

## ***Pasteurella multocida* and *Salmonella* Affections in Arabian Oryx**

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### **Abstract**

Arabian Oryx belong to bovine species, and the smallest member of the genus Oryx, native to the Arabian Peninsula. The population numbers and the Arabian Oryx frequency distribution at different sites of Bahrain (Al-Areen wildlife Park / Reserve, Al-Areen wildlife Park / Visitors' Center and Hawar Island) 70%, 14% and 16% respectively. Males represents 29.4%, while females are 70.6% of the total population. Samples have been assembled from apparently diseased Arabian Oryx from three districts in Bahrain where the Arabian Oryx found, 25 Oryx with positive respiratory signs was tested for *Pasteurella multocida*, while 30 Oryx with digestive tract signs was tested for Salmonellosis. After the microbiological evaluation, from the 25 animals showing clinical signs, four animals only tested positive for *Pasteurella multocida*, 3 of them are females and one male and all of them were younger than 1 year of age. The obtained results concluded that, Arabian Oryx were adapted to the weather in Bahrain, but they are still susceptible to diseases, especially in the young ages. More effort from relevant authorities is required to maintain control *Pasteurella multocida* and salmonella infection in Arabian Oryx.

**Keywords:** Arabian Oryx, *P. multocida*, *Salmonella*, Al Areen, Wildlife, Bahrain

### **Introduction**

Arabian Oryx (*Oryx leucoryx*) is the official national animal of Bahrain. It is known as Arabian Oryx or Oryx and has a variety of names including Al maha, Al wudhaihy and baqar al wahsh in Arabic language, while in English it is called white Oryx, which can be observed in the Sinai, the

Transjordan, Palestine, much of Iraq, and vast majority of the Arabian Peninsula. (*Talbot & Merriam, 1960*).

It was thought to be disappeared from the wild by the 1970s, but it was preserved in private preserves and zoos and reinstated into the wild in 1980. (*Massicot, 2007*). The Arabian Oryx had been classified as

endangered upon the IUCN Red List in 1986, and it was the initial mammal to be reclassified as vulnerable after being declared nonexistent in the wild in 2011. It is listed in Appendix I in the CITES list. In 2011, natural populations were estimated to number over 1,000 individuals, with 6,000–7,000 in captivity around the world. (*IUCN, 2017*).

Arabian Oryx had been re-introduced to Bahrain, UAE, Saudi Arabia, Palestine, Syria and Jordan and with a little populace in Hawar Island of Bahrain, and substantial semi-managed populaces at numerous sites in Bahrain and the UAE. In Bahrain, in 1976 H. H. the later Sheikh Zayed bin Sultan Al Nahyan sent 6 Arabian Oryx to Bahrain to H. H. Sheikh Hammad Bin Isa Al Khalifa (Was the Crown prince) to start a breeding program in Bahrain. H. H. Sh. Hammad established a protected area for the Arabian Oryx within the Southern area of Bahrain (Al Areen Wildlife Park) and started a perfect breeding program. (*Massicot, 2007*).

During the day, Arabian Oryx rest throughout the day and can identify rain and go towards it, allowing them to roam huge areas; a flock in Oman may range over three thousand km<sup>2</sup> (1,200 sq mi). Groups are of both sexes and normally include between 2 and 15 Arabian Oryx, while groups of about a hundred individuals was documented. Arabian Oryx are not usually aggressive towards each

other, which grants herds to live peacefully for certain time (*Massicot, 2007*).

Oryx may live for almost 20 years in captivity and under ideal conditions in wild areas (*The Phoenix Zoo, 2008*). Wolves remain the only hunters of the Arabian Oryx, aside from humans. During droughts, however, starvation and dehydration can dramatically shorten their life expectancy. Male fights, snake bites, sickness, and drowning during floods are amongst the other causes of death.

Disease transmission hazards are improved for the repositioned animals themselves, along with for local wildlife and local domestic species (*Woodford, 1989*). Arabian Oryx seem to be susceptible to extremely pathogenic bacteria and viruses which affect domestic ruminants (*Greth et. al., 1992*). The chief aim of this study was thus to evaluate the exposure of Arabian Oryx from various locations to selected infectious diseases found in domestic and wild species.

In Bahrain, there are 143 Arabian Oryx distributed in three sites: Al-Areen wildlife Park/ Reserve section, Al-Areen wildlife Park / Visitors Center section and Hawar Island. Therefore, the aim study to know the common bacteria causing respiratory and gastrointestinal diseases in the Arabian Oryx from Al Areen Wildlife Park – Kingdom of Bahrain.

## Materials and methods

**Study area:** The Arabian Oryx can be found distributed in three different areas in Bahrain: Al Areen Wildlife Park-Visitors Center (3 square kilometers), Al Areen Wildlife Park-Reserve district (4 square kilometers) and also Hawar Island (52 square kilometers). All these animals are included in this study.

**Sample Size:** Kingdom of Bahrain has a complete of 143 Arabian Oryx distributed as the following: 20 animals in Al Areen Wildlife Park (visitors Center), 100 animals in Al Areen Wildlife Park (Reserve) and 23 animals in Hawar Island. A total of 143 Arabian Oryx had been categorized based on their gender into males and females in the different districts (Al Areen Wildlife Park (visitors Center), Al Areen Wildlife Park (Reserve) and Hawar Island). The animals are then classified corresponding to their age into 2 groups including: Young age (1 to 12 month), Old age (over 1 years). Weight, length, height, and body temperature had been measured and the average were calculated.

**Samples' collection:** A total of 25 serum and 25 nasal discharges samples had been collected from 25 animals showing positive respiratory signs (body temperature >39.5°C, respiratory rate >40/m, with cough and nasal discharge, auscultation reveals crackle/snoring/whistle sounds), and massive percussion. For serum,

5 ml whole blood had been collected through jugular venipuncture in a vacutainer and then set aside to clot at 25°C for 20 min. The serum was separated, clarified by centrifuge at 4000 rpm for 15 min and kept at -20°C till the tests were performed. Thirty fecal samples were collected from animals showing positive gastrointestinal signs (body temperature >39.5°C, weight loss, absence of regurgitation, abdominal pain in palpation, auscultation & percussion (air and fluid), bloody diarrhea or diarrhea without blood).

### Detection and Isolation of *P. multocida*

*Pasteurella* species are spherical, ovoid or rod-shaped Gram-negative rods or coccobacilli which occur singly or in pairs or short chains. Bipolar staining commonly occurs; capsules may be present. (PHE, 2015). Nasal swabs were cultured according to Bote et al. 2017.

The 3 morphologically similar pasteurellaceae species and genera were expected to grow upon sub culturing according to Carter et al. (1990).

### Detection and Isolation of *Salmonella*

*Salmonella* isolation and identification was conducted in accordance with the guidelines of the International Organization for Standardization (ISO 6579:2017).

If growth was detected as Hadimli et al. (2017). A loopful from enrichment semi solid cultures was

streaked according to *Quinn et al. (2002)*.

### Results

Current results revealed that, animals with positive signs are 25 but the negative are 118. These 25 animals with positive respiratory signs consist of 18 females and 7 males, while by age point of view the results showed 19 animals represented with positive respiratory signs were younger than 1 year and 6 animals were older than 1 year.

Animals with elevated body temperature higher than 39.5 were 22 (5 males and 17 females) and 16 animals were at age of 1 year old and younger while 6 were older than 1 year of age. Referring to the respiratory rate, all the animals had more than 40/minute resembled in 18 females and 7 males, 17 of them were at age of 1 year and younger while 8 animals were older than 1 year of age. Animals with nasal discharge / cough were meant to be 20 / 21; 16 females and 4 males / 16 females and 5 males, 14 / 13 were

less than one year, and the 6 / 8 were older; respectively. By the percussion and auscultation methods of diagnosis it has been revealed a massive percussion while by auscultation there was crackle, snoring and whistle sounds.

Table (6) demonstrates the Prevalence percentage of the digestive tract signs of the Arabian Oryx which was 21 (14.7%) of the females represented with GIT clinical signs while the males were only 9 (6.3%). While according to age (recorded in Table (7)) 13 (9.1%) animals below than one year had positive clinical signs while 17 (11.9%) were older than one year.

The frequency distribution of clinical gastrointestinal tract signs (Body temperature >39.5°C, Weight loss, Absence of regurgitation, Palpation abdominal pain, Percussion & Auscultation: Air & fluids, bloody diarrhea, or diarrhea without Blood) recorded correspondingly in diseased Arabian Oryx on 30 animals is described in respect to sex in table (8) and age in table (9).

**Table (1)** Prevalence of *Pasteurella multocida* in Arabian Oryx after microbiological evaluation based on animals showing positive signs in respect to sex (n=25)

Items	Arabian Oryx Sex	
	Female No (%)	Male No (%)
Positive	3 (2.1)	1 (0.7)
Negative	15 (10.5)	6 (4.2)
Total	18 (12.6)	7 (4.9)

**Table (2)** Prevalence of *Pasteurella multocida* in Arabian Oryx after microbiological evaluation based on animals showing positive signs in respect to age (n=25)

Items	Arabian Oryx Age	
	< 1year No (%)	> 1year No (%)
Positive	4 (2.8)	0 (0.0)
Negative	15 (10.5)	6 (4.2)
Total	19 (13.3)	6 (4.2)

**Table (3)** Frequency distribution of *Pasteurella multocida* in Arabian Oryx after microbiological evaluation-based samples examine in respect to Sex (n=25)

Items	Arabian Oryx Sex	
	Female No (%)	Male No (%)
Positive	3 (12)	1 (4)
Negative	15 (60)	6 (24)
Total	18 (72)	7 (28)

**Table (4)** Frequency distribution of *Pasteurella multocida* in Arabian Oryx after microbiological evaluation-based samples examine in respect to Age (n=25)

Items	Arabian Oryx Age	
	< 1year No (%)	> 1year No (%)
Positive	4 (16)	0 (0.0)
Negative	15 (60)	6 (24)
Total	19 (76)	6 (24)

**Table (5)** Prevalence of *Pasteurella multocida* in Arabian Oryx according to wildlife districts

	Wildlife districts		
	Al-Areen Wildlife Park-Reserve	Al-Areen Wildlife Park - Visitors Center	Hawar island
Female No (%)	2 (2%)	0 (0%)	1 (4.4%)
Male No (%)	1 (1%)	0 (0%)	0 (0.0%)
Total	3 (3%)	0 (0%)	1 (4.4%)

**Table (6)** The prevalence (%) of GIT signs in the Arabian Oryx according to sex (n = 143)

Items	Arabian Oryx Sex	
	Female No (%)	Male No (%)
Positive Signs	21 (14.7)	9 (6.3)
Negative Signs	80 (56)	33 (23)
Total	101 (70.7)	42 (29.3)

**Table (7)** The prevalence (%) of GIT signs in the Arabian Oryx according to age (n = 143)

Items	Arabian Oryx Age	
	< 1year No (%)	> 1year No (%)
Positive Signs	13 (9.1)	17 (11.9)
Negative Signs	14 (9.8)	99(69.2)
Total	27 (18.9)	116 (81.1)

**Table (8)** Frequency distribution of clinical GIT signs recorded in Arabian Oryx according to sex (n = 30)

Items	Arabian Oryx Sex	
	Female No (%)	Male No (%)
Body Temp. >39.5°C	21 (70)	9 (30)
Weight loss	10 (33.3)	4 (13.3)
Absence of regurgitation	12 (40)	9 (30)
Palpation Abdominal Pain	3 (10)	2 (6.7)
Auscultation & percussion (Air and fluid)	17 (56.7)	7 (23.3)
Diarrhea with/without blood	20 (66.7)	8 (26.7)

**Table (9)** Frequency distribution of clinical GIT signs recorded in Arabian Oryx according to age (n = 30)

Items	Arabian Oryx Age	
	< 1year No (%)	> 1year No (%)
Body Temp. >39.5°C	7 (23.3)	23 (76.7)
Weight loss	9 (30)	5 (16.7)
Absence of regurgitation	10 (33.3)	11 (36.7)
Palpation Abdominal Pain	4 (13.3)	1 (3.33)
Auscultation & percussion (Air and fluid)	11 (36.7)	13 (43.3)
Diarrhea with or without blood	15 (50)	13 (43.3)

**Table (10)** Prevalence of *Salmonella* spp. in Arabian Oryx after microbiological evaluation according to total population in respect to sex(n=143)

Items	Arabian Oryx Sex	
	Female No (%)	Male No (%)
Positive	6 (4.2)	3 (2.1)
Negative	95 (66.4)	39 (27.3)
Total	101 (70.6)	42 (29.4)

**Table (11)** Prevalence of *Salmonella* spp. in Arabian Oryx after microbiological evaluation according to total population in respect to age (n=143)

Items	Arabian Oryx Sex	
	< 1year No (%)	> 1year No (%)
Positive	8 (5.6)	1 (0.7)
Negative	57 (39.9)	77 (53.8)
Total	65 (45.5)	78 (54.5)

**Table (12)** Frequency distribution of *Salmonella* spp. in Arabian Oryx after microbiological evaluation-based on their sex (N= 30)

Items	Arabian Oryx Sex	
	Female No (%)	Male No (%)
Positive	6 (20)	3 (10)
Negative	15 (50)	6 (20)
Total	21 (70)	9 (30)

**Table (13)** Frequency distribution of *Salmonella* spp. in Arabian Oryx after microbiological evaluation-based on samples examines, in respect to their age (N= 30)

Items	Arabian Oryx Age	
	< 1year No (%)	> 1year No (%)
Positive	8 (26.7)	1 (3.3)
Negative	19 (63.3)	2 (6.7)
Total	27 (90)	3 (10)

**Table (14)** Prevalence of *Salmonella* spp. in Arabian Oryx according to wildlife districts

	Wildlife districts		
	Al-Areen wildlife Park – reserve	Al-Areen wildlife Park – visitors Center	Hawar Island
Female No (%)	4 (4)	1 (5)	1 (4.3)
Male No (%)	1 (1)	2 (10)	0 (0)
Total	5 (5)	3 (15)	1 (4.3)

**Table (15)** Prevalence of *Salmonella* spp (n=9) isolated from Arabian Oryx based on total population (n=143)

Identified strains	Group	Antigenic structure		Prevalence
		O	H	
<i>S. Dublin</i>	B	1,9,12	g, p	4 (2.8%)
<i>S. Enteritidis</i>	C3	1,9,12	i: 1,7	4 (2.8%)
<i>S. Typhimurium</i>	D1	1,4[5],12	i: 1,2	1 (0.7%)

O: Somatic antigen

H: Flagellar antigen

## Discussion

Current results revealed that, Animals with positive signs are 25 even though the negative are 118. These 25 animals with positive respiratory signs consist of 18 females and 7 males, while by age point of view the results showed 19 animals represented with positive respiratory signs were younger than 1 year and 6 animals were older than 1 year. This may regard to strong evidence of *P. Multocida* in Calves more than adults, where these results agreed with (*Van Donkersgoed et al., 1993; Sivula et al., 1996; Singer et al., 1998; Virtala et al., 2000; Hirose et al., 2003 and Catry et al., 2006*). Regarding to the frequency distribution of clinical respiratory signs in table (8) and table (9) and other signs like atrophic rhinitis were detected these signs almost similar to those been mentioned by (*Wilson and Ho, 2013*).

Other researchers found similar findings in calves from Brazilian rural settlement cows. Lethargy was not found in BRD calves. When contrasted to healthy calves, BRD calves had atypical lung sounds (snoring / crackle / whistle), purulent / mucopurulent nasal discharge ( $P=0.002$ ), body temperature  $>39.5^{\circ}\text{C}$ , and the respiratory rate is  $>40$  breaths/min. (*Gaeta et al. 2018*).

### *Pasteurella multocida* in Arabian Oryx

Consistent with table (1) and Table (2) which showing the *Pasteurella*

*multocida* prevalence in Arabian Oryx after microbiological evaluation based on animals showing positive signs according to sex and age, correspondingly, nearly similar results reported by *Greth et al. (1992)* Out of 239 sera (7.17 percent) from 128 Arabian oryx (*Oryx leucoryx*) taken from 7 locations, they found 3 positive samples for *P. multocida* type B and type D, (Riyadh, Taif, and Mahazat as Stated, Saudi Arabia; Shaumari, Jordan; San Diego, United States of America [USA]; Bahrain and Qatar).

On the other hand, *Bote et al. (2017)* had reported lower results of *P. multocida* prevalence was 13 (3.39%) out of 384 samples tested from cattle suffered from hemorrhagic septicemia in Bambasi and Assosa districts, Benishangul Gumuz Regional state, Ethiopia.

The *Pasteurella multocida* in Arabian Oryx frequency distribution after microbiological evaluation based on animals represented with positive respiratory signs shows the same findings were achieved by *Marru et al. (2013)*. These data are resembled in Table (3) and table (4) correspondingly.

The reaction of *P. Multocida* on sheep-blood agar as non-hemolytic colonies same observation was reported by *Bote et al. (2017)* with the goals of isolating *Pasteurella multocida* from sick calves with Hemorrhagic Septicemia,



identifying it, and determining its antibiotic susceptibility profile.

Furthermore, biochemical tests for *P. multocida* isolated from Arabian Oryx has been done. The biochemical tests results are oxidase, Catalase, Indole, Glucose, and sucrose positive, while Triple-sugar-iron agar, Motility, Maltose are negative.

#### **Salmonella in Arabian Oryx**

Table (6) demonstrates the Prevalence percentage of the Digestive tract sings in Arabian Oryx, the same results were reported with *Anwarullah et al., (2014)*, while more percentage were reported on adults (older than one year) with *Borrielo et al., (2012)*. The fever often subsides precipitously with the beginning of Diarrhea, which may vary from watery greenish brown to field watery Diarrhea.

*S. Typhimurium* is commonly related along with outbreaks of enteritis in calves <2 months old, whereas *S. Dublin* has been associated with the same condition in adult cattle and older calves. In lambs and calves, *S. Dublin* is usually endemic on a certain farm, whereas *S. Typhimurium* is commonly associated with introduction of calves from infected farms and may cause sporadic unpredictable outbreaks. (*Grunberg, 2020*).

In contrast to Arabian Oryx, enteritis with septicemia is the mainly popular syndrome in newborn lambs, calves, foals,

piglets and birds, with outbreaks occurring in pigs as young as 6 months. When enteritis causes systemic disease due to shortage of immunity, the sickness can be severe, with fever (40.5°–41.5°C), depression and death within 24–48 hours. Pigs and calves may show neurological symptoms and pneumonia. Depending on the genetic background and strain virulence of the host, mortality can exceed 100%. (*Grunberg, 2020*).

The frequency distribution of clinical GIT symptoms documented in Arabian Oryx according to sex and age (tables 8 and 9) reveals the same results in this investigation. In a group outbreak, many hours may pass before the onset of diarrhoea, during which time the fever might resolve. Fibrinous, Mucus casts, strips of mucous membrane, and, in certain cases, blood can be seen in the feces, which have a fetid odour and include fibrinous, mucus casts, shreds of mucous membrane, and in certain cases, Rectal examination induces tenesmus and considerable discomfort. Dairy cows' milk production typically drops dramatically. In horses, abdominal pain is frequent and can be severe (colic). Mortality varies, but according to strain virulence, it can exceed 100%. (*Grunberg, 2020*).

Surface water use, rough-surfaced floors, the use of hoppers as feeders, and staff's walking boots remained all risk factors for *Salmonella* spp. Bacteria had been detected in the environmental and

animals' samples collected from the study sites. The obtained serological reaction revealed animal exposure and/or contact with the pathogen. Antibiotic use, which was supplemented in the food, may have produced an increase in *Salmonella* spp. antimicrobial resistance, as did management methods and infrastructure. (*Giraldo-Cardona et al., 2019*)

Table (12) stands for the frequency distribution of *Salmonella* spp. in Arabian Oryx after microbiological evaluation to the 30 examined samples, related to the sex, 9 gave the positive results including 6 females and 3 males. Regarding to age, which illustrated in Table (13), 8 of them are within 1 year and only one is older.

In the current study, for identification of *Salmonella* spp., biochemical tests are done for *Salmonella* spp. isolated from Arabian Oryx. *Salmonella* spp. is a Lactose and Glucose fermentative, motile, Hydrogen sulphide producing bacteria, and gave positive results for Lysine decarboxylation and Methyl red. On the other, Indole test, Voges – proskauer, Urease test and Oxidase test gave a negative reaction. While total populace of Arabian Oryx in Bahrain, only 9 animals identified with *Salmonella* spp. infection, they are 2.8% *S. dublin*, 2.8% *S. enteritidis* and 0.7% *S. typhimurium*. Nearly similar results reported by *Hadimli et al. (2017)*.

This study concluded that, although the Arabian Oryx is adapted to the weather in Bahrain, but they are still susceptible to diseases, especially in the young ages (younger than 1 year old). Most of these diseases are affecting the respiratory and digestive systems. The hazard in the wild animals that it is not easy to detect the diseased animal at early stages as they are in free ranged areas. The most common diseases related to the Arabian Oryx are: Pasteurellosis and Salmonellosis.

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### ملخص العربي

#### إصابات الباستريلا مالتوسيدا والسلامونيلا في المها العربي

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تم فحص ظاهرياً عدد 143 حيوان من فصيلة المها العربي في ثلاث مناطق لانتشار الحيوان بمملكة البحرين بالترتيب: محمية العرين للحياة البرية، ومنتزه العرين للحياة البرية / مركز الزوار، وجزيرة حوار حيث يتواجد حيوان المها العربي بنسبة 70% و 13.9% و 16.1% على التوالي. ويمثل ذكور تلك الحيوانات نسبة 29.4% بينما تمثل الإناث نسبة 70.6% من إجمالي عدد الحيوانات. ووضعت نتائج الفحص البيطري أن الحيوانات إيجابية النتائج للاعراض التنفسية هي عدد 25 حيوان، بينما الاعراض السلبية لذات الإصابة هو عدد 118 حيوان. ووزعت تلك الأعراض التنفسية الإيجابية بين عدد 18 أنثى و 7 ذكور، بينما أظهرت النتائج الإيجابية للاعراض التنفسية من ناحية العمر أن عدد 19 حيواناً عمر أقل من عام وعدد 6 حيوانات كان عمرها أكبر من سنة واحدة. وأكد الفحص البكتريولوجي للعينات المسحوبة من الحيوانات المصابة أن عدد 4 حيوانات كانت إيجابية لجرثومة الباستريلا مالتوسيدا، منهم عدد 3 من جنس الإناث وذكر واحد وجميعهم كانت أعمارهم أقل من سنة واحدة. كما وجد أن توزيع الاصابات بجرثومة الباستريلا مالتوسيدا في المها العربي وفقاً لمناطق الحياة البرية في مملكة البحرين عدد 2 من الإناث وذكر في محمية العرين للحياة البرية، و عدد واحد أنثى في جزيرة حوار ولا توجد أي إصابة لجرثومة الباستريلا مالتوسيدا في المها العربي في محمية العرين - مركز الزوار. ومن الناحية الأخرى تبين من النتائج أن أعراض الجهاز الهضمي في المها العربي توزعت بنسبة 14.7% للإناث ونسبة 6.3% ذكور. بينما وفقاً للعمر كان نسبة 9.1% من الحيوانات ذات الأعراض الإيجابية للجهاز الهضمي أصغر من عام، بينما كان نسبة 11.9% أكبر من عام. وبلغت نسبة الإصابة في الإناث من محمية العرين 4% بينما يمثل الذكور 1% فقط، وكانت في محمية العرين - مركز الزوار 5% للإناث و 10% للذكور. بينما كانت في جزيرة حوار عدد أنثى واحدة فقط مصابة بجرثومة السلمونيلا. وخلصت النتائج التي تم الحصول عليها إلى أن حيوان المها العربي يتكيف مع طقس مملكة البحرين ولكنه لا يزال عرضة للأمراض التنفسية والهضمية خاصة في الأعمار الصغيرة. لذلك يجب بذل المزيد من الجهود من السلطة المختصة للسيطرة على عدوى الباستريلا مالتوسيدا والسلامونيلا في المها العربي بمملكة البحرين.