

# Effects of Boiling Dairy Products on Human Brucellosis

## Süt Ürünlerinin Kaynatılmasının İnsan Brusellozisi Üzerine Etkisi

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### Abstract

**Objective:** In this study, we determined the seroprevalence of Brucella in the blood of bovine and ovine animals and in the blood of the people who raise these animals to produce cheese in two rural counties that use two different methods of cheese production in Erzurum Province in Turkey.

**Materials and Methods:** Samples are taken from 100 bovine animals, 100 ovine animals, 100 young people between the ages of 10-20 years and 100 adults between the ages of 20-60 years. The samples were tested with the Rose Bengal Plate Test (RBPT), the Serum Agglutination Test (SAT), the Coombs' Test and a micro-ELISA.

**Results:** We found the following rates of Brucella in the province that makes cheese with raw milk: bovine (3.00%), ovine (5.00%), people between 10-20 years of age (2.00%) and people between 20-60 years of age (10.00%). However, the corresponding rates in the region that makes cheese with boiling milk were 2%, 4%, 1% and 5%, respectively.

**Conclusion:** The results were analyzed descriptively and in comparison to the results from the other region. There was a significant difference found between the two regions among the Hınıs and Oltu individuals aged 10-20 and 20-60 ( $z=0.6<1.96$  with a 95% confidence interval).

**Key Words:** Boiled milk, dairy products, Human Brucellosis

### Özet

**Amaç:** Bu çalışmada; Türkiye'nin Erzurum ilinde farklı iki yöntemle peynir üreten iki ilçenin kırsalındaki büyük ve küçük baş hayvanlardan, bu hayvanların besisini yaparak peynir üreten insanlardan alınan kan örneklerinde Brucella seroprevelansı araştırıldı.

**Gereç ve Yöntem:** Araştırmaya her bölgeden örnekler; 100 sığır, 100 koyun, 10-20 yaş grubu 100 genç çocuk ve 20-60 yaş grubu 100 erişkin dahil edildi. Örnekler Rose Bengal Plate Test (RBPT), Serum aglütinasyon (SAT), Coombs ve Mikro ELISA yöntemleriyle ayrı ayrı test edildi.

**Bulgular:** Kaynatılmamış süttten peynir yapılan bölgedeki sığırlarda %3.00, koyunlarda %5.00, insanların 10-20 yaş grubunda %2.00 ve 20-60 yaş grubunda ise %10.00 oranında Brucella antikor pozitif bulundu. Buna karşılık kaynatılmış süttten peynir yapan bölgedeki pozitiflik oranları sırasıyla; %2, %4, %1 ve %5 olarak bulunmuştur.

**Sonuç:** Sonuçlar kendi aralarında ve diğer bölgedeki sonuçlarla tartışıldı. 10-20 yaş ve 20-60 yaş arası Hınıs ve Oltu bireyleri karşılaştırması  $z=0.6<1.96$ , iki bölgedeki kişiler arasında %95 güven aralığında fark vardır. İki bölge arasındaki sonuçlarda anlamlı bir farklılık saptandı.

**Anahtar Kelimeler:** Bruselloz, kaynamış süt, süt ürünleri

### Introduction

Brucella is a disease that sometimes produces chronic clinical symptoms and occasionally appears as a progressive zoonotic disease with no symptoms. To clinically diagnose this infection, which produces clinical symptoms similar to many other diseases, is quite difficult [1].

Laboratory diagnosis of Brucellosis that has an ambiguous clinical diagnosis is also complex [2]. The reproduction of Brucella is more difficult to achieve in a laboratory environment compared to other bacteria because it is fastidious. In

addition, because antibodies tend to have cross-reactivity and incomplete antibodies may be produced, many tests should be conducted simultaneously to achieve complete results for a serologic diagnosis [3].

Every day, brand new Brucella species are detected. Furthermore, the transport of animals throughout the modern world means that this zoonosis is not geographically based any longer; it may occur in any region. At the same time, we face a complicated situation in that *Brucella abortus* only affects bovine animals, *Brucella melitensis* affects ovine animals, and Brucella can occur not only in wild dogs but also in domesticated and wild ovine and bovine animals [4].

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**Table 1. The distribution of positive samples according to village (Each group consists of 100 subjects)**

	Hinis Villages		Oltu Villages		TOTAL	
	n	%	n	%	n	%
Cattle	3	3.0	2	2.0	5	2.5
Sheep	5	5.0	4	4.0	9	4.5

**Table 2. The distribution of positive samples from humans according to sex**

	Hinis Villages			Oltu Villages			TOTAL		
	n	n total	%	n	n total	%	n	n total	%
Women (10-20 years)	1	42	2.38	0	46	0.0	1	88	1.14
Men (10-20 years)	1	58	1.72	1	54	1.85	2	112	1.78
Women (20-60 years)	7	60	11.67	4	45	9.88	11	105	10.47
Men (20-60 years)	3	40	7.50	1	55	1.82	4	95	4.21
n: number of patients; n total: total number of people									

Thus, even though the number of Brucellosis cases in Turkey does not increase every day, there has not yet been a decrease either. In the east and southeast regions of Turkey, the prevalence of Brucellosis is higher than it is in other regions of Turkey. In this study, we aimed to determine the prevalence of Brucellosis in both animal breeders and the animals they breed in the two rural counties of Erzurum.

## Materials and Methods

In our study, blood samples were taken from randomly selected animals and from people who breed the animals for milk for cheesemaking purposes in two counties of Erzurum. The difference between the two counties is that in one of them (Oltu), the milk is boiled while making cheese, and in the other one (Hinis), the milk is not boiled. In our study, blood samples were taken from 100 sheep, cattle, young people (10-20 years) and adults (20-60 years) from both regions, and all the samples were tested. The presence of Brucella in the serum taken from animals and people was investigated using four different methods: the Rose Bengal Plate test (RBPT), the Serum Agglutination Test (SAT) and the Wright Method, Coombs' Antiglobulin test and a micro-ELISA [2].

## Results

Each animal group (cattle and sheep) consisted of 100 subjects. Seropositivity was present in 2.5% of the cattle and

4.5% of the sheep. The distribution of seropositivity by village is presented in Table 1.

Comparison of 10-20 age range individuals in Hinis and Oltu; there is 95% difference of reliability interval between the people of two regions. ( $z=0.6<1.96$ ). Among the 10- to 20-year-old participants in Hinis and Oltu, there was a statistically significant difference in the prevalence of Brucella between the two regions. ( $z=1.35<1.96$ ) (Table 2).

## Discussion

Brucellosis is considered to be solely an infection occurring in animals. However, consuming contaminated meat, milk or dairy products; having direct contact with infected animal blood or any other tissues through wounded skin or conjunctiva; and inhaling infectious aerosols play a large role in transmitting this disease to people. The most important transmission pathway is through the digestive tract in countries such as ours, where people consume cheese made from raw milk and the usage of pasteurized milk is not common [5, 6].

Eastern Turkey has the highest prevalence of Brucellosis in the country. The spread of Brucellosis infection is closely related to socio-economic factors and some cultural customs. Thus, its distribution differs throughout regions and communities in the world [7]. In the two rural counties (Hinis and Oltu) in Erzurum, there are obvious differences in animal breeding and the production of animal products. While milk is boiled during the production of cheese in the county of Oltu, cheese

**Table 3. The distribution of positive samples according to age (Each group consists of 100 subjects)**

	Hinis Villages		Oltu Villages		TOTAL	
	n	%	n	%	n	%
Individuals aged 10-20	2	2.0	1	1.0	3	1.5
Individuals aged 20-60	10	10.0	5	5.0	15	7.5

is produced from raw milk in the county of Hınıs in Erzurum. However, hand milking, a primitive method, is still practiced in both counties. Mechanization is not yet commonly used in these two counties. The prevalence of *Brucella* in sheep and cattle is similar between the two counties (5% of sheep and 3% of cattle in Hınıs; 4% of sheep and 2% of cattle in Oltu) because the government's animal health regulations are the same in both counties. Regarding the rates among humans, the prevalence among young people between 10-20 is 2% in Hınıs and 1% in Oltu. Among adults in the 20-60 age range, the prevalence is 10% in Hınıs and 5% in Oltu (Table 3). While the difference between the groups of young people is not particularly large, the difference between the adult groups is considerable. Milking is performed by women over 20 in both regions. Young children do not milk, regardless of their sex. Adults, especially adult women, milk sheep and cattle. Therefore, there are no significant differences in the diagnosis rates of *Brucella* between young male and female children in Oltu and Hınıs. As shown in Tables 2 and 3, *Brucella* diagnosis rates in both regions increase as age increases. Again, there are significant differences in the prevalence rates among adults in both regions. In the region where cheese is produced from raw milk, the prevalence is 2.38% among females and 1.72% among males in the 10-20 age range, while in the region where people produce cheese from boiled milk, the rates are 0.00% among females and 1.85% among males (Among individuals 10-20 years of age in Hınıs and Oltu, there is a significant difference in the prevalence rates between the two regions).

Regarding the adults, the seropositivity rate among females is 11.67%, a very high rate, in the community that produces cheese from raw milk, while the seropositivity rate among males is 7.50% in the same community. In contrast, in the society that produces cheese from boiled milk, the seroprevalence of *Brucellosis* was 9.88% among females and 1.82% among males (Among individuals 20-60 years of age in Hınıs and Oltu, there is a significant difference in the prevalence rates between the two regions.) (Comparison of 20-60 age range individuals in Hınıs and Oltu  $z=1.35<1.96$ , there is 95% difference of reliability interval between people of two regions.).

We can connect the fact that *Brucella* antibodies are found at higher rates among 20- to 60-year-old females in the two regions to the fact that young adult females actively

participate in the different stages of milking the animals and producing dairy products (These products are consumed by both males and females). The lower seropositivity rates among males compared to females can be explained by the fact that males do not take part in milking animals and producing dairy products, although they do consume these products (There is a significant difference between the females aged 10-20 and the females aged 20-60 in Hınıs).

*Brucellosis* is a systemic infection that affects many organs and tissues [8]. According to the World Health Organization (WHO), *Brucellosis* is the most common infection in the world, and 500,000 cases are reported annually [4, 9]. According to data from the Ministry of Health, there was a gradual increase in reported cases in Turkey between 1970 (0.1/100,000) and 2004 (18,563 cases; 25.6/100,000) [10, 11]. *Brucellosis* is an endemic disease in our country that is reported in low numbers (10,224 cases in 2009-2010; 13.5/100,000 incidence) because animal therapy and vaccination are performed precisely and intensively in our developing country. However, the incidence rates in the 1970s were very low (e.g., 0.1/100,000) because scientific studies were rare and the case notification system was not developed at the time.

We can draw three conclusions from the data we collected. First, in the regions where cheese and other dairy products are produced from raw milk and people consume raw milk, the rate of *Brucellosis* infection is high compared to the rates in regions that use boiled milk. Second, the *Brucellosis* infection rate is higher for females who participate in milking in both regions compared to males. Finally, the animal health measures applied to animals in both regions are equal.

Based on this study, we conclude that *Brucellosis* studies should be conducted on animal breeders and animals routinely in developing countries such as Turkey. In addition, enabling mechanized milk production will encourage all regions to produce dairy products from boiled milk.

**Conflict of interest statement:** The authors declare that they have no conflict of interest to the publication of this article.

## References

1. Rubinstein E, Baldwin C. Management and pathogenesis of *brucellosis*. In: Intracellular bacterial infections, JC Pechere (Ed), Cambridge Medical Publications, 1996; pp.87-98.

2. Notice of Infectious Diseases and Notification System Standard Diagnosis Surveillance and Laboratory Guide, Ministry of Health Basic Health Services Head Office, 4th Edn September 2005; Ankara.
3. Gall D, Colling A, Marino O, et al. Enzyme immunoassay for serological diagnosis of bovine brucellosis: A trial in Latin America. Clin Diagn Lab Immunol 1998; 5: 654-61.
4. Food and Agriculture Organization of the United Nations World Organization of Animal Health Organization Brucellosis in Human and Animals. World Health Organization, Geneva WHO/CDC/EPR 2006.
5. Edward J. Young Brucella species, Mandell GL, Bennet JE, Dolin R. Principles and Practice of Infectious Diseases, 5th Edn, New-York, 2000; 2386-93.
6. Gotuzzo E. Brucella, Gorbach SL, Bartlett JG, Blacklow NR. Infectious Diseases second edition, W.B. Saunders Company, 1998. Philadelphia; p.1837-44.
7. Gwida M, Aldohouk S, Melzer F, et al. Brucellosis regionally emerging zoonotic disease? Croat Medical Microbiology 2010; 51: 289-95. [\[CrossRef\]](#)
8. Pappas G. Changing Brucella Ecology: Novel reservoirs, new threats. Int J Antimicrob Agents 2010; 36(suppl 1): 8-11. [\[CrossRef\]](#)
9. Pappas G, Papadimitrov P, Akriditis N. The New Global Map of Human Brucellosis Lancet Infect Dis 2006; 6: 91-9.
10. Turkish Ministry of Health, Basic Health Services Head Office, Head of Department of Zoonotic Diseases Brucellosis Data.
11. Yüce A, Alp Cavus S. Brucellosis in Turkey; General Review Clinic Journal 2006; 19: 87-97.