Seroprevalence of schmallenberg virus infection as emerging disease in cattle in Iraq

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Abstract

Schmallenberg virus is an emergent disease which infect cattle, sheep and goats which cause loss of condition, diarrhea, and abortion in pregnant animals, so this study was conducted to detect the antibodies in imported calves by using cELISA, so 400 blood samples was collected calves in different ages and healthy status in a period between October 2018 to September 2019 in Nineveh province, the investigation of specific antibodies was done by competitive Enzyme Linked Immuno Sorbent Assay, the results showed that the prevalence of Schmallenberg virus in imported calves was 21% (84 positive from 400 samples), high prevalence of infection in the animals more than 6 months to 1 year old 11.5% when compare with animals less than 6 months of age 9.5%, high incidence of infection in animals suffer from various clinical signs 17% and the healthy apparent animals recorded low prevalence of infection 4%, samples which collected in spring months recorded high prevalence of infection 7.5% while the lowest prevalence of infection with the virus recorded in winter months 2%, with significant variance in spring and summer months compared with other seasons, in conclusion this study was conducted that schmallenberg virus is newly emerge in Iraq and this study is firstly recorded this virus in cattle in Iraq.

Introduction

Schmallenberg virus (SBV) is an enveloped segmented, single-stranded RNA virus, negative sense, classified to the Orthobunyavirus genus, Bunyaviridae family and it is related to Akabane, Ainoa and Shamonda viruses; which are all constitute Simbu-Sero group (1). The virus was first recorded in the German town of Schmallenberg at 2011, in cows suffer from increase body temperature, diarrhea, and decrease milk production, then in 2012 abortions and malformation in calves, kids, lambs in other European countries (2). The virus can be transmitted via arthropods (Biting midges and Culicoides, mosquitoes) that ingest the virus with infected animals blood and replicate in the arthropod’s tissues then transmitted by bite to other healthy animals by biting midges (3), the virus reported in domestic animals like ovine, caprine, bovine, equine and camilida and reported in other wild animals like zebra, bison and deer (4). The initial clinical signs in animals are diarrhea, decrease in milk production, loss of appetite, increase body temperature and abortions, congenital malformations, stillbirths as the most common and vertebral, limb deformities in the aborted, stillborn or neonatal SBV-positive calves (5). ‘Dummy syndrome’ is a behavior abnormality occurring in SBV calves, followed by some clinical signs including walking continuously, avoiding food or drink, pressing head against solid objects, inability to retreat and no response any stimuli (6). Schmallenberg virus can be isolated on several types of cell culture including baby hamster kidney cells, Vero cells with a cytopathic effect (7). RT-qPCR have been developed to discovered SBV, detection of S segment is mostly used PCR assays which showed that this method is more sensitive,
SBV RNA has been found at two-week intervals in blood samples of infected calves so the viral RNA can be used to investigate in blood samples (8). Enzyme Linked Immuno Sorbent Assay is a specific and sensitive method than other serological tests, testing for a larger number of samples in the same time (9). The indirect ELISA test is depending on SBV recombinant nucleoprotein antigen or whole SBV virus antigen, other types of ELISA which can be available commercially including competitive ELISA (10). Other character of ELISA technique that the can be applied to serum and milk samples which can be collected sporadically or bulk milk in dairy cattle farm (11). In cattle SBV was reported as emergent disease in neighboring countries including Turkey (12), Iran (13), Saudi Arabia (14), Jordan (15), in Iraq there is one study reported this virus in aborted ewes in Erbil (16). So the aim of study is the investigation the viral antibodies in imported calves in Iraq.

Materials and methods

Study animals
A total of 400 imported calves, as soon as they arrive in Iraq, the age of calves ranged from 6 months -1 year, some animals was recorded a history of loss appetite, diarrhea, fever, while another is healthy appearance, the collection of samples was collected from in animals in a period between October 2018 to September 2019 in Nineveh province.

Blood samples and laboratory testing
Blood were collected from jugular vein under aseptic conditions, the blood transferred to plain tubes, then separated the serum from clot by centrifuge at 3000 rpm for 10 minutes, then the serum was transferred to ependrof tubes and stored in -20°C until used (17).

Competitive Enzyme Linked Immuno Sorbent Assay (c-ELISA)
was used to detect specific antibodies against SBV (ID Vet France: SBVC-5P), the plate of this test coated with recombinant SBV nucleoprotein antigen and the conjugate consist of Anti-SBV nucleoprotein-HRP conjugate. The test was done according to manufacture manual of ID Vet.

Statistical analysis
The variations in the prevalence of Schmallenberg virus between the ages of animals and the animal’s status were evaluated by employing two-sided Chi-square and Fischer’s exact test in IBM-SPSS statistics version 19 program (18).

Results
The study revealed the prevalence of Schmallenberg virus in imported calves was 21% (84 positive from 400 samples). High prevalence of infection in the calves more than 6 months to 1 year old 11.5% when compare with animals less than 6 months of age 9.5% (Table 1). The relationship between the prevalence of infection with Schmallenberg virus and the health status of calves reveal high incidence of infection in animals suffer from various clinical signs (diarrhea, depression, fever) 17% and the healthy apparent animals recorded low prevalence of infection 4% (Table 2). The result showed high prevalence of infection with Schmallenberg virus in samples which collected in spring months 7.5% while the lowest prevalence of infection with the virus recorded in winter months 2%, with significant variance in spring and summer months compared with other seasons (Table 3).

Table 1: Prevalence of infection with Schmallenberg virus according to age of the animals using c-ELISA

| Age          | n blood samples | n+ samples (%) |
|--------------|-----------------|----------------|
| >6 months    | 176             | 38 (9.5)a      |
| <6 -12 months| 224             | 46 (11.5)a     |
| Total        | 400             | 84 (21)        |
| Values significantly different (P < 0.05) between different ages are labelled with the different superscript letters (a, b or c).

Table 2: Prevalence of infection with Schmallenberg virus by using c-ELISA according to health status of the animals

| Health status | n blood samples | n+ samples (%) |
|---------------|-----------------|----------------|
| Clinical Signs| 148             | 68 (17)a       |
| Healthy       | 252             | 16 (4)a        |
| Total         | 400             | 84 (21)        |
| Values significantly different (P<0.05) between health status are labelled with the different superscript letters (a, b or c).

Table 3: Prevalence of infection with Schmallenberg virus by using c-ELISA according to season of samples collection

| Season | n blood samples | n+ samples (%) |
|--------|-----------------|----------------|
| Winter | 112             | 8 (2)a         |
| Autumn | 82              | 14 (3.5)a      |
| Spring | 96              | 30 (7.5)b,a    |
| Summer | 110             | 28 (7)c,b      |
| Total  | 400             | 84 (21)        |
| Values significantly different (P < 0.05) between seasons are labelled with the different superscript letters (a, b or c).

Discussion
Schmallenberg virus (SBV) is a emerge viral disease in ruminants worldwide and Transmitted through the arthropods. it Classified to the Orthobunyavirus genus, family Bunyaviridae, infected animal suffered Increase body temperature, loss of appetite, drop of milk, diarrhea and loss of body conditions (19). In this study the prevalence of schmallenberg virus antibodies in imported calves using...
ELISA showed that total percentage of infection was 21% (84 from 400 samples), many serological studies recorded the schmallenberg virus antibodies in cattle in different countries with differences in percentage of infection (20) recorded 56.6% of cattle was infected with virus in Ethiopia using cELISA, while (11) investigate the antibodies of the virus in Netherland using virus neutralization test and the result showed the 72.5% of cattle was infected. other study (21) included seroprevalence for Schmallenberg virus in Mozambique in ovine, bovine and caprine sera using c SBV ELISA, and results showed 90% of cattle was infected with SBV, (7) found 32.2% positive results for anti-SBV antibodies in 1101 cattle serum samples and tested by virus neutralization test (VNT) in Portugal 7.92% was recorded in Russia (22), in Jordan (15) 57 from 115 serum samples positive give results, Azkur et al. (12) recorded 24.5% of Turkish cattle was infected with SBV using ELISA.

The differences between the distributions of the disease between the countries explains to several reasons which included: the differences in distributions and infestation of arthropods vectors, the variations between the farm management system and quarantine measures, the history of the disease in every country and the source of the animals (4). This study recorded high prevalence of infection in the calves more than 6 months to 1 year old compared with animals less than 6 months of age without significant differences. Armin et al. (11) founds no significant differences in age, which referred to that the SBV is emerge comes from other areas. High prevalence in older animals than young once because the younger animals is housed indoor breeding and this decrease exposure to arthropod vectors, other reasons of increase of infection in older animals that the increase of maternal antibodies in older animals with elevated in younger animals than older once. The results showed high prevalence of infection in animals suffer from various clinical signs (diarrhea, depression, fever) when compare with healthy apparent without statics differences (23) said that the infection is non-specific and symptoms is fever, severe diarrhea, and sometimes nervous signs, while (10) recorded inconspicuous clinical signs with short durations due to short-duration SBV viremia.

The results showed that the animals suffer from diarrhea, depression, fever recorded high prevalence compare with healthy once, several studies included the clinical signs in cattle which infected with SBV (19) how said that there is mild clinical signs such as reduced milk yield or increase temperature but abortion is considered while (2) recorded mild increase of body temperature, diarrhea and a decrease in milk production, other studies (20) showed that the SBV cause Reproductive disorders including abortion, and metritis, other researchers indicate that calves which SBV positive were arthrogyrosis, brachynathia inferior, torticollis, kyphosis, lordosis, scoliosis, and muscle hypoplasia (24).

The result showed high prevalence of infection in spring months and decreased in winter months, with significant variance in spring and summer months compared with other seasons. In summer 2012 acute infection of SBV occurred in Germany (25) then the virus spread specially in vector season (26). This scenario may repeated when vector is spread in some seasons and the infection is happened. Depending on status immune system, the disease will appear if immune suppress of the animal (27) The increase of temperatures supplied better climate of vector-borne disease spread compared with standard temperatures. Vector-borne disease transmission models for Schmallenberg virus commonly use mathematical (28,29), during vector transmission season of 2012 in Denmark the Schmallenberg virus was spread (30), the virus was spread in Sweden in 2012 through the investigation of antibodies in serum and bulk milk during season of vector spread, other study of spread the virus in vector season in Denmark was attempt (31). Kameke et al. (32) showed that the first arthropods find inside cattle stables which relates to appear the infection with SBV in Germany, While Martinelle et al. (33) said that the arthropods activity is evenly distributed over the seasons of the animals, actual transmission seasonality of arthropods liberalty (34).

Conclusions

This study was conducted that schmallenberg virus is newly emerge in Iraq and this study is firstly recorded this virus in cattle in Iraq

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Conflict of interest

Author declare no conflict of interests of the manuscript.

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21. Schmallenberg

القصص المصممة للإصابة بفيروس كأحد الأمراض الطارة في المناطق العربية

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الخلاصة

إن الإصابة بفيروس شمألنبرغ هو أحد الأمراض الطارة بحسب الأدلة والأعراض والطريقة في التعامل والإفرازات في الإدانة للحوم. هذه الأدلة ماركة تطبيقات تعلم عن الفيروس. التأثيرات في الحيوانات السائدة باستخدام اختبارات المزمن المناعي المرتبط بالفيروسات في الفترة بين أكتوبر 2016 إلى سبتمبر 2019 في محافظة تيزي. تم القصص عن الأسماك المضادة المختصة باستخدام

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اختبار الممتز المناعي المرتبط بالإنزيم التنافسي، أظهرت النتائج أن نسبة الإصابة بالفايروس في العجول المستوردة كانت 21% (84 إيجابية من 400 عينة)، ارتفعت نسبة الإصابة في الحيوانات التي تبلغ أعمارها 6 أشهر إلى سنة واحدة 11.5% عند مقارنتها بالحيوانات أقل من 1 شهر من العمر 9.5%. وفي الحيوانات التي تعاني من علامات سريرية مختلفة 17% في حين كانت منخفضة في الحيوانات السليمة.4%، سجلت

العينات التي تم جمعها في أشهر الربيع معدل انتشار مرتفع للإصابة 17.5%، بينما سجلت أقل نسبة إصابة بالفايروس في أشهر الشتاء 2%. مع تباين معيّن في فصل الربيع والصيف مقارنة مع المواسم الأخرى، بينت هذه الدراسة أن فيروس شمالنبرغ هو أحد الأمراض الطارئة الجديدة في العراق، وهذة الدراسة سجلت هذا الفيروس في الماشية لأول مرة في العراق.