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DEPARMENT OF NUTRITION AND DIETEICS

INVESTIGATION OF ANTIBIOTIC RESIDUES IN DAIRY PRODUCTS

MASTER THESIS

OSETYA BATIBAY

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SUPERVISOR

Assist. Prof. Dr. İSKENDER KARALTI

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	Unvanı, Adı-Soyadı (Kurumu)	İmza
Jüri Başkanı:	Yrd. Doç. Dr. İskender KARALTI	
Tez danışmanı:	Yrd. Doç. Dr. İskender KARALTI	
Üye:	Yrd. Doç. Dr. Hülya DEMİR	
Üye:	Doç. Dr. Cenk SESAL	
Üye:		

ONAY

Bu tez Yeditepe Üniversitesi Lisansüstü Eğitim-Öğretim ve Sınav Yönetmeliğinin ilgili maddeleri uyarınca yukarıdaki jüri tarafından uygun görülmüş ve Enstitü Yönetim Kurulu'nun ..10../08../2017.. tarih ve 2017/15-09...sayılı kararı ile onaylanmıştır.

İmza

Prof. Dr. Bayram YILMAZ

Sağlık Bilimleri Enstitüsü Müdürü

DECLARATION

I hereby declare that this thesis is my own work and that, to the best of my knowledge and belief, it contains no material previously published or written by another person nor material which has been accepted for the award of any other degree except where due acknowledgment has been made in the text.

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LIST OF SYMBOLS AND ABBREVIATIONS

AB (Avrupa Birliği

ABD (Amerika Birleşik Devletleri)'de

CDC: Centers for Disease Control and Prevention

FDA: Food and Drug Administration

HMF (Hidroksimetilfurfuralı)

HPLC: High Performance Liquid Chromatography

l: litre

maksimum kalıntı limitleri (MRL)

Min: Minute

ml: Mililitre

µ: Mikro

OIE: World Animal Health Organization

PBS: Phosphate-buffered saline

ppb: per part billion

WHO: World Health Organisation

ÖZET

Batıbay, O. (2017). Süt Ürünlerinde Antibiyotik Kalıntılarının Araştırılması. Yeditepe Üniversitesi, Sağlık Bilimleri Enstitüsü, Beslenme ve Diyetetik Anabilim Dalı, Yüksek Lisans Tezi, İstanbul.

Süt ve süt ürünleri insanların temel gıda maddelerinden biri olmakla birlikte; veteriner ilaçları, pestisitler, mikotoksinler, ağır metaller, genetiği değiştirilmiş organizmalı ürünler (GDO), dioksin ve benzeri kimyasal madde kalıntılarını içerebilmeleri açısından da riskli ürünler arasındadır. Sütte bulunabilen bu kimyasallar endüstriyel ya da ziraai kaynaklardan gelmektedir.

Antibiyotik içeren sütler patojen mikroorganizmaların direnç kazanmalarına neden olmaktadırlar. Bazı kişilerde ise allerjik reaksiyonlar oluşturdukları için olumsuz sonuçlara neden olmaktadırlar Antibiyotik direnci tehlikesiyle karşılaştırıldığında, gıda maddelerinde kullanımına izin verilen ilaçların neden olduğu kalıntıların yol açacağı toksisite riski oldukça düşüktür. Bununla birlikte, insanların zararlı düzeylerde kalıntılara maruz kalmalarını engellemek için, AB hayvansal gıda maddelerinde veteriner ilaçlarının Maksimum Kalıntı Limitlerini (MRL) belirlemiştir. Ülkemizde ise Türk Gıda Kodeksinin 2002/30 sayılı tebliğinde "Hayvansal Gıda Maddelerindeki Maksimum Kalıntı Düzeyleri" belirlenmiştir. Çalışmamızda 40 adet süt örneği incelenmiştir. Bu örneklerden MiRA test ile kalitatif olarak; beta laktam elisa test kiti ile de kantitatif olarak antibiyotik kalıntı deneyleri gerçekleştirilmiştir.

Sonuç olarak her iki yöntem ile de 40 örnekten 4 tanesinde (% 10) pozitiflik saptanmıştır. Bütün çalışmalar incelendiğinde süt ve süt ürünlerinde antibiyotik kalıntı sorunu halen devam etmekle birlikte, çalışmamızda elde ettiğimiz düşük limitlerdeki beta laktam değerleri umut verici olarak değerlendirilmiştir. Ancak daha fazla örnek ile bu çalışmalar genişletilmeli ve rutin olarak süt ve süt ürünleri hijyen kriterleri yönünden analiz edilmelidir. Çünkü sadece antibiyotik kalıntıları değil, bunun yanında mikotoksinler, pestisitler, hormonlar, ağır metaller gibi çeşitli kirleticiler açısından da gıda maddeleri incelenmelidir.

Anahtar Kelimeler: Süt, Beta-Lactam, MiRA Test, Antibiyotik Kalıntı, Elisa

ABSTRACT

Batıbay, O. (2017). Investigation of Antibiotic Residues in Dairy Products. Yeditepe University, Institute of Health Science, Department of Nutrition and Dietetics, MSc thesis, İstanbul.

Milk and dairy products are not only one of the basic foodstuffs, are also among the risky products in terms of containing chemical residues veterinary drugs, pesticides, mycotoxins, heavy metals, genetically modified organism products (GMO), dioxin and similar chemical products. These chemicals that can be found in milk come from industrial or agricultural sources.

Milk containing antibiotics cause pathogenic microorganisms to gain resistance. They cause allergic reactions in some people and therefore cause negative consequences. Compared with the danger of antibiotic resistance, the risk of toxicity caused by the residues caused by drugs permitted for use in foodstuffs is very low. In addition to, the EU has set the Maximum Residue Limits (MRLs) of veterinary drugs in animal feedstuffs to prevent people from being exposed to residues at harmful levels. In our country, Maximum Residue Levels in Animal Food Material; has been determined in the 2002/30 numbered communiqué of Turkish Food Codex. 40 milk samples were examined in our study. These samples were studied as qualitative analysis by MiRA test; Beta lactam ELISA test kit also performed quantitative antibiotic residue tests.

As a result, positivity was found in 4 (10%) of 40 samples by both methods. When all the studies are examined, the antibiotic residue problem in milk and dairy products still continues, the beta lactam levels in the lower limits of our study have been evaluated as promising. However, these studies should be extended with more examples and milk and dairy products should be routinely analyzed in terms of hygiene criteria. Because not only antibiotic residues, but also food substances such as mycotoxins, pesticides, hormones, heavy metals must be examined for various pollutants.

Keywords: Milk, Beta-Lactam, MiRA Test, Antibiotic Residue, Elisa

I. INTRODUCTION AND PURPOSE

Milk is one of the basic sources of nutrients for all people from their first year of life. Scientific studies have demonstrated several chemical residues in milk and their potential adverse effects on human health. The most common residues in milk and dairy products include veterinary drugs such as antibiotics and hormones, pesticides, mycotoxins and dioxins. The presence of these chemical residues in milk is a potential hazard of public health since milk products are widely consumed by children and adults worldwide. For this reason, many countries have regulated the limits for the level of chemical residues in milk and dairy products (1).

In modern livestock applications, the use of veterinary medicines is of utmost importance for the efficient and safe production of meat, fish, milk, eggs and honey. The use of these drugs is subject to strict licensing and approval procedures with a system similar to that of medicinal products. In many countries, including Turkey, growth hormones and the use of drugs that release toxic residues in animal products is strictly forbidden. The presence of detectable levels of such prohibited products is considered a violation of the law. Stilbens, stilbene derivatives, anti-thyroid agents, steroids, resorkilik acid lactones and β-agonists are prohibited drugs. In addition, the use of natural horns as growth hormones is prohibited in the EU and Turkey. USA (United States) allow to use natural hormones such as melengestrol acetate and is also widely used except for the prducts exported to EU (European Union).The use of many medicines in Turkey is allowed to use despite these restrictions and prohibitions. Antibacterial substances such as sulphanamides and quinolones, anthelmintics, anticoccidials, sedatives and non-steroidal anti-inflammatory drugs are among the drugs permitted for use. Some veterinary medicines are also used as preventive more than treatment against diseases, and such medicines are generally used by mixing them with food. In order to protect human health, the obligation to leave compulsory deadlines (breakthrough time) between the application of the drugs allowed for use and the cut of the animal, the lowest levels of residues levels in foodstuffs were accepted by law (2,3)

In our study, it was aimed to investigate the antibiotic residue levels in milk that were sold in the open and sterilized by pasteurized or UHT method.

II. LITERATURE REVIEW

II.1. Milk

Milk it is a liquid that is secreted at different times in milk beat to feed newborn offspring of female mammals, and contains enough nutrients in which all the nutrients that the baby must take until it comes to the state to feed itself. The composition of the milk differs according to the environmental conditions in which the animals live. It is important in terms of calcium, phosphorus and riboflavin (vit B2). Keep it its place amino acids and fatty acids that are vital. Only components found in milk; Lactose, milk oil, casein, lactoalbumin and lactoglobulin. The energy value of the milk differs according to its compound. Milk is a protective food because of its ingredients and properties. Milk protein can buffer acid and base vapors due to its amphoteric property, binds toxic heavy metals. Because of this, chemical industry, coal mines, coal gas and workers working in boiler rooms are given continuously milk and yogurt to protect against poisoning by legal regulations.

II.1.1. The Component of Milk

The composition of milk varies according to many factors, mainly depending on the animal spice being taken. In general, the major chemical components, such as protein, fat, sugar, are present in large quantities. However, the contribution of small amounts of components to the milk properties is great. For example, vitamins are important for nutritional value.

Lactose is a single carbon hydrate, a disaccharide composed of glucose and galactose. It is found only in milk.

Most of the milk fat is composed of complex triglycerides. The chain length of the fatty acids in the structure and the rate of saturation and unsaturation vary. Other lipid components are phospholipid, cholesterol, free fatty acids, mono and diglycerides. 80% of the proteins are made up from the casein. Casein kinds are; A S1, αS2-, β-, and κ-casein, and they are phosphorous compounds. The other 20% of the proteins form serum proteins (β-Ig). Milk also contains a large number of minor proteins and enzymes.

Na, K, Ca, Mg, Cl and phosphate are the basic mineral substances in the milk, and there are also many trace elements. Milk salts may be partially ionized. A large number of organic acids, primarily citrate, are present in the form of ions or salts. Apart from these, there are many compounds in trace amounts..

The total components, except for the water in the milk, are called dry matter. Other component distinctions; Milk is fat free dry substance and dry sunstance is fat. The chemical composition of the milk significantly affects the nutritional value. There may also some microorganisms in the milk. They affect the chemical reactions and sensory qualities of the milk (4).

II.1.2. Spices of Milk

Raw milk is a milk that milking from the cow at certain intervals, cooled, taking no components in it or added no components in it, have not been previously processed (such as heating) sent to the factory for processing.

Casein milks are milk in which at least 3 to 2 total proteins are formed by casein. Like cow, sheep, goat milk. Casein milk is resistant to high temperatures. Acids and stomach secretions and yeast give big clot so it is hard to digest.

Albumin milks, albumin and globulin total is more than 3 in 1 protein amount, this milk enters the group of albumin milk. Like human, horse, donkey dog, pig milk. These milks are less resistant to high temperatures. Acids and stomach secretions and yeast give a smaller grain and soft clot, digestion is easy.

Colostrum (Mouth Milk), is the first product of breast milk starting to give. It is secreted in 5-7 days immediately after birth. The yellowish-brownish or reddish brown color has an abnormal odor and salty taste.

The mouth milk does not show a particular composition, but the composition changes continuously as time progresses. Compared to normal milk, the dry matter, casein albumin, globulin and mineral substances are rich but fat and sugar are poor. Colostrum is particularly rich in protein fractions. The greatest feature of colostrum is that the amount of globulin is very high. About 17% of the globulin is also involved in the transport of antibodies. Immunoglobulins from these antibodies are immunogenic, thus allowing the offspring to gain resistance to external influences and diseases. The amount of immune substances in the colostrum is largely dependent on the number of lactations of the mother. Use of them shows a rapid decline as the calf ages. Magnesium

salts and catalase are rich in peroxidase amylase and lipase-like enzymes. Vitamin and antibody levels are high. Vitamin A in particular can reach 20 times the amount of normal milk (4).

II.1.3. Factors Affecting the Formation of Milk

The changes that occur after milking are usually microbiological. The chemical composition of the milk may vary from cooling, storage and applied technological processes. Also; Drugs used in the treatment of disease, antibiotics, pesticides, detergents and disinfectant residues, pregnancy and anger states, duration of driying and body secretions are also effective. Factors that affect the composition of raw milk before and during milking are:

a) Race of Animal

The ratios of milk yield and of the ingredients in the composition are dependent on the races. Differences can be found even among different individuals of the same race. For this reason breeding studies have been carried out to breed races with high fat content.

b) Lactation

The lactose ratio during lactation is generally constant. However, fat and protein fall in the first 3 months of lactation and increase towards the end.

c) Age of Animal

Metabolism starts weakening as the animal gets older thus ability to synthesize milk lost at certain extent. As a result, a small amount decrease in the dry matter rate is observed.

d) State of Health of Animal

The state of health and diseases of the animal causes the animal to weaken and the efficiency to decrease. In some diseases, such as breast infection (mastitis), the composition of the milk varies. In mastitis infection, microorganisms pass through the alveoli. The ability of milk-producing cells to synthesize fat, protein, lactose is impaired. Other changes in the milk: Since the osmotic pressure must be kept at a

certain level, some ions formed in the blood pass through in to milk and the amount of chlorine in the milk increases. At the same time, there is a decrease in the amount of lactose. Along with the increase in the amount of chlorine, electrical conductivity of the milk increases. Partially damaged tissues pass through the serum protein form and increase their amount. The amount of catalase enzyme and the viscosity increase. Acidity of milk reduces. Mastitis also creates problems technologically. Milk's cloting with cheese and yoghurt yeast is difficult and the yield is reduced. Microorganisms causing this disease: Streptococcus agalactiae, Streptococcus disagalactiae, Streptococcus salivarius, Staphylococcus aureus, Corynebacterium pyogenes, Clostridium perfringens, Aerobacter aerogenes.

e) Temperature (season)

Optimum yield is usually obtained at a temperature of 5 to 20C. Particularly at high temperatures, when relative humidity is high, there is a decrease in the fat rate.

f) Time of Milking and Style of Milking

The milk yield of an animal increases with increasing daily milking number. It should be milked 3 times on average in a day. With proper milking, massage will be good for animal's breast and mammary glands and increases the efficiency. It is important that the milk is completely milked during the milking.

g) Food

Substances are taken from the body through blood during the synthesis of the milk. If these substances are not given to the animal constantly, the animal meets the necessary substances from its own body and the animal is getting weaker. After a while, the yield decreases considerably and the composition becomes weaker. The ability of animals to benefit from feed is a genetic feature. Some of them turn the food into better milk and meal. Green fodder, corn and wheat bran, sunflower pulp increase milk yield. Rape and sesame reduce yield (4).

II.2. Unwanted Substances in Milk

Milk is a clear to contamination food for various reasons. Foreign substances may be present in the composition of the milk from various sources that are not naturally present. Being present in component of milk unnaturally, not added during the processing, but the substances that are consumed with this milk and its products from various sources are referred to as foreign substances or residues and contaminants. As an example of these resources; Many substances that are transmitted from the soil, from the grass and the air, and industrial wastes, some of the ingredients used in animal feeding, some medicines used to control disease in livestock, and some of the substances that pass the milk, some of the residues after cleaning and disinfection. But on the other hand, the same expression is often used for substances that are added to the substance for reasons such as cheating or unfair earnings that are not required to use technology, In fact, such items need to be used in the form of foreign materials that participate in fraud. Due to the danger of human health, amount in the product is restricted to legal regulations in many countries (4).

II.2.1. Antimicrobial Substances

The presence of antibiotic residues in milk is important for economic, technological and human health. The reasons for using in dairy animals are:

1. In the treatment of infectious diseases, it can be given by injection or drinking water.

2. During periods when there is infection threat; It can be used when there is threat, when the production is intensive or at the time of transplant or when the end of lactation. This is called strategic application.

3. It is applied at certain times to prevent the disease from reaching the clinical level and to prevent its spread in the flock.

4. It is used to increase the rate of utilization of food, to increase growth rate and milk yield. The dose is very low amount here.

II.2.1.1.Use of Antimicrobial Substances in Food

The effect of antibiotics on bacteria is two-ways;

a) Bacteriostatic effect that prevents the growth and proliferation of bacteria
b) Bactericidal effect that kills the bacteria

Many antibiotics, especially penicillin, are used in the treatment of mastitis in dairy animals. Micrococcus and Strptococcus species infiltrate the milk ducts in the breast, causing inflammation of the mammary glands. 30-80% of the antibiotic used is absorbed by the tissue, the rest is thrown out through the blood and lymph through the back. In the treatment of mastitis apart from penicillin; Streptomycin, chlortetracycline, oxytetracycline, simplexin, chloramphenicol, neomycin, polymyxin, subtilin, clormycitin are used. The drug is given directly through the breast or through the veins, muscles, or mouth. Very little pass in the second given.

As a general rule, an animal's milk treated with antibiotics should not be used for 4 days following treatment.

The pass rate of antibiotic to milk / existence in milk depend on the following factors

1. The state of the disease: antibiotics given to healthy animals are throw out in a short time

2. Kind of Antibiotics: for example continues to pass in to milk in penicilin for 72-92 hours.

3. Number of Milking: The transitions will be more for the first milkings. In general after injection the transition of antibiotic to milk start between 10 minutes and 4 hours.

4. Dose of antibiotic

5. Given style: Given from breast are more common than muscle, vein, and oral given.

6. Season: The amount is higher in the winter and spring months when the diseases increase (4,5,6).

II.2.1.2. Why is Comsuption of Antibiotic Milk Harmful?

1. It causes allergic reactions in terms of health-sensitive people.

2. Microorganisms do not gain resistance to antibiotics in humans as a result of continuous consumption of anibiotics milk.

3. Problems arise in the production of products such as cheese, yogurt, butter in milk technology and quality control of milk.

Yoghurt bacteria are very sensitive to antibiotics. The symbiotic relationship between bacteria is deteriorated, acidity development slows down and the incubation period extends.

Antibiotic milks in cheese technology are not at a level that can prevent them during production. However, it causes some quality disorders during ripening period. As the initial heat treatment in cheese production does not make all the antibiotics inactive, the activity of the starter bacteria added to the cheese is slowed down. As a result, development of sufficient acidity can not be obtained, the whey is not sufficiently separated from the clot, and the soft cheese is obtained by leaving a lot of water in the clot. On the other hand, antibiotic-resistant bacteria during both ripening and in processing, lead to taste and structure disorders as a dominant mediator. For example, coliforms form gas by multiplying. Also, in hard cheese, the butter bacteria that are more resistant to antibiotics during ripening cause the formation of holes and cracks. Some lactic acid bacteria may naturally produce antibiotics in the milk as a result of their activity.

Bacteria used in butter production are also affected by antibiotics. The acidity is slowed down, the churning time is long, and the taste and aroma substances do not occur in sufficient quantities.

Antibiotic residues in the other dried and condensed products will be a threat in terms of public health when they are concentrated.

The results of microorganism test are misleading because of the effect on the microorganism in antibiotic milks. The antibiotic in the milk prevents the phosphatase test. The pasteurized milk gives blue colour due to penicillin and tetracycline and processed as if it is not pasteurized. (4,7).

II.2.2. Radioactive Substance Residues

Radioactive isotypes of some elements are found in milk and almost all food. It is called radioactive substance residues from atomic reactors, nuclear tests, centrals and weapons that are transmitted to nature (soil, plant, water), then animal.

There are two types of radioactivity:

a) Natural radioactivite: The radiation generated by the natural radionuclides in the interplanetary space is naturally found in the human body in the environment. They have nothing to do with the contagion.

b) Unnaturel radioactivite: Radioactive substance radiate from TV, X-ray apparatus and equipments that have entered our daily life and nuclear tests are residues that are emitted from central and atom reactors and are also named as radioactive fallout. In a nuclear explosion, degradation products come out at high altitudes, spread by air currents and come down to the earth with precipitation; and contamine to soil, plants, air and water. In this way, it is absorbed by the mucous of the digestive and respiration organs of the animal, and through the blood, contamine to the body and mammary glands.

The disintegration rate of these substances is important. There are some definitions.

Physical half-time: Time for the amount of isotope to reduce by half .

Biological half-time: It's a time that half of the radioactive material taken in the body will be thrown away from the body in natural ways. It is important for nutrition. Atomic bomb tests Radioactive Sr and I, nuclear reactor accidents also cause Cs contamination. Sr is the longest remaining element in the body, and since it shows the same properties as Ca, it is found in the milk and its products in excess. 38% of the Sr taken with all foodstuffs is taken with dairy products. Much of the iodine in the milk is present as dissolved iodide. Cs in the milk is like K + and Na +. If there is a possibility of contamination of these substances to milk, the best solution is to treat the products with low radioactive element concentration. The presence of Ca and K in the butter indicates that Sr and Cs will pass to the butter in small quantities. Sr 1% Cs 2% in the milk passes to butter. Since Sr is related to Ca, 45-83% of the Sr in the milk passes to cheese. Ion exchangers, electrodialysis is used to clear the contaminated milk. The binding agents such as potassium perchlorate can be mixed in the food or calcium phosphate supplements can be added to milk.

II.2.3. Heavy Metals

Milk and its products contain toxic and non-toxic metal ions in different chemical structures. They naturally originate from pasture feeding techniques and contamination from industrial and human activity. The most important elements are As, Cd, Pb, Hg, Sn.in terms of food safety. However, in terms of nutrition, Cu, Fe, Se, and Zn are also important at normal concentrations.

Negative effects of heavy metals;

Lead: neurophysiological development

Cadmium: nephrotoxic

Mercury: nerve poison, nephrotoxic

Chromium: genotoxic

Selenium: nerve poison

Arsenic: carcinogen

Where can Heavy Metals Contaminate From?

1. Equipment for milk and milk products, boilers, boats, etc. Here, the passage of Metaloids; Chemical reactions occurring on the surface and electron exchange on the metal surface. Lactic acid in products also accelerates corrosion

2. Use water is also a good source of contamination. The amount of Fe can be found in domestic water are limited to 0.3 mg / Lt

3. Contamination of food with high / high levels of metal

To prevent contamination;

1. 1. Industrial production zones and agricultural-based industrial zones should be separated
2. Disposal of food containing metal ions above the maximum concentration
3. Milk and its products should only be in contact with inert surfaces (4,8).

II.2.4. Cleaning Materials and Disinfectants

Detergents are used to clean all kinds of spaces where milk and products come into contact and disinfectants are chemical substances used for the purification of microorganisms from any surface that they come in contact with until they are packaged from the production of the milk. After this process, when the cleaning and disinfectant materials are not removed from the surface effectively, the residues also pass to milk. Cleaning is not only carried out in production units but also during production, storage, transport and marketing of raw milk. When the surface disinfectant residue comes into contact with the surface, it also contaminate to milk. These residues especially affect the development of lactic acid bacteria negatively. For this reason, after cleaning and disinfection rinsing with water can achieve a more positive result if hot water and steam are observed. Measures to take:

1. Develop more suitable compounds to soften water
2. Develop new formulations for cleaners
3. More effective sanitation methods
4. Measures to avoid the use of corner materials by developing the used tool equipment
5. Proper use of both equipments and cleaning agents

II.2.5.Pesticides

In agricultural activities, pesticides are called chemical compounds that are used to kill pests and harmful diseases and combat diseases, pesticide residues are called drug residues that are transmitted to milk and milk products. They are possible to be examined in 6 groups:

1) Insecticide: kill the insects
2) Fungicide: parasitic fungi
3) Hetbicide: weeds
4) Molluscicide. insect mollusc
5) Nematocide. effective on parasites
6) Rodenticide. against rodent

In agriculture, arsenic, nicotine sulfur and copper medicines were used against harmful organisms until 1940s. DDT and organic phosphorus compounds were then used. The expected toxic effect against pesticides expected from pesticides should not

be seen in humans and animals. It has been proven that the residues are harmful for human health. It has been determined that pesticides retain their properties for a long time in the soil. For example; DDT continues its activity within 10 years, dieldrin 8 years, lindan 6.5 years. Water solubles are not excreted by urine and feces, whereas oil solubles are stored in fatty tissues. There is no direct use of pesticides in milk and dairy products. Dairy animals are usually take the pesticides with food. It passes to milk through the blood and then to the people. Depending on the conditions of use, residues remaining in the soil accumulate in the tubers of the plants such as patatoes end beets and in the leaves.

There may be pesticides in the food crops of grassland, drinking water. Pesticide residues entering the animal's body through the skin contaminate to milk and from there to human. If you drug for the parasites in the barn and if the animals are taken to the barn immediately, or if you do when they are inside, it passes to animal through the skin. Medicines used to combat animal parasites also cause transmission through the bloodstream. The use of contaminant additives in composite dairy products also causes transmission (eg strawberry yogurt). The technological processes applied to the milk are not very effective on pesticide residues. If it passed to the milk ıt will definetely pass to product. In the case of chronic poisoning and environmental pollution in humans, strict control of this subject is required..

85 pesticides were identified in the milk and products by the subcommittees of the Codex Alimentarius commission pesticide and drug residues from the committees affiliated to the FAO / WHO organization and Limitations have been introduced regarding their residual levels (4).

II.2. 6. Mycotoxins

Mycotoxins are mold metabolites that cause pathological changes such as mutagenic, carcinogenic, teratogenic, acute and vetoxic effects in humans and animals. Milk and dairy products are the most sensitive group for the contamination of mycotoxins. Aflatoxin M1 and aflatoxin B1 are mycotoxins which are the major milk contaminants. Contamination takes place in 2 ways:

Indirect contamination: Containment occurs with consumption food of dairy cattle. Aflatoxin B1 is synthesized by Aspergillusflavus and Aspergillus paraciticus in agricultural products (Oilseeds, cereals, dried coconut) which are used as feed and feed

additives under appropriate temperature water activity and nutrient conditions. In Milk, M1 is the 4th hydroxy derivative of B1. For this reason, restrictions have been introduced regarding aflatoxin levels in statutes and regulations.

Direct contamination: Contamination of dairy products. In particular, aflatoxins which are both benefited from maturation of cheese and are the result of unwanted mold contamination of cheese. Certain strains of Penicillium roqueforti and Penicillium camemberti are used in mold-ripened cheeses. These types of cheeses include mycotoxins such as mycophenolic acid, rocfortine, cyclapiazonicacid. Also ocratoxin A secreted by Penicilliumssp kinds, penicillic acid and patulin are the other mycotoxins on the cheeses. Other than these, sterigatocystin, synthesized by Aspercillus versicolor, is also a contaminant found in hard cheeses.Food may also contain fumonisin B1 and B2 from fusarium toxins (4).

II.3. Analysis Methods Used in Monitorıng Drug Residues

There are numerous and different types of drug residues in animal wastes, and many analyzes are required for effective monitoring. For this reason, the use of screening assays is essential (9).

In the identification and characterization of microorganisms, a long-standing antibody-antigen reaction is applied. Immunological methods are preferred when determining food contaminants such as mycotoxins, pesticides or veterinary drugs with low molecular weight. The antigen-antibody reaction is a powerful system for rapid identification of all pathogens. Some systems are automatized at high speed while others are simple to use. These tests can be classified as follows (10,11).

II.3. 1. Latex Agglutination Tests

Antibody coated and dyed latex or colloidal gold particles are used for rapid serological identification or identification and typing of pure bacterial culture isolates. Visible agglutination is formed after antigen and antibody conjugation. The reverse latex agglutination tests are for soluble antigens and are often used to search for toxins (11).

II.3. 2. Automatic and Manual ELISA (Linked Immunosorbent Assay) Methods

The most common application is the use of Enzyme-Linked Immunosorbent Assay (ELISA) systems. This technology is extremely sensitive due to the use of antibodies developed according to the target molecule. Because of its high orderability, the results of the analysis are reliable. It is possible to analyze a large number of samples for different drug residues as soon as they contain easy sample preparation procedures. In this method, the antibody is labeled with an enzyme and the immunological reaction is measured as an enzymatic activity result. The most commonly used sandwich ELISA method is the ELISA test, although there are different forms such as direct, indirect and sandwich ELISA. Several ELISA tests have been developed to identify pathogenic microorganisms and toxins Many ELISA kits currently in use have a high standard and automatically increase speed and efficiency for their work and reduce human error (11).

II.3. 3. Lateral Migration Immunoassay Method

Another development in the field of immunology is the use of Laterel Flow Technology which is based on the antigen-antibody relationship. It's a real quick test. Bacillus anthracis has been developed for the rapid detection and identification of various samples of pathogens such as E. coli 0157, Salmonella, Listeria and Avian influenza (11).

II.3. 4. Immuno-Magnetic Separation (IMS) Technology

The IMS system saves at least one day from the enrichment and pre-enrichment steps by identifying pathogens from the grains. Recently several diagnostic systems (ELISA) have been combined with the immuno-magnetic coating system. In this case, the incubation period was shortened and the sensitivity increased (11).

II.3. 5.Fast Scan Kits

Some fast screen kits are available for the detection of antibiotic residues. MeRA test is one of them. Some antimicrobial agent groups, such as beta-lactams and tetracyclines, are sensitive to heat; Molecules belonging to these chemical classes are inactivated shortly at the growth temperature of thermophilic bacteria.The MeRA test

involves a rapid pre-incubation step that allows growth and multiplication of Geobacillus stearothermophilus. Following this step, the interaction between the vegetative form of *G. stearothermophilus* and heat-sensitive antibiotics, if present in the sample, is carried out at room temperature. Finally, the test tubes are subjected to a final incubation and color change is observed (12).

II.3. 6. High Performance Liquid Chromatography (HPLC)

Chromatographic methods can be described as separation techniques involving mass transfer between the stationary phase and the mobile phase. One of these methods, liquid chromatography, Although this method was found at the beginning of the 1900's, it has been the subject of extensive research for the development of methods for residue analysis since 1960's. HPLC is a very sensitive method in which the liquid phase soluble chemical substance mixture can be easily and rapidly separated into its components. Today, HPLC is widely used in many areas. Its primary uses are chemical separation, purification, identification and concentration determination (13,14).

II.4. Microbiological Criteria for Milk Hygiene

Standards Needed for Raw Milk and Microbiological Criteria: There are some Standards and microbiological criteria while collecting and accepting raw milk from production facilities:

A.Raw cow milk: Raw cow milk to be used in the production of heat-treated drinking milk, dairy products and milk-based products must comply with the following standards:

Total number of live bacteria 30°C (ml'de) ≤ 100.000*

Somatic cell count (ml'de) ≤ 500.000** *

*Geometric average of at least two samples per month for a period of two months

** Geometric average of at least one sample per month and a three month period

B. Raw Mandate Milk: The raw banana milk to be used in the manufacture of dairy products and dairy based products must comply with the following standards:

Total number of live bacteria 30°C (ml'de) ≤ 1.000.000*

Somatic cell count (ml'de) ≤ 500.000**

* Arithmetic average of at least two samples per month for a period of two months

** Arithmetic average of at least one sample per month and a three month period

C. Raw Goat and Sheep Milk: Raw goat and sheep milk to be used in the manufacture of drinking milk, dairy products and milk based products must comply with the following standards:

Total number of live bacteria 30°C (ml'de) ≤ 1.000.000 *

* Geometric average of at least two samples per month for a period of two months (Good Hygiene Practices Guide for Raw Milk Production, 2010).

II.5. General Criteria for Milk Hygiene

Drug use in animals, It should be in accordance with Ministry of Agriculture and Rural Affairs 2005/74;Circular on Control of Drug Use in Animal Husbandry Established by Food-Rated Animals. The milk obtained from animals treated with veterinary medicaments which are supposed to be administered on top of each other must be disposed of for a specific period of time for each veterinarian, should not sent to milk processing facility. The amount of the drug, the day it was given, and the identity of the animals applied should be kept and maintained veterinarian and / or animal owner. To be added to animal feed Permitted food additives, premixes should be used within the context of Communiqué on the Production, Import, Export, Sale and Use of Feed Additives and Premixes; of the Ministry of Agriculture and Rural Affairs no 2002/66.

Before the use of pesticides; Efforts should be made to prevent the presence of insects, flies and rodents. Although stables and milking places are places that attract insects, precautions should be taken such as building the building according to insects control, cleaning and maintenance, removing fecal waste and therefore the number of insects can be reduced. No fertilizer accumulation should be allowed, especially near the milking areas.

Animal food stores are places that attract rodents. Therefore, feed storage should be placed at an appropriate place and the food should be stored in suitable closed containers (containers) to ensure protection against insects. Measures must be taken in food deposits and effective insects fighting should be done. When using chemical medicines such as mouse poison, it is important to note that these products are officially approved for use in food businesses and it must be used in accordance with the manufacturer's instructions. When it is necessary to apply chemical insects control measures, it should be noted that these products are authorized and approved for use in food enterprises, it must be used in accordance with the manufacturer's instructions. Insects control chemicals should be stored so as not to cause contamination in the milking areas and in the vicinity. These chemicals should not be stored in damp or close to food deposits. The use of solid snack foods should be preferred. Insects control chemicals should never be applied during the milking process.

Milking should be carried out on hygienic conditions. Before milking the animal, the nipple, nipple, groin, flank and abdomen should be cleaned. Tools and equipment used at milking should be cleaned and disinfected. Damage to the breast and nipple tissue during cleaning and milking should be avoided. In order to prevent contamination of animal wastes or dust to milking equipment and the milking environment, operations such as feeding of animals and removal of animal litter should not be performed before the milking. The person milking should carefully monitor the conditions of the animals being milking; should control each animal for the volume for sensory and physicochemical signs; examine animal records. In an abnormal situation, the consumption of milk should not be presented. The first milk from each nipple should be thrown away or collected separately and not used for human consumption, as the safety and quality of the milk are not affected. Instruments and equipment that come into contact with milk used at the right should be immediately cleaned, disinfected and stored under appropriate conditions following their use. The person who makes a milking person should wash his arms, wrists and elbow between his arms before milking and keep it clean throughout the milking process. For this purpose, the person who is milking near the milking place must have a suitable sink and disinfectant to wash his / her hand and arm.

Places where milk is cooled or stored, storage tanks, stumps and other related equipment should be designed and constructed to prevent the risk of contamination. It should have floor and walls that can be easily cleaned and disinfected. It must be resistant to corrosion. Milk should be a feature that can not be a material transition in quantities that will cause a health risk to the consumer. Liquid waste must have a ground that can be easily trained. Appropriate ventilation and lighting conditions must be avaliable. All sources of contamination, such as toilets and fertilizers, must be distinguished. Connection devices must be easy to wash, clean and disinfect. There must have appropriate milk refrigeration equipment. It must be protected against negative external factors such as pesticides. Storage tanks and stools should not be used to store any harmful material that may cause contamination of the milk. Storage tanks and stools should be regularly cleaned and disinfected adequately.

Immediately after milking, milk should be collected in a clean place without adverse effect on the quality of the milk. and if the milk is not collected within 2 hours after milking, it should be cooled to 8 °C, if collected daily below 8 °C, and if not collected daily below 6° C. If milk is not transported to processing and production facilities within two hours after milking, it should be ensured that the temperature does not exceed 10 °C while being transported to processing and production facilities. However, cooling may not be performed if it is delivered within two hours after milking.

The collection center should be designed and operated in a way that prevents contamination of the milk. The milk must have cooling equipment or appropriate equipment to cool it, if it is stored in the milk collection center, it should have cold storage equipment. If the milk is purified in the storage center, there must be other separators or other equipment suitable for physical purification of the milk. Adequate precautions should be taken at the entrance of staff and vehicles to the collection center so that the milk hygiene is not adversely affected. In particular, access to the collection center must be clean from fertilizers, silage, etc. The milk collecting / transport operations should not make pathogenic microorganisms causing the disease susceptible. The milk collector / carrier must have received adequate training in the protection of raw milk hygiene and should wear clean clothing. When the milk collector's clothes and shoes are contaminated with fertilizer, the contaminated clothing and shoes must be cleaned or cleaned before continuing to work. Milk collector / bearer must not enter

where the animals are and where the fertilizer is located. Before collection, the milk collector / conveyor must check each manufacturer's column to determine if there are signs of spoilage or signs of spoilage. Milk should not be collected if there are signs of deterioration and spoilage in the milk. Milk should be collected under hygienic conditions. Milk collector / carrier must not enter production locations in milk operation. You should only communicate directly with the relevant dairy operation personnel without passing through the milk processing areas or the staff dealing with processed milk and dairy products, distribution of milk samples, dressing, resting place etc. rules must be set out in order to give necessary permission for. (15,16).

II.5. Antibiotics

Antibiotics are naturally occurring substances that inhibit or kill bacteria. They are grouped according to various characteristics. Frequently used antibiotics in milk and dairy products are beta lactam group.

II.5. 1. Beta-Lactam Group of Antibiotics

Beta-lactam antibiotics; is a wide range of antibiotics group with different pharmacokinetic properties, antibacterial domains, chemical structures. Common characteristics of the members of this group are; The presence of a beta-lactam ring in all of its structure, the mechanisms of action, and the ways of resistance against them. Antibiotics included in this group; Penicillins, cephalosporins, monobactams, carbapenems and betalactam / betalactam inhibitor combinations. All beta-lactam antibiotics; Blocking the transpeptidase activity of penicillin-binding proteins (PBP) responsible for cell wall synthesis in bacteria, inhibiting peptidoglycan synthesis. As a result, the bacteria that can not synthesize the cell wall lysis and die. Beta-lactam antibiotics are bactericidal.

II.5.1.1.Penicillins: It was first recognized in 1928 by Fleming that a fungus called Penicillium notatum secreted an antibacterial substance that caused the staphylococcus lysis, and this was called penicillin. It was then purified in the 1940's and used clinically.

II.5.1.2. Cephalosporins: Cephalosporins are divided into four groups by considering the spectrum of action. Roughly speaking, as the number of generations increase Gram-negative activity increases, decreases, Gram-positive activity increases.

II.5.1.3. Monobactatns: This group of antibiotics is the only example of clinical use of aztroenam. It Has strong Gram-negative activity.. However, there is no activity against Gram-positive bacteria and anaerobic bacteria. It is a narrow-spectrum antibiotic. Therefore, it should not be used alone in polymicrobial infections and in the empirical treatment of serious infections of unknown cause. Bacteremia / sepsis, nosocomial pneumonia, nosocomial urinary system infections, osteomyelitis, septic arthritis caused by susceptible Gram-negative bacteria constitute are the main clinical uses.

II.5.1.4.Carbapenemler: It is the group with the broadest influence among the antibiotics currently available. Two agents in clinical use in this group were imipenem and later meropenem. There is no significant difference between domains. Meropenem Gram-negative, imipenem Gram-positive activity is slightly more. There is little difference between the activities of the self-airway.

II.5.1.5 Beta-Lactam/Beta-Lactamase Inhibitör Combinations: The most common resistance mechanism that bacteria use against beta-lactam antibiotics is the production of beta-lactamase enzymes that inactivate these antibiotics. These enzymes can neutralize one or several of the penicillin, cephalosporin, monobactam or carbapenem groups by enzymatic hydrolysis. There are many different and different forms of beta-lactamase enzymes synthesized by different bacteria under the control of plasmid or chromosome. They are increasingly a clinically important problem. Beta-lactamase inhibitors have been developed for this purpose. However, it is known that these inhibitors are not effective against all enzyme types (17,18).

III. MATERIAL AND METHODS

III.1. Material

For our work, 20 pieces of 20 different brand milk samples were collected. Packed samples were bought from the market, while open samples were bought from the dairy farm. Accepted samples were delivered to the laboratory in sterile conditions in the cold chain conditions and stored at 2-8 ° C until the day of operation.

III.2. Method

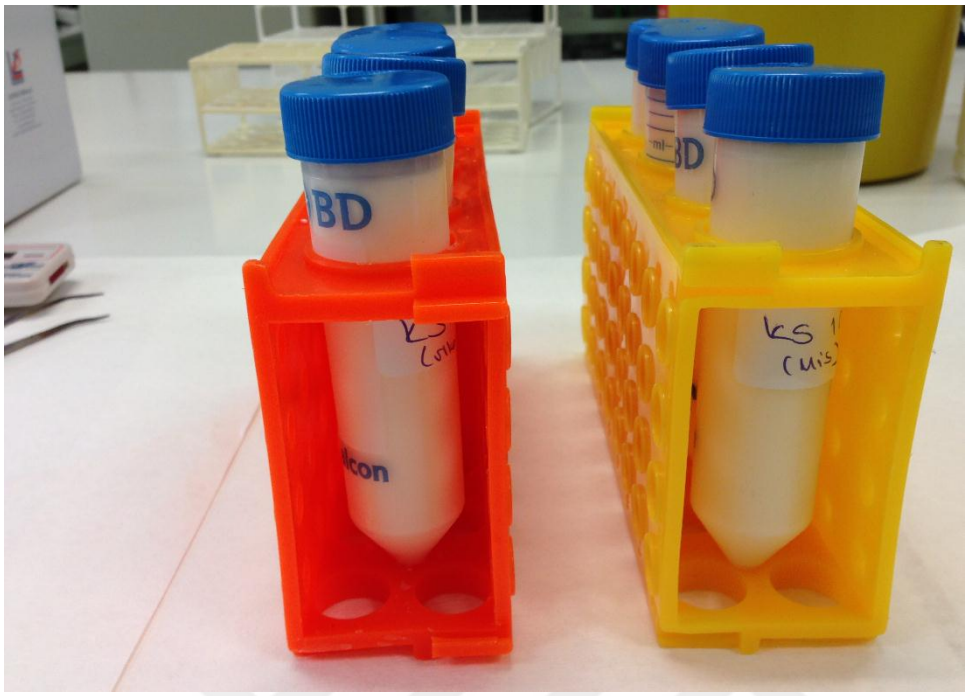
Firstly, the antibiotic residues in the samples were qualitatively determined by MiRA Test and then beta lactam level is quantitated by by elisa. Beta lactam group antibiotic residues are frequently found in milk and dairy products.

III.2 .1. Antibiotic Residue Assay Method with MiRA Test Kit

First, the screening test was performed with Mira Test. It is a microbiological test containing Geobacillus stearothermophilus spores for the detection of antimicrobial substance residues in milk.

Some antimicrobial agent groups such as beta-lactams and tetacyclines are sensitive to heat. Molecules belonging to these chemical classes are inactivated shortly at the growth temperature of thermophilic bacteria. The MiRA test involves a rapid pre-incubation step that allows growth and multiplication of G. stearothermophilus. Following this step, the interaction between the vegetative form of G. stearothermophilus and the heat-sensitive antibiotics, if present in the sample, is carried out at room temperature. Finally, the test tubes are subjected to a final incubation. This incubation step of the MiRA test is a critical step in achieving extremely low detection limits. The work steps are described below:

1) 2 examples were taken from the milk samples to be tested and 6 ml of distilled water was transferred to a 10 ml test tube (milk: water ratio 1: 3) (Figure II.1).



Resim III.1: The appearance of milk samples

2) The test sample in the tube was homogenized with the vortex for a few second

3) The homogenized sample was centrifuged at 4000 rpm for 15 min.

4) One sports disk was added to the food solution.

5) Pre-incubation of the sports diskin was performed with a 20 min solution at 64 ° C (Figure III.2).

6) After the incubated solution reached room temperature after incubation, 1ml of homogenized supernatant (test sample) was transferred into the incubated solution, The solution was allowed to stand at room temperature for 20 minutes in order to effect the antimicrobial agent in the test sample (if present).

7) The test sample was incubated for 3 to 3. 5 hours on a water bath or thermoblock at 64 ° C.

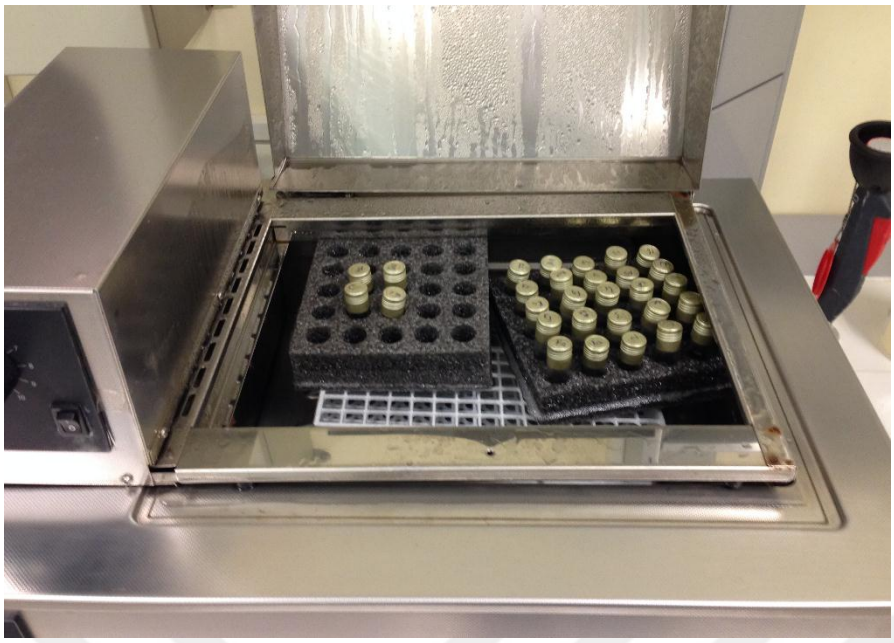


Figure III.2.: MiRA Test Water Bath Stage

8) If there is no discoloration after observing the color change in the tube (Blue-Green Color): The concentration of the antimicrobial agent in the sample is accepted above the detection limits. If there is color change (Yellow Color): No antimicrobial agent or concentration is accepted below the detection limits (Figure III.3) (12)

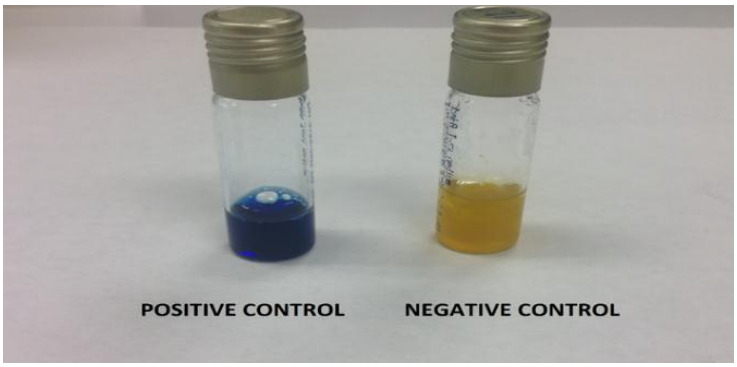


Figure III.3. Positive Control ve Negative Control

III.2.2. Elisa Testi:

At the same time milk samples were analyzed for the antibiotic residue levels of the beta lactam group by elisa method. .

III.2.2.1. Beta Lactam Elisa Test Working Method:

Preparation of Samples: 50 ml was taken from milk samples were transferred to tubes and 20 ml of 50 mM succinic acid was added. Then the balcony tubes were shaken in a 15 min shaker incubator at room temperature. It was then centrifuged for 15 min at 4000 g for 15 min. The supernatant after centrifugation was diluted 1/10 (100 µl supernatant, 900 µl PBS-Phosphate buffered saline). 50 µl of this mixture was used in the experiment.

Preparation of Standards: Standards were diluted bucause of they are concentrated. Each standard (50 µl) was diluted with 450 µl sample buffer 1. Standards were prepared on the working day because tehey must be fresh.

Preparation of Elisa Test: The solution and plate in the kit were brought to room temperature before the operation and the following steps were followed step by step and the operation was completed (Figure III.4).

Figure III.4: Elisa test kit

1. The test sample was placed in the plate as much as the sample and standard number.
2. Standard and samples were pipetted into 50 µl wells respectively.
3. 50 µl anti-tetracycline antibody was pipetted into each wells. It was incubated for 1 hour at room temperature.
4. In the automatic elisa washer, 250 l wash buffer was washed in each wash 3 times.
5. 100 µl of conjugate was added to each buffer with the help of a multi-channel pipette, shake, and incubated for 15 min at room temperature (Figure III.5).
6. In an automatic elisa washer, 250 l wash buffer was washed 3 times in each wash.
7. 100 µL of sucrose / chromogen was added to each wells, shaked, and incubated at room temperature for 15 minutes
8. 100 µl stop solution was added and Elisa was read in reader using a 450 nm filter. A standard curve graph was drawn using the Rida Soft Win program Absorbance sample / zero Values of tetracycline values in ppb were calculated using the standard absorbancex100 formula (19)

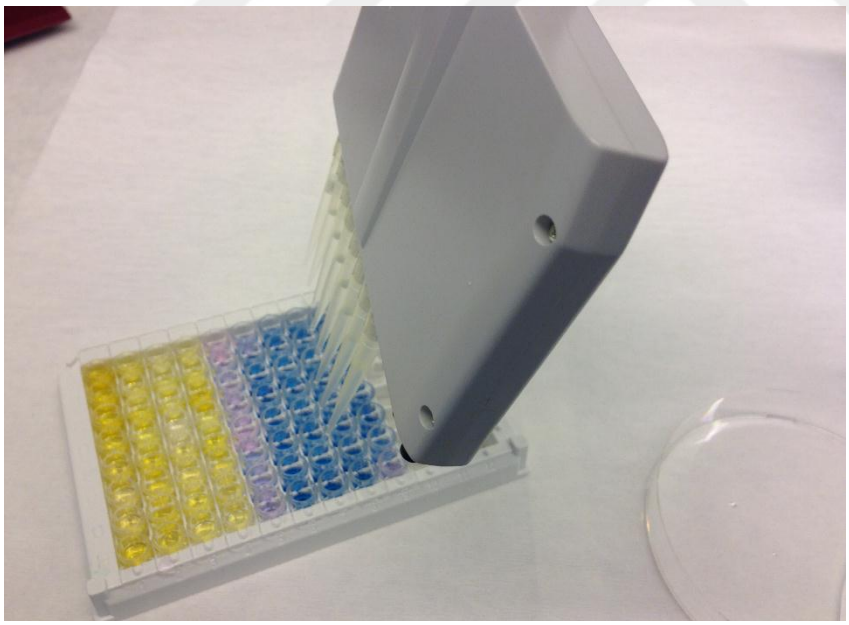


Figure III.5: Study of elisa test

IV. RESULTS

IV.1. Qualitative Antibiotic Residue Test Results

The MiRA test qualitatively detects antibiotic residues. Apart from the beta lactam group, this test kit can also detect traces of antibiotics, including tetracycline, and other antibiotic groups such as macrolides. These antibiotics and the limits of detection of this kit are given in table IV.1.

Table IV.1. Maximum Acceptable Limits and MiRA Test Kit Detection Limits.

ANTIBIOTIC/SULPHAMIDE	*MRL* for milk [1] MAXIMUM RESIDUE LIMITS (µg/kg)	**MiRA Test** Sensitivity in 3h 30' DETECTION LIMITS (µg/kg)
BETA-LACTAMS		
Penicillin G	4	2-4
Ampicillin	4	2-4
Oxacillin	30	<10
Cloxacillin	30	<10
Dicloxacillin	30	<10
Amoxicillin	4	2-4
Benzylpenicillin	4	2-4
Nafcillin	30	15-30
Penethamate	4	2-4
Cefalexin	100	50-100
Cefazolin	50	25-50
Ceftiofur	100	50-100
Cephapirin	60	5-10
Cefquinome	20	10-20
TETRACYCLINES		
Tetracycline	100	50-100
Clorotetracycline	100	50-100
Oxytetracycline	100	50-100
MACROLIDES		
Erithromycin	40	<10
Tylosin	50	25-50
Tilmicosin	50	20-40
Spiramycin	200	100-200
LINCOSAMIDES		
Lincomycin	150	75-150
Pirlimycin	100	<50
AMINOGLYCOSIDES		
Gentamicin	100	50-100
Neomycin	1500	<100
Streptomycin	200	<100
Dihydrostreptomycin	200	<100
SULPHAMIDES		
Sulfadiazine	100	<150
SULFANILAMIDES		
Sulfadimidine	100	<200
BENZIL PIRIMIDINE		
Trimethoprim	50	25-50
QUINOLONES		
Flumequine	50	50-100
Enrofloxacin	100	50-100
NOVOBIOCIN		
Novobiocin	50	100-200

According to the MiRA test results, positivity was found in one of the packaged samples and 3 in the open milk samples (Table IV.2.).

Table IV.2: MiRA Test Results

Mira Test Results			
Packed Milk Sample No	**Result**	**Unpacked Milk Sample No**	**Result**
1	Negative	1	Positive
2	Negative	2	Positive
3	Negative	3	Positive
4	Negative	4	Negative
5	Positive	5	Negative
6	Negative	6	Negative
7	Negative	7	Negative
8	Negative	8	Negative
9	Negative	9	Negative
10	Negative	10	Negative
11	Negative	11	Negative
12	Negative	12	Negative
13	Negative	13	Negative
14	Negative	14	Negative
15	Negative	15	Negative
16	Negative	16	Negative
17	Negative	17	Negative
18	Negative	18	Negative
19	Negative	19	Negative
20	Negative	20	Negative

The samples studied with this test kit are evaluated according to the color change. At the end of the incubation, the blue-green tube was positive; yellow ones are considered as negative (Figure IV.1, Figure IV. 2).

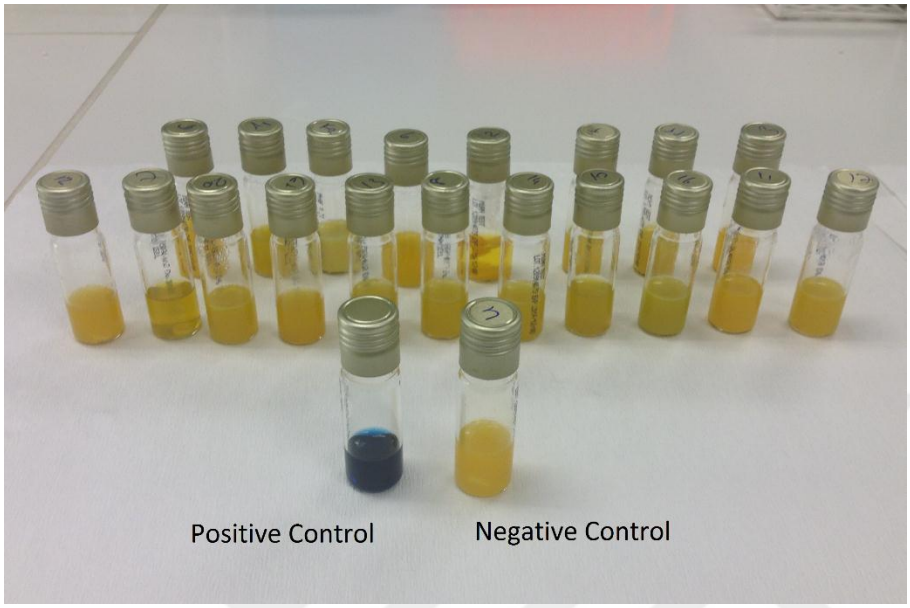


Figure IV.1. Evaluation of Test Results**.**

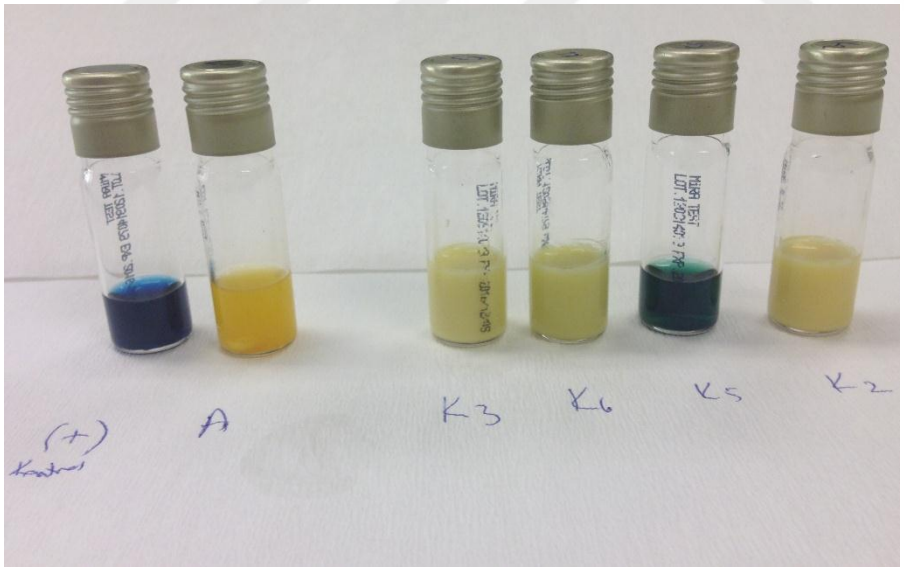


Figure IV.2. Appearence of Positive Sample.

IV.2. Quantitative Antibiotic Residue Test Results (Beta Lactam Elisa Test Results)

Beta lactam elisa test results showed that the same results as the MiRA test results were obtained. That is, only one sample (5th sample) from packaged milk samples and 3 sample samples (1-3 samples) from open milk samples were positive. According to national and international acceptable limits, the maximum amount of beta lactam antibiotics in milk products is 4 ppb (Table IV.1).

Table IV.3: Beta Lactam Elisa Test Results.

Packed Milk Sample no	**Numerical Value**	**Result**	**Unpacked Milk Sample No**	**Numerical Value**	**Result**
1	0,21	Negative	1	7,9	Positive
2	0,21	Negative	2	6,9	Positive
3	0,21	Negative	3	4,8	Positive
4	0,21	Negative	4	0,21	Negative
5	4,2	Positive	5	0,21	Negative
6	0,21	Negative	6	0,21	Negative
7	0,21	Negative	7	0,21	Negative
8	0,21	Negative	8	0,21	Negative
9	0,21	Negative	9	0,21	Negative
10	0,21	Negative	10	0,21	Negative
11	0,21	Negative	11	0,21	Negative
12	0,21	Negative	12	0,21	Negative
13	0,21	Negative	13	0,21	Negative
14	0,21	Negative	14	0,21	Negative
15	0,21	Negative	15	0,21	Negative
16	0,21	Negative	16	0,21	Negative
17	0,21	Negative	17	0,21	Negative
18	0,21	Negative	18	0,21	Negative
19	0,21	Negative	19	0,21	Negative
20	0,21	Negative	20	0,21	Negative

V. DISCUSSION AND CONCLUSION

Milk and dairy products are not only one of the basic foodstuffs, are also among the risky products in terms of containing veterinary drugs, pesticides, mycotoxins, heavy metals, genetically modified organism products (GMO), Dioxin and similar chemical residues (Khanikiet al, 2007). These chemicals that can be found in milk come from industrial or agricultural sources (20).

Most of the chemicals that show the ability to dissolve in oil are easily absorbed in human fat tissues (21). Milk and dairy products are an important source of nutrients for many people and are an indispensable food source for people in every age of life. However Studies indicate that residues of milk and dairy products may adversely affect public health if not produced under the appropriate conditions and prevent the negative exterior factors. FAO and WHO, as well as the official agencies of many countries, are trying to take action in this regard. Authorized organization for the residues of animal products including milk and dairy products in our country is the Ministry of Food, Agriculture and Livestock. Effective inspections within the scope of the residual monitoring program being carried out by the relevant ministry are important for the protection of public health. However, veterinarians, animal breeders, animal food producers, drug producers and distributors have important roles and responsibilities in this regard (1,22).

Milk containing antibiotics cause pathogenic microorganisms to gain resistance. In some people they cause allergic reactions and therefore cause negative consequences (23,24).

Compared with the danger of antibiotic resistance, the risk of toxicity caused by the residues caused by drugs permitted for use in foodstuffs is very low. Nevertheless, the EU has set the Maximum Residue Limits (MRLs) of veterinary drugs in animal feedstuffs to prevent exposure to residues at harmful levels. The regulation of the EU Commission on Active Pharmacological substances of 37/2010 and 22.12.2009 lists the maximum residue limits in foodstuffs of animal origin (3,25).

In the European Union there are numerous regulations on veterinary medicines, as well as changes and updates to these regulations. Basic regulations regulate the use of hormonal, thyrostatic, and β-agonists in animals, creating legal grounds for the prohibition of growth support. In the case of an old directive on medicines that are

allowed to be used, a list was recently updated to list the permitted medicines and their maximum residue limits. All of these directives are adapted to Turkish Laws. In the 2002/30 numbered communiqué of the Turkish Food Codex, ;Maximum Residue Levels in Animal Food Material have been determined. The violation of the residue levels and the legal infrastructure are regulated by Law No. 3285. If there is a prohibited substance in the inspections of the farm, the related matter is confiscated and the legal action is taken about the concerned ones. During periods when legal breakthroughs are not anticipated in live animals sent to the slaughterhouse, the animals are prevented from being slaughtered and the animal is under official supervision during this time. In accordance with Law 5179, Crops or animal products and fish in the presence of prohibited substance or MRL level above the level of the product is withdrawn from the market by hand, money and imprisonment punishment is applied. In addition to these, there are two legal regulations in the EU. The first directive sets out the Union and National Refrence Laboratories to work on veterinary medicines, The second directive sets the number of mandatory screening tests per animal species, each Member State, by regulating drug residue monitoring programs, which are mandatory for member states. Screening tests play an important role in national monitoring programs. In the slaughterhouses of member countries, screening analyzes are carried out on a large number of urine, bile and stool samples, within the framework of national residue monitoring programs. In Turkey, the Veterinary Research Laboratories within the Ministry of Agriculture conduct their National Reference Laboratories in their area of expertise. Also in Turkey since 2000; honey, fish, poultry and eggs, National Residue Monitoring Programs have started to be implemented and the results have been regularly reported to the EU (3,26).,

When we look at the Mira test results in our study, it is seen that there is one positive sample. This sample (sample 5) was positively detected with Elisa. Three samples (1-3 samples) were found positive in the samples sold outdoors. The same samples were also found to be positive in experiments with beta lactam elisa kit. At this point, the test performance of Mira chitin was determined to be appropriate for beta lactam (Figure V.1).

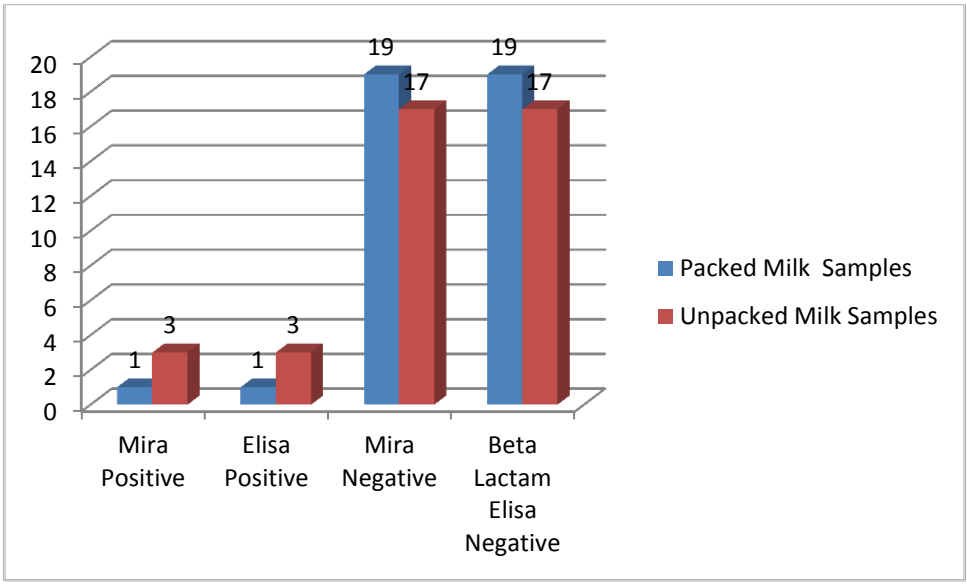


Figure V.1. Comparison Chart of Elisa and Mira Test Results.

In a study of 240 samples in Ankara penicillin G, oxytetracycline, streptomycin, gentamicin and neomycin amount was researched and both our countries encountered high rates of antibiotic residues given the international limits (27). However, as in our study, a small number of positive samples were detected. In a different study conducted in Ankara, penicillin residues were investigated in 200 milk samples and 11 samples were positive (28). In our study, 4 of 40 samples were positively detected.

In a study conducted with raw milk, a total of 170 raw cow milk samples were analyzed with two different methods to identify antibiotic residues. No tetracycline group antibiotics could be identified in any of the samples analyzed with the specific rapid test kit, antimicrobial residues were found at low concentrations in all samples in aliquots of High Performance Liquid Chromatography of the same samples (29). In our study, residual amounts of beta lactam antibiotics were found at very low levels (4.2-7.9, average).

In another study made with 204 raw milk and 103 pasteurized milk, disc diffusion method was used and positivity was detected with 44% and 29% respectively (30).

In other studies conducted in our country, various antibiotic residues in milk samples were investigated and positive results were obtained (31,32,33).

When all the studies are examined, the antibiotic residue problem in milk and dairy products still continues, The beta lactam levels in the lower limits of our study have been evaluated as promising. However, with further examples these studies should be expanded and routinely analyzed for milk and milk products in terms of hygiene

criteria. Because not only antibiotic residues, but also food substances must be examined for various pollutants such as mycotoxins, pesticides, hormones and heavy metals.(34,35)

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VII.CURRICULUM VITAE

Name	Osetya	**Surname**	Batıbay
Place of birth	Ankara	**Date of Birth**	07.10.1990
Nationality	Turkish	**Phone Number**	+905320615696
E-mail	osetyabatibay@gmail.com		

Education

Degree	School/Department	Graduation Year
Master	Yeditepe University/ Nutrition and Dietetic	2017
Associate	Anadolu University/ Management of Health Institutions	2016
Undergraduate	Yeditepe University/ Nutrition and Dietetic	2013
High School	Haydarpaşa Anadolu Lisesi	2008

Work Experience

Position	Place	Year
Foundress/Dietitian	O Diyet Beslenme ve Danışmanlık Merkezi	2016- continue
Dietitian	PinnerTest Sağlıklı Yaşam	2015-2016
Dietitian	Emsey Hospital	2014-2015
Dietitian	Anadolu Sağlık Merkezi	2013-2014

Foreign Language	Level	Score (KPDS/UDS/IELTS/TOEFL)
English	Advanced	-

Computer Programs

Program	Level
Microsoft Office Programs	Advanced
Nutrition Data Systems (Bebis)	Advanced