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## Serosurvey for Zoonotic Viral and Bacterial Pathogens Among Slaughtered Livestock in Egypt

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### Abstract

**Introduction**—Zoonotic diseases are an important cause of human morbidity and mortality. Animal populations at locations with high risk of transmission of zoonotic pathogens offer an opportunity to study viral and bacterial pathogens of veterinary and public health concern.

**Methods**—Blood samples were collected from domestic and imported livestock slaughtered at the Muneeb abattoir in central Egypt in 2009. Samples were collected from cattle ( $n = 161$ ), buffalo ( $n = 153$ ), sheep ( $n = 174$ ), and camels ( $n = 10$ ). Samples were tested for antibodies against *Leptospira* spp. by a microscopy agglutination test, *Coxiella burnetii* by enzyme immunoassay, *Brucella* spp. by standard tube agglutination, and Rift Valley Fever virus (RVFV), Crimean–Congo hemorrhagic fever virus (CCHFV), sandfly fever Sicilian virus (SFSV), and sandfly fever Naples virus (SFNV) by enzyme-linked immunosorbent assay.

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No competing financial interests exist.

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**Results**—Antibodies against *Leptospira* spp. were identified in 64 (40%) cattle, 45 (29%) buffalo, 71 (41%) sheep, and five (50%) camels; antibodies against *C. burnetii* in six (4%) buffalo, 14 (8%) sheep, and seven (70%) camels; and antibodies against *Brucella* spp. in 12 (8%) cattle, one (1%) buffalo, seven (4%) sheep, and one (10%) camel. Antibodies against RVFV were detected in two (1%) cattle and five (3%) buffalo, and antibodies against CCHFV in one (1%) cow. No antibodies against SFSV or SFNV were detected in any species.

**Discussion**—Results indicate that livestock have been exposed to a number of pathogens, although care must be taken with interpretation. It is not possible to determine whether antibodies against *Leptospira* spp. and RVFV in cattle and buffalo are due to prior vaccination or natural exposure. Similarly, antibodies identified in animals less than 6 months of age may be maternal antibodies transferred through colostrum rather than evidence of prior exposure. Results provide baseline evidence to indicate that surveillance within animal populations may be a useful tool to monitor the circulation of pathogens of veterinary and public health concern in Egypt.

### Keywords

Zoonoses; Rift Valley Fever virus; Crimean–Congo hemorrhagic fever virus; *Coxiella burnetii*; *Brucella*; *Leptospira*; Abattoir; Livestock; Egypt

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### Introduction

Zoonotic pathogens transmitted from animals to humans by inhalation, ingestion, direct contact, or arthropod vectors, result in human morbidity and mortality and economic loss. Considerable attention has been drawn to these pathogens in recent years as their incidence and distribution have increased (Coker et al. 2011, Pal 2013).

In agricultural nations of North Africa and the Middle East, livestock are important to the economy and social structure as sources of labor, meat, dairy, leather, and wool and hair. However, trade in live animals and animal tissues poses a risk to both human and animal health (Ibrahim et al. 1997, Seimenis 1998, World Health Organization 2006) due to the potential emergence and spread of zoonoses (Armelagos 1998, McCarthy and Moore 2000).

Dense comingling of human, animal, and vector populations creates high-risk environments for the spread of zoonotic infections (Maronpot and Barsoum 1972, Vaira et al. 1988, Ibrahim et al. 1997, Gubler, 2004). Locations such as abattoirs, quarantine facilities, and live animal markets offer opportunities to monitor disease spread at sites where humans are in close contact with animals and hematophagous arthropods involved in disease transmission. Focusing surveillance efforts on animal populations within these environments offers an opportunity for the detection and subsequent mitigation of pathogens of concern to both veterinary and public health (McCluskey 2003, Racloz et al. 2006). Although the use of animals as sentinels for human disease is a surveillance technique that is often underused, this methodology is a potentially important and effective tool to monitor emerging pathogens of public health significance (Chomel 2003, Rabinowitz et al. 2005, Halliday et al. 2007).

A baseline serosurvey was conducted to evaluate the feasibility of a surveillance system using animals as sentinels in Egypt. The survey examined the presence of antibodies against a panel of zoonotic pathogens among livestock with a focus on pathogens affecting human health. These included bacterial pathogens (*Leptospira* spp., *Coxiella burnetii*, and *Brucella* spp.) and arboviruses (Rift Valley fever virus [RVFV], Crimean–Congo hemorrhagic fever virus [CCHFV], sandfly fever Sicilian virus [SFSV], and sandfly fever Naples virus [SFNV]).

## Materials and Methods

A serosurvey was conducted at the Muneeb abattoir in the Giza governorate in central Egypt. Livestock slaughtered at this abattoir include cattle, buffalo, sheep, and camels originating in Sudan, Somalia, and governorates throughout Egypt. Slaughter is performed according to Islamic tradition, which requires a deep incision to the animal's throat.

Sample collection took place over a 2-week period in July, 2009. Study teams visited the abattoir on the 2 days each week when the number of animals slaughtered was expected to be highest. On each visit, study teams attempted to collect samples from all livestock slaughtered during routine operational hours of the abattoir. No measures were taken to target specific animals or subgroups of animals. Samples were collected in 50-mL conical tubes as blood drained from the jugular vein or carotid artery of each animal immediately after slaughter. Age, sex, and location of origin (country and/or governorate) were recorded for each animal on the basis of information provided by caretakers after sample collection with verification of sex and approximate age by visual inspection by collaborating veterinarians. Despite attempts to collect vaccination history for each animal, this information was unavailable.

Samples were stored on site in an insulated container and then transported by vehicle to US Naval Medical Research Unit No. 3 (NAMRU-3) in Cairo, Egypt, where serum was separated within 6–8 h of sample collection and stored at – 20°C until analyses were performed.

To assess *Leptospira* spp. exposure, microscopic agglutination test (MAT) was performed according to procedures described elsewhere (Kurtoglu et al. 2003, Parker et al. 2007). Briefly, test serum dilutions were separately mixed with individual *Leptospira* cultures and incubated for 1 h at room temperature. All samples ( $n = 498$ ) were tested against *L. interrogans* serovar Grippotyphosa (serogroup Grippotyphosa, strain Moskva V), *L. borgpetersenii* serovar Hardjo (serogroup Sejroe), *L. interrogans* serovar Icterohaemorrhagiae (serogroup Icterohaemorrhagiae, strain RGA), and *L. interrogans* serovar Pomona (serogroup Pomona, strain Pomona). Samples negative for those serovars ( $n = 99$ ) were then tested against *L. interrogans* serovar Australis (serogroup Australis, strain Ballico), *L. borgpetersenii* serovar Ballum (serogroup Ballum, strain Mus 127), *L. interrogans* serovar Bataviae (serogroup Bataviae, strain Van Tienen), *L. interrogans* serovar Bratislava (serogroup Australis, strain Jez Bratislava), *L. interrogans* serovar Canicola (serogroup Canicola, strain Ruebush), *L. weilii* serovar Celledoni (serogroup Celledoni, strain Celledoni), *L. interrogans* serovar Djasiman (serogroup Djasiman, strain

Djasiman), *L. santarosai* serovar Georgia (serogroup Mini, strain LT 117), and *L. interrogans* serovar Pyrogenes (serogroup Pyrogenes, strain Salinen). Samples negative for both sets of serovars ( $n = 38$ ) were then tested against *L. santarosai* serovar Alexi (serogroup Pyrogenes, strain HS616), *L. interrogans* serovar Autumnalis (serogroup Autumnalis, strain Akiyami A), *L. santarosai* serovar Borincana (serogroup Hebdomadis, strain HS622), *L. kirschneri* serovar Cynopteri (serogroup Cynopteri, strain 3522 C), *L. santarosai* serovar Borincana (serogroup Hebdomadis, strain HS622), *L. borgpetersenii* serovar Javanica (serogroup Javanica, strain Veldrat Batavia 46), *L. interrogans* serovar Mankarso (serogroup Icterohaemorrhagiae, strain Mankarso), *L. borgpetersenii* serovar Tarassovi (serogroup Tarassovi, strain Perepelicyan), and *L. interrogans* serovar Wolffi (serogroup Sejroe, strain 3705). Samples were considered positive if dark-field microscopy showed at least 50% agglutination or lysis at a titer of at least 1:800 for one or more serovars. This cutoff was used to minimize nonspecific and multiple cross-reactions among *Leptospira* serovars and strains (Felt et al. 2011). Cultures used for the test were provided by the National Center for Emerging and Zoonotic Infectious Diseases at the US Centers for Disease Control and Prevention (CDC, Atlanta, GA).

An enzyme immunoassay was used for the detection of *C. burnetii* antibodies in serum samples according to manufacturer's instructions and recommended cutoff values (IDEXX Laboratories, Inc., Westbrook, Maine). Standard tube agglutination (STA) was used to assess exposure to *Brucella* spp. following manufacturer's instructions (SA Scientific Inc., San Antonio, TX) and using antigen for *Brucella abortus* (strain USDA #119-3). Samples were considered positive by agglutination titers greater than 1:160 (Moustafa et al. 1998).

Serum was tested by indirect enzyme-linked immunosorbent assay (ELISA) for immunoglobulin G (IgG) antibodies against RVFV, SFSV, and SFNV using in-house antigen assays (Botors et al. 2004, Ellis et al. 2008, Riddle et al. 2008). Likewise, detection of IgG antibodies against CCHFV was performed using ELISA (US Army Medical Research Institute for Infectious Diseases, Fort Detrick, MD) (Mustafa et al. 2011). Modifications involved the use of anti-bovine IgG horseradish peroxidase (Kirkegaard & Perry Laboratories, Inc., Gaithersburg, MD) for sera from cattle, buffalo, and camels, whereas anti-sheep IgG horseradish peroxidase (Kirkegaard & Perry Laboratories, Inc., Gaithersburg, MD) was employed for sera from sheep. In a subset of samples, the anti-ruminant IgG conjugate supplied with *C. burnetii* kits (IDEXX Laboratories, Inc., Westbrook, ME) and protein A-horseradish peroxidase conjugates (HRP-Protein A, Life Technologies-Invitrogen Corporation, Camarillo, CA) were also used for sera from camels for confirmation (Wernery et al. 2012). Optical density (O.D.) of the reactions was measured at a wavelength of 405 nm. Samples with O.D. values greater than three standard deviations above the mean of the negative control sera were considered positive. Positive samples were confirmed by a two-fold serial dilution.

Descriptive data and laboratory results were entered into a Microsoft Access database using double data entry to ensure accuracy. Data were analyzed using SAS software, version 9.3, of the SAS System for Windows (SAS Institute Inc., Cary, NC). The proportion and standard error (SE) of antibody-positive animals was calculated for each pathogen of interest, in the overall study population and within each species. Investigators examined

whether any species of animals was more likely than any other species to be exposed to specific pathogens of interest or multiple pathogens of interest. These bivariate analyses used Mantel–Haenszel estimates to calculate odds ratios (OR) and 95% confidence intervals (CI) and the Mantel–Haenszel chi-squared test to assess statistical significance. Associations were considered significant by a  $p$  value less than 0.05.

## Results

The study population consisted of 498 animals including 161 (32%) cattle, 153 (31%) buffalo, 174 (35%) sheep, and 10 (2%) camels, as shown in Table 1. All cattle, buffalo, and sheep were reported to have originated within Egypt. Camels were the only imported species, with eight (80%) coming from Sudan and two (20%) from Somalia.

Antibodies against at least one pathogen of interest were identified in 221 (44%, SE = 2%) animals, and antibodies against multiple pathogens of interest were found in 19 (4%, SE = 1%) animals, as shown in Table 2. Among samples in which antibodies against multiple pathogens of interest were identified, antibodies against *Leptospira* spp. were present in all except two samples.

Camels had the highest proportion of exposure, with nine (90%, SE = 10%) animals demonstrating antibodies against at least one pathogen and four (40%, SE = 16%) animals with antibodies against multiple pathogens. No differences were found in camel results when ELISA was performed using anti-bovine, anti-ruminant, or protein A conjugates. Eighty-four (48%, SE = 4%) sheep, 74 (46%, SE = 4%) cattle, and 54 (35%, SE = 4%) buffalo showed antibodies against at least one pathogen of interest, whereas antibodies against multiple pathogens of interest were identified in seven (4%, SE = 1%), five (3%, SE = 1%), and three (2%, SE = 1%) sheep, cattle, and buffalo, respectively. All 97 animals under 6 months of age were buffalo. Of these, 19 (20%, SE = 4%) animals demonstrated antibodies against *Leptospira* spp., five (5%, SE = 2%) against *C. burnetii*, and four (4%, SE = 2%) against RVFV.

The highest titers in MAT-positive cattle were identified for *L. borgpetersenii* serovar Ballum ( $n = 5/26$ ), *L. interrogans* serovar Bataviae ( $n = 4/26$ ), and *L. kirschneri* serovar Cynopteri ( $n = 3/8$ ), whereas *L. borgpetersenii* serovar Ballum ( $n = 7/29$ ), *L. interrogans* serovar Bataviae ( $n = 5/29$ ), and *L. interrogans* serovar Bratislava ( $n = 4/29$ ) demonstrated the highest titers among MAT-positive buffalo. Among MAT-positive sheep, *L. kirschneri* serovar Cynopteri ( $n = 6/22$ ) and *L. borgpetersenii* serovar Ballum ( $n = 10/43$ ) were most frequent, followed equally by *L. interrogans* serovar Australis ( $n = 3/43$ ), *L. interrogans* serovar Bataviae ( $n = 3/43$ ), and *L. interrogans* serovar Pyrogenes ( $n = 3/43$ ). Only one serovar, *L. interrogans* serovar Icterohaemorrhagiae, demonstrated high titers in the single MAT-positive camel ( $n = 1/10$ ). High-titer MAT results are shown in Table 3, which also illustrates the high levels of cross-reaction between serovars and serogroups analyzed.

Camels were more likely than other species to harbor antibodies against *C. burnetii* (OR = 54, 95% CI 13–227,  $p < 0.01$ ), whereas cattle were more likely than other species to demonstrate antibodies against *Brucella* spp. (OR = 3, 95% CI 1–7,  $p = 0.02$ ). Buffalo were

less likely than other species to show antibodies against *Leptospira* spp. (OR = 0.6, 95% CI 0.4–0.9,  $p = 0.02$ ) or *Brucella* spp. (OR = 0.1, 95% CI 0.0–0.8,  $p = 0.03$ ). The species most likely to show antibodies against multiple pathogens of interest was camels (OR = 21, 95% CI 5–82,  $p < 0.01$ ). No other associations between species and pathogens of interest were statistically significant, perhaps due to limited sample size.

## Discussion

Results of the current survey may not be representative of the livestock population in Egypt, yet they provide a snapshot of zoonotic pathogens circulating in a country where livestock represents 25% of the agricultural gross domestic product (El-Nahrawy 2011). Antibodies against bacterial pathogens (*Leptospira* spp., *C. burnetii*, and *Brucella* spp.) were common among animals in this study, but antibodies against arboviral pathogens (RVFV, CCHFV, SFSV, and SFNV) were more limited. Whereas antibodies against *Brucella* spp. and RVFV among cattle and buffalo may be attributed to prior vaccination, immunization among other livestock species and against other pathogens of interest is not performed in Egypt. Therefore, prior infection is most likely responsible for other observed antibodies, with the exception of livestock under 6 months of age, among which maternal antibodies transferred through colostrum may have been detected.

*Leptospira* spp. was the most common pathogen against which antibodies were identified in this study. The proportion of antibody-positive animals was 40% (SE < 1%) among cattle and 29% (SE = 4%) among buffalo, comparable to previous findings of 44% and 20%, respectively, in 2006–2007 (Felt et al. 2011). The proportion of sheep demonstrating antibodies against *Leptospira* spp. was 41% (SE = 4%), apparently higher than that previously identified (Felt et al. 2011), although this discrepancy may be due to regional variations in sampling (Felt et al. 2011). The identification of antibodies against *Leptospira* spp. in 50% (SE = 17%) of camels can only be compared to data from nearly 40 years ago, which noted a much lower proportion of animals exposed (Maronpot and Barsoum 1972). Given that vaccination against *Leptospira* spp. is not performed among ruminants in Egypt, it is likely that livestock demonstrating antibodies against *Leptospira* spp. have been naturally infected with the bacteria except for the 19 antibody-positive buffalo less than 6 months of age, which may carry maternal antibodies at this age (Ryan et al. 2012).

MAT is the only reliable method for identifying *Leptospira* spp. serovars in animals, but the use of serology results to identify an infecting serovar in individual cases is less appropriate than previously thought. However, results may be useful in predicting immunodominant serogroups circulating within a population (Bharti et al. 2003, Levett 2003). Interpretation of results is complicated by common cross-reactions between serogroups and paradoxical reactions that direct the initial immune response toward an alternative serovar or serogroup (Levett 2003). Several serogroups were highly reactive in the current study, although many samples were responsive to numerous other serovars; some samples demonstrated antibodies against as many as nine serovars, despite the use of cutoff values designed to minimize detection of cross-reactions. It is not possible to determine whether these results occurred as a result of natural exposure to multiple serovars or as a result of cross-reactivity during

testing. Although individual infecting serovars cannot be determined, results may still provide support for serogroups circulating within the study population.

*C. burnetii* was the next most frequent pathogen identified in the study, although the overall proportion of animals with antibodies against *C. burnetii* was lower than in a 2007 study (Mazyad and Hafez 2007), with the exception of camels. Discrepancies between these studies may indicate variation in the geographic distribution of *C. burnetii* due to regional differences in sampling, although differences in laboratory methods and seasonality of sample collection may also explain these differences. Of the six buffalo with antibodies against *C. burnetii*, maternal antibodies may have been detected in the five (83%) less than 6 months of age. Camels were more likely than other species in the current study to harbor antibodies against *C. burnetii*, perhaps due to increased opportunity of exposure during migration from Sudan and Somalia.

Proportions of antibodies against *Brucella* spp. among animals in the current study are comparable to those reported by a 1998 study, with slight variations that may be attributed to differences in laboratory methods and interpretation (Refai 2002). It is also possible that vaccination practices and coverage, which would influence antibody levels, have shifted over the past decade. Vaccination against *Brucella* spp. is currently practiced in Egypt, employing *B. abortus* vaccine (strain 19) and *B. melitensis* vaccine (strain Rev. 1) to vaccinate cattle, which were more likely than other species in this study to have antibodies against *Brucella* spp., and buffalo. Thus, it is not possible to determine whether positive STA results in these species indicate prior vaccination or natural infection. *B. abortus* vaccine (strain RB-51), which would induce immunity without producing antibodies that react in this test, is not currently available in Egypt. Because sheep and camel are not regularly vaccinated against *Brucella* spp., it is expected that antibody presence in these species reflects prior infection.

Antibodies against RVFV were detected in two (1%, SE = 1%) cattle and five (3%, SE = 1%) buffalo in this study. Although RVFV has been responsible for two major outbreaks in Egypt, first in 1977 and again in 1993 (Darwish et al. 1994, Corwin et al. 1992, Arthur et al. 1993, Laughlin et al. 1979), cattle and buffalo are vaccinated against infection with RVFV so it is likely that antibodies identified in this study reflect prior vaccination rather than recent infection. Furthermore, four (80%) of the antibody-positive buffalo were under the age of 6 months and thus may demonstrate maternal antibodies (Swanepoel, no date). It is notable that no sheep or camels, species that would not be vaccinated against RVFV, demonstrated antibodies against RVFV.

In this study, antibodies against CCHFV were identified in only one cow (1%, SE = 1%), which is unexpected given the higher levels of exposure reported in previous studies, including a serologic study in 2004–2005 that showed antibodies in 3% of cattle, < 1% of buffalo, 6% of sheep, and 1% of goats (Mohamed et al. 2008) and a study from 1986–1987 that showed antibodies in 14% of imported camels but no domestic cattle or sheep (Morrill et al. 1990). Furthermore, a previously undocumented variant of CCHFV was identified in ticks collected from five camels within this same study (Chisholm et al. 2011), although these ticks may have fed on different animals before transferring to unexposed animals

sampled in this study. The sample size in the current study is also low compared to previous studies, although geographic or temporal variations between the studies may also account for the differences in antibody levels.

No animals in the current study harbored antibodies against SFSV or SFNV. Data on vertebrate reservoirs of SFSV and SFNV are limited (Depaquit et al. 2010), and no information on animal infection is available in Egypt. Community-based surveys of human exposure in the Nile Delta of Egypt showed proportions of 4% and 2%, respectively, for SFSV and SFNV under normal conditions (Corwin et al. 1993), and 46% and 21%, respectively, during extreme floods (Darwish et al. 1994). If these limited data suggest a low general circulation of sandfly fever viruses in Egypt, it is not surprising that no positive samples were identified within this small study population.

Camels were more likely to have antibodies against multiple pathogens, which may be related to their increased age or the greater distance these animals traveled from Sudan and Somalia, which would have allowed opportunity for exposure in a variety of geographic and climatic environments. However, the sample size for camels was particularly small, so it is difficult to determine whether these findings are skewed by limited data.

## Conclusions

Results from this study provide an update on exposure to viral and bacterial pathogens among a sample of livestock in Egypt. Although results of this small serosurvey are not representative of the broader livestock population in the country, the study's successful implementation indicates that surveillance within animal populations may be a useful tool to monitor the circulation of pathogens of veterinary and public health concern in Egypt. However, the methodology used in this study is not appropriate for examining *Brucella* spp. and RVFV among cattle and buffalo, or other pathogens or species for which vaccination is routine, unless animals' vaccination status can be determined with adequate confidence to distinguish between prior infection and vaccination. Future studies may also consider excluding young animals that may demonstrate maternal antibodies absorbed from consumption of colostrum.

Further efforts are needed to evaluate the circulation of pathogens examined in this study in the broader livestock population within Egypt. The methodology used in this study provides a relatively simple tool for future cross-sectional studies or ongoing active surveillance. Agencies involved in human and animal health should consider the use of this methodology as a tool to monitor the spread of endemic pathogens and to identify emerging infections of concern to both human and animal health.

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Table 1

## Study Population Characteristics by Species

	Cattle		Buffalo		Sheep		Camels		Total	
	n	%	n	%	n	%	n	%	n	%
Age										
< 6 months	0	0%	97	63%	0	0%	0	0%	97	19%
6 months to 1 year	10	6%	2	1%	169	97%	0	0%	181	36%
> 1 year	133	83%	48	31%	4	2%	7	70%	192	39%
Unknown	18	11%	6	4%	1	1%	3	30%	28	6%
Sex										
Male	156	97%	111	73%	174	100%	7	70%	448	90%
Female	5	3%	42	27%	0	0%	0	0%	47	9%
Unknown	0	0%	0	0%	0	0%	3	30%	3	1%
Location of origin										
Lower Egypt	129	80%	112	73%	123	71%	0	0%	364	73%
Greater Cairo	3	2%	13	8%	15	9%	0	0%	31	6%
Upper Egypt	21	13%	21	14%	4	2%	0	0%	46	9%
Unknown Egypt	8	5%	7	5%	32	18%	0	0%	47	9%
Sudan	0	0%	0	0%	0	0%	8	80%	8	2%
Somalia	0	0%	0	0%	0	0%	2	20%	2	0%
Total	161	100%	153	100%	174	100%	10	100%	498	100%

Numbers (n), Percentages (%), and Standard Errors (SE) of Animals with Immunoglobulin G Antibodies against Viral and Bacterial Pathogens by Animal Species

Table 2

	Cattle (n = 161)			Buffalo (n = 153)			Sheep (n = 174)			Camels (n = 10)			Total (n = 498)		
	n	%	SE	n	%	SE	n	%	SE	n	%	SE	n	%	SE
<i>Leptospira</i> spp.	64	40%	0%	45	29%	4%	71	41%	4%	5	50%	17%	185	37%	2%
<i>Coxiella burnetii</i>	0	0%	0%	6	4%	2%	14	8%	2%	7	70%	15%	27	5%	1%
<i>Brucella</i> spp.	12	7%	2%	1	1%	1%	7	4%	2%	1	10%	10%	21	4%	1%
RVFV	2	1%	1%	5	3%	1%	0	0%	0%	0	0%	0%	7	1%	1%
CCHFV	1	1%	1%	0	0%	0%	0	0%	0%	0	0%	0%	1	0%	0%
At least one pathogen of interest	74	46%	4%	54	35%	4%	84	48%	4%	9	90%	10%	221	44%	2%
<i>Leptospira</i> spp. + <i>C. burnetii</i>	0	0%	0%	1	1%	1%	5	3%	1%	3	30%	15%	9	2%	1%
<i>Leptospira</i> spp. + <i>Brucella</i> spp.	3	2%	1%	0	0%	0%	2	1%	1%	0	0%	0%	5	1%	0%
<i>Leptospira</i> spp. + <i>C. burnetii</i> + <i>Brucella</i> spp.	0	0%	0%	0	0%	0%	1	1%	1%	0	0%	0%	1	0%	0%
<i>Leptospira</i> spp. + RVFV	1	1%	1%	2	1%	1%	0	0%	0%	0	0%	0%	3	1%	0%
<i>Leptospira</i> spp. + CCHFV	1	1%	1%	0	0%	0%	0	0%	0%	0	0%	0%	1	0%	0%
<i>C. burnetii</i> + <i>Brucella</i> spp.	0	0%	0%	0	0%	0%	2	1%	1%	1	10%	10%	3	1%	0%
Any combination of pathogens of interest	5	3%	1%	3	2%	1%	9	5%	1%	4	40%	16%	21	4%	1%

SE, standard error; RVFV, Rift Valley fever virus; CCHFV, Crimean–Congo hemorrhagic fever virus.

Table 3

Serovars with High-Titer Reactions Identified among MAT-Positive Results

<i>Leptospira</i> serovar	Cattle			Buffalo			Sheep			Camels			Total		
	n	%	Total	n	%	Total	n	%	Total	n	%	Total	n	%	Total
<i>L. interrogans</i> serovar Grippityphosa	4	2%	161	3	2%	153	5	3%	174	0	0%	10	12	2%	498
<i>L. borgpetersenii</i> serovar Hardjo	4	2%	161	3	2%	153	5	3%	174	0	0%	10	12	2%	498
<i>L. interrogans</i> serovar Icterohaemorrhagiae	4	2%	161	3	2%	153	5	3%	174	1	10%	10	13	3%	498
<i>L. interrogans</i> serovar Pomona	4	2%	161	3	2%	153	5	3%	174	0	0%	10	12	2%	498
Total high-titer samples in group 1	4	2%	161	3	2%	153	5	3%	174	1	10%	10	13	3%	498
<i>L. interrogans</i> serovar Australis	1	4%	26	3	10%	29	3	7%	43	0	0%	1	7	7%	99
<i>L. borgpetersenii</i> serovar Ballum	5	19%	26	7	24%	29	10	23%	43	0	0%	1	22	22%	99
<i>L. interrogans</i> serovar Bataviae	4	15%	26	5	17%	29	3	7%	43	0	0%	1	12	12%	99
<i>L. interrogans</i> serovar Bratislava	2	8%	26	4	14%	29	2	5%	43	0	0%	1	8	8%	99
<i>L. interrogans</i> serovar Canicola	1	4%	26	2	7%	29	1	2%	43	0	0%	1	4	4%	99
<i>L. weilii</i> serovar Celledoni	1	4%	26	2	7%	29	1	2%	43	0	0%	1	4	4%	99
<i>L. interrogans</i> serovar Djasiman	1	4%	26	2	7%	29	2	5%	43	0	0%	1	5	5%	99
<i>L. santarosai</i> serovar Georgia	1	4%	26	2	7%	29	1	2%	43	0	0%	1	4	4%	99
<i>L. interrogans</i> serovar Pyrogenes	3	12%	26	3	10%	29	3	7%	43	0	0%	1	9	9%	99
Total high-titer samples in group 2	9	35%	26	12	41%	29	13	30%	43	0	0%	1	34	34%	99
<i>L. kirschneri</i> serovar Cynopteri	3	38%	8	0	0%	7	6	27%	22	0	0%	1	9	24%	38
Total high-titer samples in group 3 <sup>a</sup>	3	38%	8	0	0%	7	6	27%	22	0	0%	1	9	24%	38

<sup>a</sup> All samples tested for the following serovars (eight cattle, seven buffalo, 22 sheep, one camel) were negative: *L. santarosai* serovar Alexi, *L. interrogans* serovar Autumnalis, *L. santarosai* serovar Bortincana, *L. santarosai* serovar Borincana, *L. borgpetersenii* serovar Javanica, *L. interrogans* serovar Mankarso, *L. borgpetersenii* serovar Tarassovi, *L. interrogans* serovar Wolffii.

MAT, microscopic agglutination test.