

Contents

Subject	Page
Introduction.	1
Review of Literature.	5
❖ Historical Review.	5
❖ Milk Ring Test (MRT) and Serial Dilution MRT.	6
❖ Whey Serological tests.	25
❖ Public Health Significance of Brucella.	31
Material and Methods.	48
Results.	61
Discussion.	90
Conclusion and Recommendation.	105
Summary.	109
References.	112
Arabic Summary.	

Conclusion and Recommendations

No one can deny that brucellosis is a great problem in most countries of the world, so great efforts must be done to eradicate this disease. Serological examination of the animals is the corner stone in the diagnosis and eradication of the disease. Under the condition of this investigation and according to the data obtained, it is concluded that:

By examination of cattle milk and milk whey samples it is evident that, MRT and serial dilution MRT were more sensitive than wRBPT, wBAPAT, wRiv.T and wTAT. While, within the whey serological tests it is clear that wTAT was more sensitive than wRBPT, wBAPAT and wRiv.T

In sheep milk and milk whey samples it is clear that, MRT and serial dilution MRT were the most sensitive tests as they gave higher positive results than whey serological tests. Also, within whey serological tests it is obvious that, wRiv.T gave the highest positive results than wRBPT, wBAPAT and wTAT.

Concerning goat's milk and milk whey samples it is noted that, MRT and serial dilution MRT showed the highest positive results than the whey serological tests. While, within the whey serological tests it is evident that, wRBPT and wBAPAT were more sensitive than wRiv.T and wTAT. In addition, wRiv.T was more sensitive than wTAT.

With regard to buffalo's milk and milk whey samples, MRT, serial dilution MRT, wRBPT, wBAPAT, wRiv.T and wTAT showed negative results to all of examined milk as well as milk whey samples which reflect that, these tests were not reliable for buffalo's examination or may be due to the resistance of buffaloes to brucellosis.

Incidences of brucellosis in cattle, sheep, goat's and buffaloes's by using milk ring test (MRT) were 12.38, 10, 15 and 0%, respectively.

Milk ring test (MRT) and serial dilution MRT are highly sensitive, rapid screening and easy tests for diagnosis of brucellosis.

Brucellosis continuous to be a common health problem in communities where the consumption of unpasteurized milk and milk products is common.

The high economic loss and public health implications of brucellosis necessitates the need for effective surveillance as well as appropriate preventive and control measures among human and animal populations.

The control of milk-borne brucellosis depends on its eradication from dairy animals and an adequate heat treatment of milk and milk products. Therefore, the control includes:

I-Control in the farm.

- All animals eligible for testing (females of one year old and above, bulls, rams and bucks) are permanently identified by an ear tag.
- Animals are tested either by MRT or Rose Bengal plate test, which are recommended as screening tests, with positive samples being, retested by complement fixation test, which is recommended as confirmed test.
- Reactors should be removed from the herd as soon as possible and slaughtered.

- Adequate compensation should be paid to farmers for slaughtered animals.
- The herd retested after 30-60 days, where no reactors are found, the herd is retested after 6 months and if no reactors are found, this may be accepted as evidence that the herd is brucellosis-free. In certain circumstances, another retest after a further period of 6 months may be considered advisable before a herd can be certified as free from brucellosis.
- The sales of female over one year old from infected herds, should be prohibited unless they are to be sent to direct for slaughter.
- Regulations to prevent the introduction of infection to brucellosis-free premises are also required.
- Vaccination of animals is the only way to reduce the rate of brucellosis infection in high-prevalence areas and it may be accepted to apply calfhood (between 3-6 months) vaccination with *Brucella abortus* strain 19 vaccine, while lambs and kids (between 3-8 months) are vaccinated with *Brucella melitensis* strain Rev. 1 vaccine. The vaccinated animals must be kept in isolation till developing immune response. In this way vaccinated young animals will slowly replace the non-vaccinated members.

II-Control of dairy products:

Because the costs of animals eradication programmes for brucellosis in a country like Egypt may be beyond the financial resources, prevention and control should be concentrated on food hygiene environmental protection and public health education as follows:

- Efficient pasteurization or thorough boiling of milk must be applied.

- All dairy products must be manufactured from heat treated milk and prevention of recontamination.
- The sales of certain dairy products made from raw milk is prohibited until after a reasonable storage time to allow any pathogenic bacteria to die.
- Domiati cheese manufactured from raw milk should not be consumed before at least 15 days after preservation at room temperature or 25 days if kept in refrigerator (4°C).
- Restriction of the consumption of sweet cream.
- Strict hygienic measures should be imposed during milk production, handling and processing to prevent such contamination.
- Periodical inspection of dairy farms as well as dairy plants by specialists.
- Educational programs to those sharing in milk production and handling as well as in processing of dairy products should be encouraged.
- Periodical examination of persons who are in contact with milk and milk products (health certificate).

Summary

A total of 500 milk samples were collected from different animals at different localities in Assiut Governorate. These samples represented by 210, 120, 120 and 50 of raw milk as well as milk whey samples obtained from cattle, sheep, goats and buffaloes, respectively.

The incidence of brucella antibodies in milk samples were estimated by MRT and serial dilution MRT and by wRBPT, wBAPAT, wRiv.T and wTAT in their corresponding whey samples.

I-Cattle milk and milk whey samples.

Out of 210 milk samples examined by MRT, 12.38% were positive (constituting 4.76, 2.38 and 5.24% were positive in grade (++) , (+++) and (++++), respectively), 7.62% were doubtful and 80% were negative.

The serial dilution MRT gave 4.76, 0.95, 1.43, 0.48, 1.43, 0.95, 0.48, 0.95 and 0.95% positive reactors in titres of 1:1, 1:2, 1:8, 1:16, 1:32, 1:64, 1:128, 1:512 and 1:1024, respectively, with total reactors 12.38% while, 87.62% non-reactors.

In the corresponding milk whey samples by whey serological tests: wRBPT, wBAPAT, wRiv.T and wTAT gave 4.29, 4.29, 4.29 and 5.24% positive, while, the negative results were 95.71, 95.71, 95.71 and 94.76%, respectively.

The MRT was taken as a reference standard for evaluating other tests. The sensitivity were (34.62%), (34.62%), (34.62%) and (42.31%), while the specificity were 100% for each and the agreement were (84.29%), (84.29%), (84.29%) and (85.24%) for wRBPT, wBAPAT, wRiv.T and wTAT, respectively.

II-Sheep milk and milk whey samples.

Out of 120 milk samples examined by MRT, 2.5 and 7.5% gave positive ring and ring & disc, respectively, with 10% total positive and 90% negative.

The serial dilution milk ring test showed 0.83, 2.5, 2.5, 0.83, 1.67, 0.83 and 0.83%, positive reactors in titres of 1:1, 1:2, 1:4, 1:8, 1:16, 1:32 and 1:128, respectively, with 10% total reactors and 90% non-reactors.

In case of milk whey samples, wRBPT, wBAPAT, wRiv.T and wTAT gave 1.67, 1.67, 3.33 and 1.67% positive results and 98.33, 98.33, 96.67 and 98.33% negative results, respectively.

The MRT was chosen as a reference standard test for calculation of sensitivity, specificity and agreement of other tests, which were, 16.67, 16.67, 33.33 and 16.67%; 100% for each and 91.67, 91.67, 93.33 and 91.67% for wRBPT, wBAPAT, wRiv.T and wTAT, respectively.

III-Goat's milk and milk whey samples.

Out of 120 milk samples examined by MRT, it is evident that 2.5, 10.83 and 1.67% were positive showing ring, ring & disc and disc, respectively, with total positive of 15% and 85% negative.

The serial dilution MRT showed 2.5, 9.17, 0.83, 1.67 and 0.83% positive reactors in titres of 1:1, 1:2, 1:4, 1:16 and 1:64, respectively. The total reactors and non-reactors were 15 and 85%, respectively.

Moreover, whey serological tests wRBPT, wBAPAT, wRiv.T and wTAT gave 3.33, 3.33, 2.5 and 1.67% positive results, while, the negative results were 96.67, 96.67, 97.5 and 98.33%, respectively.

The sensitivities were (22.22%), (22.22%), (16.67%) and (11.11%), while the specificities were 100% for each and the agreements were (88.33%), (88.33%), (87.5%) and (86.67%) for the whey serological tests, respectively.

IV-*Buffaloe's* milk and milk whey samples.

All of the examined *buffaloe's* milk as well as milk whey samples found to be negative to MRT and serial dilution MRT as well as to all whey serological tests.