

## Determination of Beta Lactam Antibiotics Residues in Cow Raw Milk by Beta Star Test

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**Abstract:** Drug residues in milk have a potential hazard for the consumer. One hundred fifty cow raw milk samples were collected from Tabriz suburb milk collection centers from April 2010 to September 2010 by systematic random sampling methods. All samples were examined by Beta star screening kit (Neogen, USA). Of all samples, 8(5.33%) were positive for beta lactam antibiotic residues in cow raw milk in the northwest region of Iran. This study showed that the effective monitoring program must be run in the northwest region of Iran for control of beta lactam antibiotic residues presence in milk.

**Key words:** Beta-lactam · Residues · Cow Raw Milk · Tabriz

### INTRODUCTION

A number of antibiotics are used in livestock health care in Iran. Several hundred antibiotics have been isolated from various sources. Antimicrobial agents are used in treatment of cattle and cause the presence of drug residues in milk. Mastitis is the most prevalent disease in cattle which requires antimicrobial treatment [1, 2]. Drug residues in milk have a potential hazard for the consumer and may cause allergic reactions, interference in the intestinal flora and resistant populations of bacteria in the general populations, thereby rendering antibiotic treatment ineffective [3]. Consumers want to be confident that their food supply is free of contamination by herbicides, pesticides, drugs or antibiotics.

Approximately 5-10 percent of the populations is hypersensitive to Penicillin at a concentration as low as 1 ppb or other antibiotics and suffers allergic reactions (skin rushes, hives, asthma, anaphylactic shock). Concentration of 1 ppb delay starter activity during butter and yoghurt making. Antibiotics also decrease the acid and flavor production associated with butter manufacture and they reduce the curdling of milk and cause improper ripening of cheese [4].

Antibiotics have been used in cows for many years to treat such common infections as mastitis. The past several years has seen increased pressure on milk producers to increase milk production from each and every cow. This pressure often results in more infections

and the increased use of antibiotics to counter these infections. The beta lactam group of antibiotics is responsible for approximately 95% of all milk antibiotic contamination. Beta-lactam is a group of antibiotics which are frequently used for the treatment of animals in Iran. Beta-lactam antibiotic residues in milk cause problems in dairy industries and in human health. The maximum residues limit (MRL) for some beta-lactam antibiotics in European Union, for example Penicillin G 4 µg/L, Ampicillin 4 µg/L, Dicloxacillin 30 µg/L, Cephalexin 100 µg/L and Cepharin 60 µg/L [5].

There is no national antibiotic residue program in Iran. This is the first report, as far as we are aware, of beta-lactam antibiotic residues in cow raw milk in Iran. We were unable to find reports in our search of the literature.

### MATERIALS AND METHODS

One hundred fifty cow raw milk samples were collected from Tabriz suburb milk collection centers from April 2010 to September 2010 by systematic random sampling methods. All samples were examined by Beta star screening kit (Neogen, USA). BetaStar US is a receptor assay for rapid detection of the beta-lactam antibiotics penicillin, ampicillin, amoxicillin, cloxacillin and cephalosporin. Extensively used in the prevention and treatment of dairy cattle disease, particularly mastitis. This test is validated for use with raw, commingled cow's milk.

Table 1: MRL examples for milk in Europe, in the USA and for the Codex (ppb) [8]

Family	Molecule	Milk			
		MRL EU	MRL Codex	MRL USA	Beta star kit Tolerance/Safe limit
Beta lactam antibiotics	Penicillin G (Penethamate)	4	4	5	5
	Ampicillin	4	-	10	10
	Amoxicillin	4	-	50	10
	Cloxacillin	30	-	10	10
	Cephapirin	60	-	-	10

The test involves a specific beta lactam receptor linked to gold particles. It is a dipstick test that detects penicillins and cephalosporins. The milk sample (0.2 ml) is added to a vial containing the test reagents (receptor protein linked to gold particles), mixed and incubated at 47.5°C in the incubator for 3 minutes. During incubation, the receptor will react with the free beta lactams contained in the sample. After 3 min of incubation, the dipstick is added and incubation is continued (2 min at 47.5°C). The mixture is transferred to a strip of immunochromatography paper where it migrates towards the test field. With milk samples free of beta lactam residues, the receptor protein will be captured by a biomolecule immobilized at the test field of the chromatography paper. Since the receptor protein is linked to gold particles, the captured protein-gold complex will appear as a pink-colored band. With the sample where the receptor protein has interacted with free beta lactam molecules, the receptor protein will not be captured at the test field and no band will occur. The color intensity of the test band is visually compared with that of the reference band: if the color intensity of the test band is weaker than that of the reference band, the sample is classified as positive [6-7] (Table 1).

**RESULTS**

Of all samples, 8(5.33%) were positive for beta lactam antibiotic residues in cow raw milk in the northwest region of Iran (Fig. 1).

**DISCUSSION**

Most of beta lactam antibiotics can be detected with Beta star kit. The result showed that beta lactam antibiotic residues in milk were 5.33 percent which is high for cow raw milk in Iran. Since this is the first report on beta lactam antibiotic residues in cow raw milk in Iran, It could be follow by other research in all of the the country.

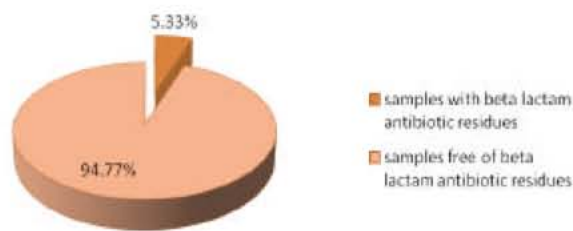


Fig. 1: Percent of positive samples for beta lactam antibiotic residues in cow raw milk in the northwest region of Iran

Movassagh [9] showed that 4.66% of Iranian ultra high temperature Milk was positive for beta lactam antibiotic residues in 2010. In 2006, Khaskheli *et al.* [10] showed that of all samples 36.5% were contaminated by beta lactam antibiotic residues in cow raw milk in Pakistan.

In Turkey, there are only a limited number of antibacterial residues studies in milk. In a study by Ceyhan and Bozkurt from a total 200 milk samples collected from Ankara region, 5.5% was positive for antibiotic residues [11, 12].

Aydin *et al.* [12] in 204 raw milk samples, 44% was positive for antibiotic residues in Turkey. Yamaki *et al.* [13] in 2686 ewe raw milk samples, 1.7% was positive for antibiotic residues in Spain.

Adesiyun *et al.* [14] studied the prevalence of antimicrobial residues in preprocessed and processed cow milk in Trinidad and showed that 10.8% of all samples were positive. Shitandi [15] showed 21% of 1109 milk samples were positive for antibiotic residues in Kenya.

In a 1988 study 71% of test samples from farms tested positive for antibiotics residues in the United States [16].

Alomirah *et al.* [17] showed 5.4% of imported pasteurized milk samples were positive for antibiotic residues in Kuwait and Out of the 308 samples analyzed for beta lactams, 62 samples (20.1%) were above the maximum residue level (MRL) (4 ppb). One hundred twenty-one samples (37.3%) were above the Tetracycline MRL (100 ppb) from a total of 324 analyzed

samples. Sixty-one samples (29.4%) were positive for Chloramphenicol (no MRL, prohibited antimicrobial drug) from a total of 207 analyzed samples.

Sasanya *et al.* [18] analyzed 384 cow milk for presence of residues of penicillin G and of all samples 13% were positive for penicillin G.

In addition to allergic reaction there is some indication in the science literature to suggest that antibiotics can induce cancer and other non-cancerous hazardous effects on the body.

This study showed that the effective monitoring program must be run in the northwest region of Iran for control of antibiotic residues presence in milk.

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