively. While, Kamberi and Sulaj [31] recorded a lower incidence (4.4%) than this present study.

Mean Concentrations of Ampicillin and Penicillin G Residues in Raw Milk Samples: Ampicillin and penicillin G were more prominent members of Beta Lactam antibiotic group in this study. Ampicillin residues level in raw milk samples ranged between 408.5 to 1986.23µg/L and averaged 902.72± 193.969. The mean value was much higher than maximum residue limit established either by European Union (EU) and Food and Drug Administration (FDA) regulations which are 4, 10 µg/L, respectively. While, Penicillin G residues in raw milk samples ranged between 9.54 and 9.55 µg/L with average 9.54± 0.005 µg/L. The mean value was 2.38 fold higher than MRL of EU regulation (4 µg/L) and 1.9 fold higher than MRL of FDA regulations (5µg/L). There was no significant difference detected between concentration of ampicillin residues in market raw milk and raw milk from sick individual animals as described in (Table 4).

The mean value of ampicillin residues in milk samples in our study was higher than that detected by Khaskheli *et al*. [10]. While, it was lower than that recorded by Ramamohana Rao *et al.* [32]. From the presented data, the mean value of penicillin G residues was 6.24 fold lower compared to value recorded by Khaskheli *et al.* [10] and much lower than that reported by Ghidini *et al*. [7]. On the other hand, it was higher than that documented by Ghanavi *et al*. [28].

Mean Concentrations of Tetracycline and Oxytetracycline Residues in Raw Milk Samples: Tetracycline residues level in raw milk samples ranged between 357.41 µg/L to 3809.8 µg/L with average 1659.3± 685.81 µg/L. The mean value was 16.59 fold higher than MRL of EU regulation (100 µg/L) and 5.53 fold higher than MRL of FDA regulation (300 µg/L). Oxytetracycline residues were found in only 2 milk samples from all positive examined samples. its amount were 1445.396 and 3597.76 µg/L with average 2521.6± 1076.18 µg/L which was much higher than EU and FDA maximum residue limit. There was no significant difference detected between concentration of tetracycline residues in market raw milk and raw milk from sick individual animals as described in (Table 4).

In the present study, Tetracycline residue levels exceeded the MRLs and higher than that measured by Abbasi *et al.* [11], Elizabeta *et al*. [25], Navratilova *et al*. [29], Tona and Olusola [30] and Bilandzic *et al.* [33].

The mean value of oxytetracycline residues in milk was 2521.6± 1076.18 µg/L which was much higher than that recorded by Abbasi *et al.* [11] and Elizabeta *et al*. [25]. Also, it exceeded EU and FDA recommended levels 100µg/L, 300µg/L, successively.

CONCLUSIONS

The antibiotic contamination of food is one of the greatest obstacles to public health and economic concern that is faced not only Egyptian people, but also worldwide people. Beta lactams and tetracyclines are the highly predominant antibiotics used in animal husbandry. For the first time, Betastar® combo HS kits used for detection of beta-lactams and tetracyclines antibiotics in raw milk. In comparsion to Delvotest® SP NT kits, Betastar® combo HS kits is very easy, more specific for beta-lactams and tetracyclines antibiotics only, incubation period is very short (3 minutes) and the results is read easily. The most important feature of Betastar® Combo HS kits is having the ability to distinguish between Beta lactam and Tetracyclines antimicrobial residues in milk.

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