ZOONOTIC HEALTH HAZARDS CONCERNING BRUCELLOSIS AND LEPTOSPIROSIS IN EGYPTIAN SWINE AND HUMAN IN CONTACT

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ABSTRACT

Brucellosis and Leptospirosis are the most symbolistic swine occupational bacterial zoonosis constitutes direct occupational hazards for manipulating employees or animals that licked or eaten the aborted fluid or membranes. Occupational workers appear to be the most risk groups of contracting the diseases from pigs. It's of ecological impact and on target zoonosis to detect the sero-prevalence of those two zoonosis in human in contact with free living swine, reflect the human bio-hazards Received at: 26/6/2012 are due to direct contact with contaminant swine subset. Pigs are naturally exposed to zoonotic agents in their subset with subsequent direct occupational human hazards. Brucella and Leptospira are infectious agents probably reflects a major bacterial hazards for human in contact with the free living swine in addition the Accepted: 2/8/2012 economical drawbacks consequences to swine abortion and piglet depletion. A total of 250 free rearing pigs blood samples were collected from different localities in large Cairo governorate, Egypt. Also, 150 serum samples were collected from occupational workers. Human and swine sera were tested for antibodies against the two zoonotic diseases by using Egyptian locally serological antigens and commercial kits. Antibodies against Brucella were detected in 42% of 250 pig samples using RBPT. The most sensitive confirmatory test was SAT 37.6% The seropositive percent of human in contact using SAT 4.7%. Antibodies against three Leptospira serovars were detected in 66.8% of the serum samples from pigs using the Microscopic Agglutination Test (MAT) at titers of 1≥200. The highest seroprevalence was recorded for L. Pomona serovar 32.4% followed by L. grippotyphosa 20.8% and then the lowest prevalence detected by Licterohaemorrhagiae 13.6%. While in human 25.3% were positive with the highest incidence corresponding to L. Pomona 11.3%.

Key words: Zoonotic Health hazard, swine, Brucellosis, Leptospirosis.

INTRODUCTION

More than 30 diseases are considered common to man and pig. Most of them, such as salmonellosis, trichinellosis, toxoplasmosis, Q fever, leptospirosis etc. may involve other animal species as well, while others are specifically porcine-related, such as *Taenia solium* (the cestode causing human cysticercosis), Streptococcus suis and Brucella suis (Shimshony, 2009). Brucellosis is one of the most important bacterial zoonosis worldwide (Young, 1995). The alarm toward Brucellosis that is highly contagious disease have Egyptian authorities priority, it's an endemic disease among Egyptian humans and animals (WHO, 1998). Laboratory testing is an absolute prerequisite for a proper diagnosis of human brucellosis and for detection and confirmation of brucellosis in animals. Laboratory diagnosis of brucellosis in animals or man may be achieved either through blood culture or serological testing (Smits and Culter, 2004).

Poor reproducibility has been demonstrated with a frequently used serological screening test, the Rose Bengal test (RB), when performed at different study sites (Maichomo *et al.*, 1998). Specificity issues have also plagued the (RB) test. Consequently, positives should be confirmed in a more specific test such as the serum agglutination test, complement fixation test, or the *enzyme* linked immunosorbent assay (Omer *et al.*, 2002; Al Dahouk *et al.*, 2003).

Leptospirosis has been recognized as an emerging global public health problem because of its increasing incidence in both developing and developed countries (Slack *et al.*, 2008).

Leptospirosis is considered as one of the most widespread zoonoses worldwide (Levett, 2001). The disease is caused by spirochetes of the genus *Leptospira*, Most of leptospira serovars can infect different animal species, but there genus is a primary host reservoir for each serovar, which ensures the survival and dissemination of the organisms (Birnbaum *et al.*, 1998).

The standard method for serological diagnosis of leptospirosis is the microscopic agglutination test (MAT), usually with a cut-off value at 100 (Faine *et al.*, 1999). To the cut-off value at 100, the MAT sensitivity is belived to be only moderate (Mousing *et al.*, 1995) and infected animals may have titers below the widely accepted minimum significant titer of 100, but the specificity is good (Ellis, 1999).

The economic loses and zoonotic potential of leptospiral infections in pigs are recognized throughout the world. The most commonly incriminated serovars are Pomona and tarassoni (Hathaway and little, 1981). Pigs of all ages are susceptible to Leptospirosis. Death or retarded development may occur in young pigs. Abortion and stillbirth are also consequences of infections (Faine, 1994).

For the pervious reasons this study done for throw the focus on Bacterial zoonotic diseases infect human in contacts with swine as Leptospirosis & Brucellosis in cairo, Egypt.

MATERIALS and METHODS

A total of 250 blood samples were collected from free rearing pigs from different herds scattered at Cairo, Giza & Qalyobia Provinces. Also, 150 blood samples were collected from human in contact.

Samples were centrifuged within 24 hours from collections and sera were collected. Serum of each sample was numbered, divided into 2 obliquities and stored at -20°C until tested. Sera were tested at the Department of Zoonotic Diseases National Research Centre, Dokki, Giza, Egypt and Leptospirosis Unite belonging to Department of Mastitis & Neonates Diseases at Animal Reproduction Research Institute, Haram, Giza, Egypt.

Human and swine sera were tested for antibodies against the two zoonotic diseases. the test methods and antigen types used were given in the following table

| Diseases | Test method | Antigen types | References Rose & Roepke (1957) | |
|---------------|--|-----------------------------|---------------------------------------|--|
| Brucellosis | - Rose Bengal Plate Test (screening test) | Whole antigen of B. abortus | | |
| | -Serum Agglutination Test (confirmatory test) | S99 | Alton et al (1988) | |
| ····· | | Viable serovars | COLE et al. | |
| Leptospirosis | - Microscopic Agglutination Test | 1-L.grippotyphosa | (1973) | |
| | | 2-L.icterohaemorrhagiae | | |
| | | 3-L.pomona | | |

 Table 1: Test method and Antigen types

A- Serology of Brucellosis in pigs &human sera:

- Rose Bengal plate Test (RBPT):

The Rose Bengal stained Brucella antigen is used for the early detection of Brucella agglutinins (Brucella abortus, melitensis and suis) according to Rose and Roepke (1957).

- Serum Agglutination Test (SAT):

The most widely used test and was performed as described by Alton et al. (1988)

B- Serology of Leptospirosis in pigs &human sera:

- Microscopic Agglutination Test (MAT):

Improved microtechnique for detection of leptospira antibodies, which Performed as described by Cole et al. (1973).

RESULTS

| | No of tested Pig sera | NO of seropositive tested samples | Seropositive % | No of tested Human sera | NO of seropositive tested samples | Seropositive% |
|--------------------|-----------------------------|---|-------------------|----------------------------|--|---------------|
| Brucella spps | | 105 (RBPT) | 42% | | 9(RBPT) | 6 % |
| | 250 | 94 (SAT) | 37.6% | 150 | 7 (SAT) | 4.7% |
| Leptospira spps | | 167 (MAT) | 66.8% | | 38 (MAT) | 25.3% |

Table 2: Seroprevalence of Brucellosis and Leptospirosis in Human and pig sera:

RBPT= Rose Bengal Plate Test

SAT = serum agglutination test

MAT = Microscopic Agglutination Test.

Table 3: Detection of leptospira serovars antibodies in pigs sera using Microscopic Agglutination Test:

| serovars | 250 tested pigs | | 1≥200 MAT Titers (total immunoglobulin) | | | | |
|-----------------------|-----------------|------|---|-------|-------|--------|--------|
| | Seropositive | % | 1/200 | 1/400 | 1/800 | 1/1600 | 1/3200 |
| L.pomona | 81 | 32.4 | 9 | 16 | 21 | 27 | 8 |
| L.icterohaemorrahgiae | 34 | 13.6 | 4 | 7 | 9 | 11 | 3 |
| L.grippotyphosa | 52 | 20.8 | 8 | 12 | 16 | 9 | 7 |
| Total | 167 | 66.8 | | | | | |

Table (3) summarizes the results obtained by Microscopic Agglutination Test, it was noticed that leptospiral agglutinins against three leptospiral serovars were detected in 167 out of 250 pig sera (66.8%); meanwhile, L. *pomona* antibodies was the most predominant in pig sera with (32.4%) of the total number of the examined sera followed by L.grippotyphosa (20.8%) and then L.icterohaemorrhagiae (13.6%).

Table 4: Detection of leptospira antibodies in human sera using Microscopic Agglutination Test:

| serovars | 150 tested persons | | 1≥200 MAT Titers (total immunoglobulin) | | | | |
|-----------------------|--------------------|------|---|-------|-------|--------|--------|
| | Seropositive | % | 1/200 | 1/400 | 1/800 | 1/1600 | 1/3200 |
| L.pomona | 17 | 11.3 | 2 | 4 | 4 | 6 | 1 |
| L.icterohaemorrahgiae | 10 | 6.7 | 4 | 2 | 1 | 3 | 0 |
| L.grippotyphosa | 11 | 7.3 | 3 | 4 | 2 | 2 | 0 |
| Total | 38 | 25.3 | | | | | |

Table (4) summarizes the results obtained by Microscopic Agglutination Test, it was noticed that leptospiral agglutinins against three leptospiral serovars were detected in 38 out of 150 human sera (25.3%); meanwhile, *L.Pomona* antibodies was the most predominant in human sera with (11.3%) of the total number of the examined sera followed by *L.grippotyphosa* (7.3%) then *L.icterohaemorrahgiae* (6.7%).

Table 5: Detection of Brucella antibodies in pig sera using Rose Bengal Plate Test:

| Serovars | 250 tested pi | gs |
|--------------|---------------|----|
| | Seropositive | % |
| Brucella SPP | 105 | 42 |

Table 6: Detection of Brucella antibodies in pig sera using Serum Agglutination Test:

| Serovars | 250 test | ed pigs | | |
|--------------|--------------|---------|-------|-------|
| <u></u> | Seropositive | % | 1/160 | 1/320 |
| Brucella SPP | 94 | 37.6 | 50 | 44 |

Table 7: Detection of Brucella antibodies in human sera using Rose Bengal Plate Test:

| Serovars | 150 teste | d persons |
|--------------|--------------|-----------|
| | Seropositive | % |
| Brucella SPP | 9 | 6 |

Table 8: Detection of Brucella antibodies in human sera using Serum Agglutination Test:

| Serovars | 150 tested | human sera | · | | |
|--------------|--------------|------------|-------|-------|--|
| | Seropositive | % | 1/160 | 1/320 | |
| Brucella SPP | 7 | 4.7 | 5 | 2 | |

Antibodies against Brucella were detected in 105 (42 %) of 250 pig samples using RBPT. The most sensitive confirmatory test was SAT 94 (37.6 %). The seropositive percent of human in contact 7(4.7 %).

DISCUSSION

Pigs included in this study appeared to be in good health when blood samples were taken; perhaps confirming the belief that, in pigs, equilibrium exists between the diseases agents and the host, but serologically there is evidence of zoonotic disease agent antibody activity. Serologic procedures to detect antibodies against Brucella in infected pigs are the most practical and most common means of diagnosis. Apparent healthy seropositive swine may be chronic shedders for Brucella and Leptospira via urine and other body fluids. Leptospirosis has been known as a zoonotic disease, caused by pathogenic spirochetes of the genus Leptospira. Zakład Chorób Swiń (2011).

In the present study a combination of Rose Bengal Plate Test and Serum Agglutination Test appeared to be advantageous in detecting individual reacting pigs, Brucella antibodies were found in 37.6% of the pigs tested, probably reflecting high prevalence of infection. This finding is in full agreement with results obtained by Zaki (1948) on Brucella infection in pigs in large breeding establishment in upper Egypt.

The discrepancy between the high reactors detected by RBPT in Tables (5&7) than the results obtained by SAT in Tables (6&8) may be due to the fact that it is a highly sensitive test, which can detect low titer as in case of chronic brucellosis that can not be considered positive by the quantitative tests (Nicolett, 1982). On the other hand, Serum Agglutination Test is 10 times more sensitive to IgM than IgG1 and IgG2 antibodies (Alton *et al.*, 1988).

In this study, 37.6 % and 4.7 % of the examined swine and contact persons sera of Serum Agglutination Test had Brucella antibodies respectively, the compatibility between swine and human percentages are due to the fact that Brucella is of highly contagious characters. Where humans in contact mainly contracted infection from shedder swine, and usually predisposes farmers, shepherds, butchers, laboratory workers, veterinarians

and slaughterhouse workers. Also, indicate that swine behavior in clay water pools may play vital role for swine and human communication via droplet infection and through intact or abraded skin. Of 80 veterinarians, 92.5% had serological evidence of past or present Brucella infection (Pappas et al., 2006). Also, lower results of brucella antibodies in human were recorded by Omer et al. (2002) who found that the prevalence of brucellosis among high risk occupational groups using Rose Bengal test is among occupational personnel (4.5%). Mudaliar et al. (2003) recorded prevalence of brucellosis of 5.33% in animal handlers and advised that the clinician should keep in mind the possibility of an occupational or environmental exposure in cases of fever of unknown origin.

Human brucellosis seroprevalence of 3.8% has been reported from Chad (Schelling *et al.*, 2003), 13.3% in Uganda (Mutanda, 1998) and in eastern Nigeria a 5.2% were seropositive (Baba *et al.*, 2001).

Leptospirosis, a worldwide distributed zoononis caused by bacteria of the genus Leptospira (antigenically classified into serovars), may be directly or indirectly transmitted through infected urine or environment. Several domestic and wild animals are leptospirosis reservoirs. The disease presents occupational character since it is widely reported in professionals that work in humid environments. In developing countries, outbreaks are related to lack of sanitation, overcrowding in inadequate housing and climatic conditions.Leptospirosis causes numerous public health problems and educational activities are very important to its control (Ullmann and Langoni, 2011).

Leptospirosis affects professionals that are constantly in contact with animals and their residues, especially reservoirs (Tiwari, 2008).

Concerning leptospirosis, in this study, the percent of infected swine and contact humans are compatible 66.8 % and 25.3 %, respectively. The higher incidence in workers may be clarified due to the fast changeability in swine herd individuals, in contrast to long term stability with the same occupational employees.

The report of Burriel *et al.* (2003) in Greek swine herds indicated a prevalence rate for leptospira interrogans serovars (28.2%) a figure which is lower than that given in the present study. Among the serovars reported in other countries, serovar Pomona is the most important in pigs (Faine *et al.*, 1999; De lang *et al.*, 1987; Van Til and Dohoo, 1991), agreeing with the present work. It has been suggested that swine are important maintenance host for serovar Pomona (Faine *et al.*, 1999).

In the present study leptospirosis seroprevalence in human samples were 25.3% the result was in agreement with the study found in Israeli Troops near Jordan River (Hadad *et al.*, 2006). On the other hand the seroprevalence was 9.5% in slaughter workers in New Zealand (Benschop et al., 2009).

In the present study, the rectant serovars and respective frequencies were L. icterohaemorrhagiae (13.6%) and L. grippotyphosa (20.8%) a figure which is higher than reported by DE. AZEVEDO et al. (2008), who found that seropositivity for L. icterohaemorrhagiae (9.5%) and L. grippotyphosa (4.8%).

Infected sows may be aborted consequence to infection by Brucella and Leptospira while their foeti, fetal fluid, and membranes are harboring the causative pathogen. So, the aborted swine constitute direct occupational health hazard for manipulating employees.

Apparent healthy seropositive swine may be chronic shedders for Brucella and Leptospira via urine and other body fluids.

Venereal transmission from carrier boars and sows may play a role in maintenance of the disease (Bharti *et al.*, 2003).

It can be concluded that serological assays concerning brucellosis and leptospirosis verifies direct occupational exposure for high risk group manipulating carrier swine or their pollutant conditions. Also, this study reflect the need of public health worry by Egyptian veterinary authorities' toward swine on such condition that constitutes spots for spread out epidemics.

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المخلط الصحية لأهم الأمراض المشتركة كالبروسيلوزس والليبتوسبيروزس في الخنازير المصرية والمخالطين لها

أشرف يركات ، المبيد رضوان ، سيلفيا احمد

تربى الخذازير فى مصر دون تفتيش صحى بيطرى فلخذازير بطبيعتها تتعرض لكثير من المعسبات الممرضة نتيجة رعيها على المخلفات الادمية. هذاك العديد من الميكروبات التى تؤدى إلى حدوث الأمراض المشتركة مثل البرويسلوزس والليبتوسبيروزس واللتان تعدان من أهم الامراض المشتركة التى تنتقل من الخذازير للعاملين المخالطين لها . كما تعتبر من أهم الامراض التى تسبب الاجهاض فى اذك الخذازير مصببة خصائر اقتصادية فادحة تهدف الدراسة عن كشف انتشار تلك الأمراض المشتركة الخطيرة فى مصل دم الخذازير الحية التى تنتفى على المخلفات الادمية والإشخاص المخاطين لها. تم جمع ٢٠٠ عينة دم من الخذازير خلال عينات دم من اماكن متفرقة من القاهرة الكبري كذلك تم جمع عينات دم من والإشخاص المخاطين لها. تم جمع ٢٠٠ عينة دم من الخذازير خلال عينات دم من اماكن متفرقة من القاهرة الكبري كذلك تم جمع عينات دم من الليبتوسبيرا . كانت النتائج انجابية للاجسام المعندة. حيث تم عمل اختبار سيرولوجية فى مصل الدم الخنازير والمخاطين لها ضد البروسيلا و الليبتوسبيرا . كانت النتائج الجابية للاجسام المعنداة حيث تم عمل اختبار سيرولوجية فى مصل الدم الخنازير مين الى مصر الإنسان (1 ٪). بينما اظهرت النتائج (٦٦.٪) فى عينات الخنازير باستخدام اختبار التلازن الدموى المعدل عند التير عنه عن من العنازير من المعارين التنائي الم المنان الائمين الليبتوسبيرا . كانت النتائج الجابية للاجسام المعنداة ضد البروسيلا باستخدام اختبار روز بنجال (٢٤.٪) فى أمصال الخنازير بينا فى مصل الا وسجلت اعلى حدوث وبائي لليبتوسبيرا بومونا (٢٠٢.٪)، يليه جريبو تيفوزا (٢٠٠٪) ثم انتشار أدنى اكثيروهيمراجى (٢٠٠٪). بينا فى وسجلت اعلى حدوث وبائي لليبتوسبيرا بومونا (٢٠٢.٪)، يليه جريبو تيفوزا (٢٠٠٪) ثم انتشار أدنى اكثيروهيمراجى (٢٠٠٪). بينما فى وسجلت العلى حدوث وبائي لليبتوسبيرا بومونا (٢٠٠٪)، يليه جريبو تيفوزا (٢٠٠٪) ثمان المن (٢٠٠٪). يليه جريبو تيفوزا (٢٠٠٪) ثم انتشار أدنى اكثيروهيمراجى (٢٠٠٪). الإتسان فكانت النتائج كالتالى (٢٠٠٪) إيجابية وأعلى نسبة كانت مديروفار بومونا (٢٠٠٪). يليها جريبو تيفوزا (٢٠٠٪) ثم اختشار

الغلاصة

يعتبر مرض البروسيلوزس والليبتوسبيروزس من أهم الامراض المشتركة وكانت نسب الإصبابة في الخنازير. التي نتغذى عل المخلفات الادمية. ايجابية وتمثل مصدر لانتشار الامراض للعمال المخالطين من خلال الحيوانات الحاملة للامراض.