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Evaluation of the Immunological Response of Local and Imported Vaccines of Lumpy Skin Disease Virus in Cattle as Preventive Step

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Abstract

VACCINATION process against lumpy skin disease (LSD) is considered the only effective method for the prevention and control of LSD in endemic areas, where neutralizing antibodies play an important role in long-term protection after vaccination. A total of 260 sera samples were collected from 20 bulls, their ages ranged from 9 to 12 months. These bulls were divided into groups (each group containing 10 bulls) in beef farms at Monshaٔt El kanater Aboghaleb, Giza Governorate, Egypt. This study was carried out from November 2022 to May 2023. One of these groups was vaccinated with the local strain (SERVAC CAPRI-C®) and the other was immunised with the imported strain (BOVIVAX LSD®). The serum sampling was carried out before vaccination and till 6 months with intervals 2 weeks after vaccination. Our study aimed to determine the efficacy of both vaccines in cattle immunity for evaluating vaccinated cattle's humeral immune response against the Neethling strain vaccine. Our results revealed that the titer of antibodies against BOVIVAX LSD® appeared to highly significantly increase (P>0.05) throughout the experiment when compared with (SERVAC CAPRI-C®) which showed a slight numerical increase and then significantly decreased at 24 weeks post-vaccination. Likewise, the vaccination process might slightly affect some parameters of liver dysfunctions mainly the alanine transaminase test. In conclusion, live attenuated BOVIVAX LSD® vaccine has a great ability to induce a high level of antibody titer with prolongation of immunity duration, so it is considered the best choice of vaccine for prevention and control of LSD that can applied in the field.

Keywords: Lumpy skin disease, Neethling strain, Vaccine, Immunity, Prevention.

Introduction

Lumpy skin disease (LSD) is an arthropod-born viral disease that causes high economic impacts and it is listed as one of the most notifiable diseases that affects cattle and buffaloes [1]. LSD is caused by poxviruses that belong to the genus of Capripoxvirus in the subfamily Chordopoxvirinae within the family Poxviridae [2].

In Africa, LSD first appeared in Zambia in 1929. This disease recently appeared in other African areas and the Middle East countries [3]. In Egypt, The first infected cases were reported in and around Ismailia and Suez governorates in 1988 [4]. Because of the importation of infected cases from Ethiopia to the private quarantine of the Ismailya governorate [5].The LSD virus is transmitted mechanically through the female mosquitoes (Aedes aegypti) and stable flies (*Stomoxys calcitrans*) from the infected to susceptible animals [6] and potentially transmitted through the ticks [7].

The Infected cattle suffered from fever, nasal discharge, hypersalivation and lacrimation, followed by characteristic skin eruptions that may involve the other parts of the body. The nodules were well round, firm, circumscribed painful slightly raised, also may involve the internal organs. The skin nodules contain a firm, yellow or creamy-gray mass of tissue. Regional lymph nodes were swollen, and the edema developed in the legs, udder and brisket. The incubation period of LSD was 4–14 days [8].

The disease was associated with high morbidity and low mortality, it causes high economic losses due to its severity and the cost of treatment whereas, decreased milk production, feed intake and weight conversion rate, also

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caused infertility, abortion and damage to the hides of infected cattle [9].

Vaccination was the only method for preventing the spread of the disease in the endemic and newly affected areas, so the selection of the most effective vaccine was the major challenge for veterinary authorities and farmers in case of outbreaks [10].

LSDV was closely related to both viruses within the Capri-poxvirus genus (sheep pox virus and goat pox virus) [11]. Likewise, LSDV, sheep pox and goat pox virus had appeared to be antigenically indistinguishable, so vaccination of cattle with a vaccine against one of the Capri-poxvirus strains protects the vaccinated cattle against the other three strains [12].

Live attenuated vaccines were the most commercially available vaccines against LSD, based on a sheep pox virus (SPPV) or goat pox virus (GTPV) and LSDV strain, however the first inactivated vaccines had recently appeared in the markets [10].

The vaccines against LSDV depend on the dosage, volume, the titer of vaccine, seed virus and the attenuation levels in the final products, so the quality of vaccine products is major important for farmers and cattle welfare [10].

Evaluation of the immune responses against LSDV vaccines in the field, the trial was highly important for the assessment of the status of the available vaccine strains for the selection of the best vaccine strain that is highly effective for protecting cattle against LSDV [13].

LSD vaccines must be very safe for use in all ages, all breeds and bovine species and both sexes [14]. No challenging experiments of the LSD vaccine had been carried out in the domestic buffaloes, but the vaccination protocol had been recommended for cattle [15]. Newly purchased cattle intended to be moved, must be vaccinated at least 28 days before transportation [10].

Vaccination of cattle leads to disturbance of the liver function, which is observed in an increase the limit of cholesterol, bilirubin and ALP activity so, the use of immune stimulant which has a hepatoprotective effect is very important due to vitamin E content that can reduce the toxic effects of biological products on the liver cells [16]. Also, AST increased after vaccination of cattle [17].

The objective of this study is the compare local (SERVAC CAPRI-C®) and imported vaccines (BOVIVAX LSD®) and to determine the efficacy of both vaccines in cattle immunity for evaluation of the humoral immune response of vaccinated cattle against the Neethling strain vaccine for prevention and control for LSDV.

Material and Methods

Study population

The study was conducted on a beef cattle farm at Monshaٔt El kanater Aboghaleb, Giza Governorate, Egypt. This study was carried out from November 2022 to May 2023. The selected cattle were 20 cross-breed, males, their ages ranged from 9 to 12 months and their weight ranged from 80 to 90 kg. Bulls raised in a semi-intensive shed with a sandwich panel roof. This farm depends on natural ventilation without a cooling system. The feeding system was open, bulls were given a total mixed ration (forage and concentrated food). Furthermore, free access to fresh and clean water. These bulls were divided into two groups (each group containing 10 bulls) for the application of used vaccines and monitoring of antibody titers in vaccinated cattle.

Ethical approval

This study was approved by the Institutional Animal Care and Use Committee of the Faculty of Veterinary Medicine, Benha University, Qalubia, Egypt (Ethical approval number: BUFVTM 17- 06-23).

Vaccination

This study was applied to the two groups of bulls. One of these groups was vaccinated subcutaneously in the neck of animals with 2 ml of local live Neethling LSD strain vaccine (SERVAC CAPRI-C®) also, the other was vaccinated subcutaneously in the neck of animals with 2 ml of imported live Neething LSD strain (BOVIVAX LSD®).

Vaccines

SERVAC CAPRI-C®: obtained from Pox Department, Veterinary Serum and Vaccine Research Institute, Abbasia, Cairo, Egypt.

BOVIVAX LSD®: obtained from an international commercial company for free exchange, Morocco.

Sampling

A total of 260 sera samples were collected from vaccinated beef animals with different vaccines. The serum samples were collected from each group of these animals at zero days and after vaccination at 2-week intervals till 6 months after the vaccination process. The samples were collected at zero days before vaccination and 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, and 24 weeks post-vaccination.

The collected blood samples without anticoagulant from each group of vaccinated animals were allowed to be clot at room temperature for 3 hrs and then the serum was extracted by centrifugation at 2000 RPM for 20min

and aliquot in 1.5 ml centrifuging tubes. All serum samples were stored until examination at - 20 °C.

Serum analysis

Serological analysis

Evaluation and monitoring of LSD antibody titers after administration of used vaccines

The collected sera samples containing LSDV antibodies were tested against LSDV antigen by using the indirect ELISA technique for titration the titers of LSD antibody released after administration of used vaccines and assessment the humeral immune response against these vaccines consequently, evaluation the efficiency of both vaccines in vaccinated cattle.

Enzyme linked immunosorbent assay (Indirect ELISA)

Evaluation of LSDV antibodies in the collected sera samples was tested by using the indirect ELISA technique as demonstrated by [4]. Briefly, 100 µl of purified antigen of LSDV that prepared according to [18] was diluted 1/100 in carbonate-bicarbonate buffer (pH 9.6) and incubated in the plates at 4 ºC overnight and then washed with the washing buffer (PBS, pH 7.4 containing 0.05% tween 20), then removed the washing solution and the plates were dried on filter paper. The dried plates were blocked with 100 µl of blocking buffer (1% bovine serum albumin in PBS containing 0.05% tween 20) for one hour at 37 ºC. Then, 100 µl of 1/100 diluted sera samples were added into the coated plates. The control positive and negative sera were included in the test. The plates were incubated for one hour at 37 ºC then washed three times. The anti-bovine IgG (whole molecule) horse radish peroxidase conjugate, Sigma chemical company (100 µl/ well) at a dilution of 1/5000 was added for one hour at 37 ºC then washed three times. Finally, 100 µl of substrate (o.Phenylene diamine), Sigma chemical company was added to each well and the plate was incubated at room temperature in a dark place for 10 minutes, the reaction was stopped by the stopping solution (sulphuric acid 97%). The absorbance of these contents of each well was read at 450 nm by using ELISA reader.

Serum biochemical analysis

Aspartate transaminase test (AST), alkaline phosphatase (ALP) and total bilirubin analyses were carried out using JENWAY 6051 Colorimeter U.K device with (Spectrum GmbH company kits; CAT. NO. (260 001; 216 001; 222 001) according to methods formerly [19, 20, 21] respectively.

Alanine transaminase test (ALT) and Cholesterol analyses were carried out using Chem7, USA device with kinetic kits (Centronic GmbH company kits; CAT. NO. (GF10000050; MG230 001) according to methods formerly [22, 23] respectively.

Statistical analysis

The statistical analysis was carried out according to [24] using Two-way ANOVA using SPSS, ver. 27 (IBM Corp. Released 2013). Data were treated as a complete randomization design. Multiple comparisons were carried out applying Duncun test. The significance level was set at < 0.05.

Results

The antibody titers of vaccinated cattle with local and imported LSD vaccines were mentioned in (Table 1 and Fig. 1). The results revealed that there is a highly significant difference between antibodies titer against the local (SERVAC CAPRI-C®) and imported (BOVIVAX LSD®) vaccines.

The antibodies titer against local SERVAC CAPRI-C® vaccine showed non significance differences during the period of experiment till 24th weeks after vaccination, while it started to numerically increase at 2nd week after vaccination (0.415±0.081) then reach to peak at 12th week (0.440±0.052) then start to significantly decline to 24th weeks after vaccination (0.264±0.021).

The antibodies titer against imported BOVIVAX LSD® vaccine showed significance differences all over the period of experiment from week to another. Likewise, it achieved to increase at 2nd week after vaccination (0.601±0.071) then gradually increased till the 16th week (0.872±0.059), followed by sharp significant increase in antibodies titer to 24th weeks after vaccination (1.281±0.102).

The serum biochemical analysis of vaccinated cattle with the local and imported LSD vaccine were mentioned in (Table 2).Vaccination process might be slightly affect some parameters of liver dysfunctions (alanine transaminase test (ALT), aspartate transaminase test (AST), alkaline phosphatase (ALP), cholesterol and total bilirubin) in vaccinated cattle. Our results, depicted in (Table 2), demonstrate a significant variation of these parameters across the period of experiment in local (SERVAC CAPRI-C®) and imported (BOVIVAX LSD®) vaccines.

The results revealed that type of vaccine (local and imported) had no significant difference on the ALT level. While the time of vaccination had gradual significant increase in ALT level began with (27.20±0.58) and (23.57±0.50) for local (SERVAC CAPRI-C®) and imported (BOVIVAX LSD®) vaccines at 2nd week after vaccination, respectively till (30.42±4.46) and (34.45±1.11) at 24th weeks after vaccination, respectively as showed in (Fig. 2).

The level of AST significantly affected by type vaccine, but it also showed not significant differences after vaccination at 2th week (19.67±2.03) and (13.00±1.73) till the 24th weeks after vaccination (27.00±2.31) and (52.00±1.15) for local and imported vaccines, respectively as showed in (Fig. 3).

The levels of ALP and total bilirubin are significantly higher in cattle vaccinated with local than cattle vaccinated by imported vaccines. The level of ALP showed (37.15±0.20 and 28.70±3.46) and (42.55±1.65 and 46.95±0.32) at 2th and 24th weeks post-vaccination for local and imported vaccine, respectively. While Total bilirubin showed (0.86±0.13) and (0.54±0.06) for local (SERVAC CAPRI-C®) and imported (BOVIVAX LSD®) vaccines, respectively at 2th week and (1.13±0.03) and (1.24±0.03), respectively at 24th weeks post-vaccination throughout the period of experiment showed in (Figs. 4, 5).

Cholesterol level significantly affected by type vaccine, they also showed significantly increases from 2th week (110.32±12.01) and (273.75±40.15) till the 24th weeks after vaccination (172.50±19.80) and (800.69±45.94) for local and imported vaccines, respectively as showed in (Fig. 6).

Discussion

Vaccination is very important to know that LSD prevention is most beneficial than its treatment to avoid high economic losses due to the cost of treatment, loss of milk due to mastitis, skin damage and loss of animal products due to deaths, fever, abortion and myiasis. So from this point of view, selection of the best effective vaccine was the major challenge in case of the outbreak, so in this study the efficacy of local and imported homologous live attenuated Neethling strain vaccines were compared and evaluated.

The recorded results about the local vaccine revealed that the first numerical increasing in antibody titers appeared at 2ed week. In this concern, The protective level of antibody titers against local vaccine of LSD (SERVAC CAPRI-C®) started from (0.415) at 2th week after vaccination and the protective duration continued for only 4 months after vaccination then gradually declined after this period. This result agreed with Klement et al. [25] reported that protective immunity developed throughout 14 days after vaccination. Also, Gari et al. [26] who reported that antibodies decreased after 63 days following annual vaccination so, duration of detectable humeral immune response could be lower than a year and Moje [27] reported that a fewer number of collected sera samples showed the presence of specific antibodies against LSD both pre-and post-vaccination.

Some vaccinated animals did not show a measurable amount of protective antibodies [28]. This may attributed to the fact that the magnitude and timing of immune response against LSDV vary from one animal to another animal [29] or due to over-attenuation of the Neethling strain during manufacturing of local vaccine or some other defects in vaccine products that causes poor immunogenicity after vaccination. Moreover, this may be due to failure in the vaccination process or inadequate doses of vaccine administration. The immune response of LSDV vaccines is variable so, the antibodies can be undetectable subsequently following vaccine administration. This may be attributed to the type of vaccine used or the nature of some cattle breeds [26].

In contrast, the protective level of antibodies titer against the imported vaccine (BOVIVAX ®) started from (0.601) in the 2nd week and then gradually increased till the 16th week (0.872), followed by a sharp increase at 24th weeks after vaccination (1.281). This means that the protective duration continued along the period of experiment till 6 months in compared with local vaccine. These results were not consistent with Samojlovic et al. [30] who recorded that the antibody titer was detected after 21 days post-vaccination, while the highest level of neutralizing antibodies was determined 35 days after vaccination and Kumar et al. [28] revealed that all the 10 vaccinated animals, except one animal, developed antibodies against LSDV at the thirty day post vaccination. Also disagreed with El-gbily et al. [31] demonstrated that antibody titers were significantly increased post-vaccination from week to week till the tenth week and became highly protective against LSD from after the third week after vaccination.

Likewise, our results agreed with Klement et al. [25] mentioned that the protective immunity of homologous LSD live attenuated vaccine in vaccinated cattle in Albania gradually increased till 2ed week post-vaccination by a level (of 0.6) then fluctuated till the 16th week reached (0.68) and subsequently occurred sharp increase at the 18th week became (1) while, disagreed with Norian et al. [32] said that vaccinated calves reached the protective level (1.5) at 3rd week and increased gradually to (2) after 4th week from vaccination and Bamouh et al. [33] revealed that the average of neutralizing antibodies titer recorded in 4th-week post-vaccination was (1.7) at a high dose and (1.2) at a low dose for the Neethling strain vaccine.

In our turn, the antibody titers of the BOVIVAX ® vaccine were evaluated and showed that the antibody titers were significantly increased all over the 24-week experiment period from one week to another.

This finding agrees with Milovanović et al. [34] reported that the specific antibodies of Capripoxvirus were detected after vaccination by 46 to 47 weeks by using ELISA showed that the humeral immune response may be lasts for more longer than 7 months and Suwankitwat et al. [35] reported that antibody response was appeared up to 15 months with higher levels of protective antibodies, also Haegeman et al. [36] revealed that Lumpyvax vaccine gave high protection for a period reached up to 1.5 year with little side-effects.

This higher value of antibody titer can be attributed to the better humeral immune responses of vaccinated cattle against the BOVIVAX LSD® vaccine.so, vaccination by BOVIVAX LSD® was shown as the most efficient measure to control LSD and avoid spreading LSD in the endemic areas.

The vaccination process might be slightly affecting some parameters of liver dysfunctions. Our results revealed that ALT and Cholesterol have a gradual significant increase after vaccination by local (SERVAC CAPRI-C®) and imported (BOVIVAX LSD®)vaccines while, AST, ALP and total bilirubin levels were significantly affected by type vaccine, but they not showed significant differences after vaccination. These results came by Burkov et al. [16] revealed vaccination against LSD causes liver damage in vaccinated cattle whereas, cholesterol levels increased by (19.9%) ($p < 0.01$) in vaccinated cattle.

On the contrary, Bulatov et al. [17] observed an attenuated heterologous goat pox virus vaccine caused a slight increase in AST after 2 weeks after vaccination while, the levels of ALT and cholesterol remained within the physiological levels in vaccinated cattle and Burkov et al. [16] revealed the content of bilirubin increased by (39.3%) after vaccination. These results might be attributed to the expected reactions due to the immune system stimulation post-vaccination.

Notably, ALT, AST, ALP, cholesterol and total bilirubin are highly significantly increased at 24th weeks post vaccination in vaccinated cattle. This may attributed to subsequent infection or may be due to the climatic change and environmental stress factors because of the change of environmental temperature along the seasons during the period of experiment. In other words, the liver is an important organ responsible for the metabolism of glucose, lipids and proteins [37], all of which have been disturbed in cattle exposed to heat stress [38]. Compromised liver function is proven in heat-stressed cattle by reducing the activities of liver enzymes [39].

Conclusion

In conclusion, live attenuated Neethling BOVIVAX LSD® vaccine gives a potential immunogenic ability to induce a higher level of antibodies titer with prolongation of immunity duration, so it is considered the best choice of vaccine for prevention and control of the LSD disease that can applied in the field with the use of immune stimulants to avoid the liver damage in vaccinated cattle.

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Conflict of interest

The authors announce that they have no Conflict of interest.

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TABLE 1. The antibodies titer of vaccinated cattle with local (SERVAC CAPRI-C®) and imported (BOVIVAX LSD®) vaccines against LSD (mean ± SE).

Sampling time	Local vaccine (SERVAC CAPRI-C®)	Imported vaccine (BOVIVAX LSD®)
Zero day (control)	0.308 ± 0.033^{abB}	0.469 ± 0.041^{fA}
2nd weeks	0.415 ± 0.081^{aB}	0.601 ± 0.071^{efA}
4th weeks	0.400 ± 0.083^{abB}	0.627 ± 0.076^{eA}
6th weeks	0.410 ± 0.062^{aB}	0.604 ± 0.053^{eA}
8th weeks	0.418 ± 0.064^{aB}	0.641 ± 0.068^{eA}
10th weeks	0.391 ± 0.049^{abB}	0.706 ± 0.048^{deA}
12th weeks	0.440 ± 0.052^{aB}	0.689 ± 0.059^{deA}
14th weeks	0.437 ± 0.092^{aB}	0.798 ± 0.082^{cdA}
16th weeks	0.398 ± 0.038^{abB}	0.872 ± 0.059^{bcA}
18th weeks	0.381 ± 0.028^{abB}	1.026 ± 0.093^{bA}
20th weeks	0.314 ± 0.020^{abB}	1.012 ± 0.159^{bA}
22th weeks	0.311 ± 0.019^{abB}	1.296 ± 0.127^{aA}
24th weeks	0.264 ± 0.021^{bB}	1.281 ± 0.102^{aA}

TABLE 2. Serum biochemical analysis of vaccinated cattle with local and imported vaccines (mean±SE)

Parameter	Period (week)	Local vaccine	Imported vaccine
ALT	Zero day (control)	16.59±5.55bA	12.21±2.01cA
	2nd weeks	27.20±0.58aA	23.57±0.50bA
	12th weeks	19.20±2.02bB	25.28±3.50bA
	24th weeks	30.42±4.46aA	34.45±1.11aA
AST	Zero day (control)	19.00±3.46bA	16.00±1.73bA
	2nd weeks	19.67±2.03bA	13.00±1.73cB
	12th weeks	19.33±2.03bA	13.67±2.33bcB
	24th weeks	27.00±2.31aB	52.00±1.15aA
ALP	Zero day (control)	37.35±0.09bA	32.95±2.63bcB
	2nd weeks	37.15±0.20bA	28.70±3.46cB
	12th weeks	30.50±1.21cA	33.45±3.61bA
	24th weeks	42.55±1.65aB	46.95±0.32aA
Total bilirubin	Zero day (control)	0.97±0.04abA	0.76±0.12bB
	2nd weeks	0.86±0.13bA	0.54±0.06cB
	12th weeks	0.54±0.01cB	0.86±0.13bA
	24th weeks	1.13±0.03aA	1.24±0.03aA
Cholesterol	Zero day (control)	144.70±9.30abA	151.45±16.77dA
	2nd weeks	110.32±12.01bB	273.75±40.15cA
	12th weeks	150.15±5.92abB	476.88±16.73bA
	24th weeks	172.50±19.80aB	800.69±45.94aA

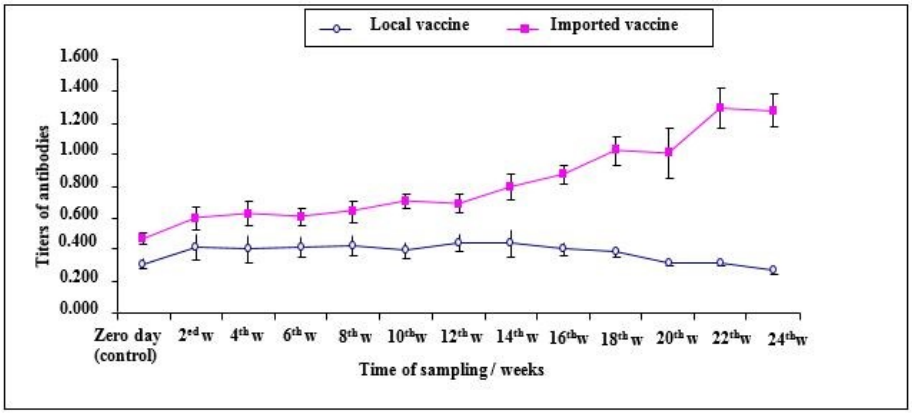

Fig.1.The antibodies titer of vaccinated cattle with Local ((SERVAC CAPRI-C®)) and imported (BOVIVAX LSD®) vaccines against LSD.

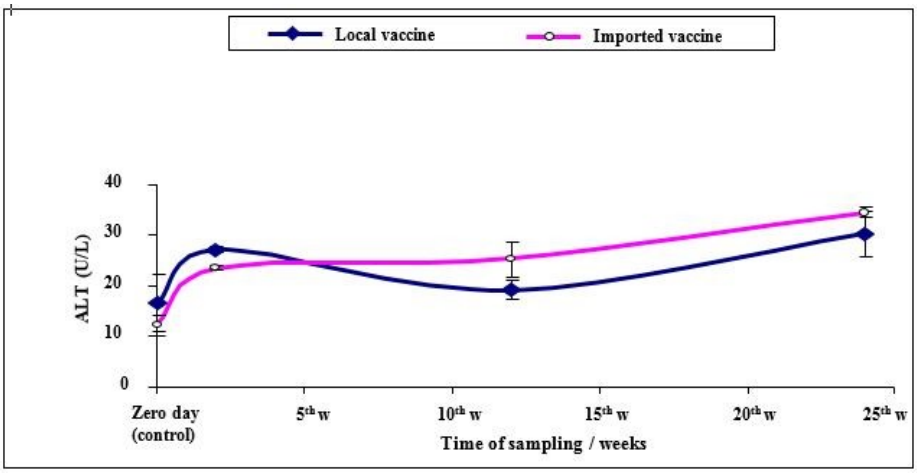

Fig.2. Levels of ALT of vaccinated cattle with Local ((SERVAC CAPRI-C®)) and imported (BOVIVAX LSD ®) vaccines against LSD.

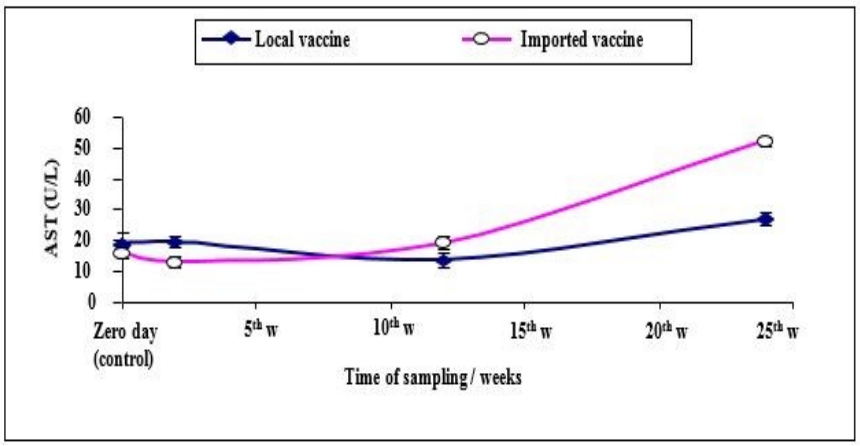

Fig.3. Levels of AST of vaccinated cattle with Local ((SERVAC CAPRI-C®)) and imported (BOVIVAX LSD®) vaccines against LSD.

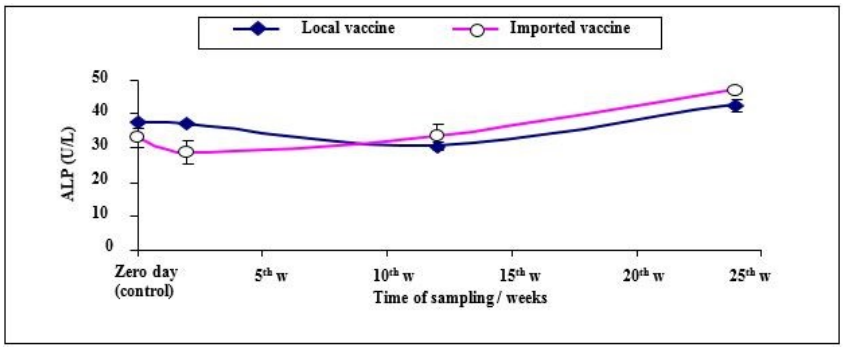

Fig.4. Levels of ALP of vaccinated cattle with Local ((SERVAC CAPRI-C®)) and imported (BOVIVAX LSD®) vaccines against LSD.

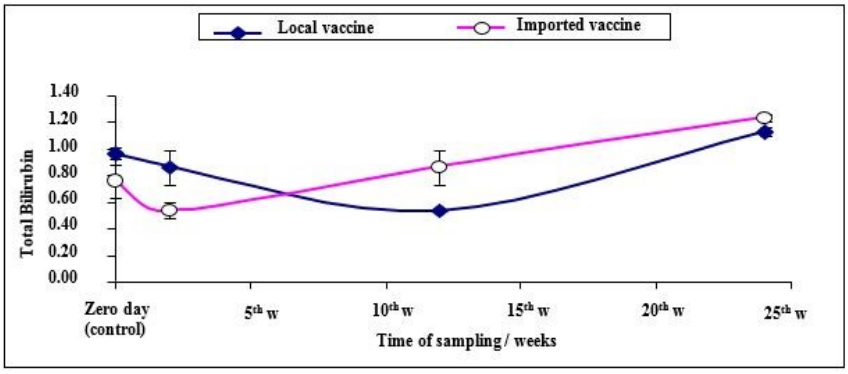

Fig.5. Total bilirubin of vaccinated cattle with Local ((SERVAC CAPRI-C®)) and imported (BOVIVAX LSD®) vaccines against LSD.

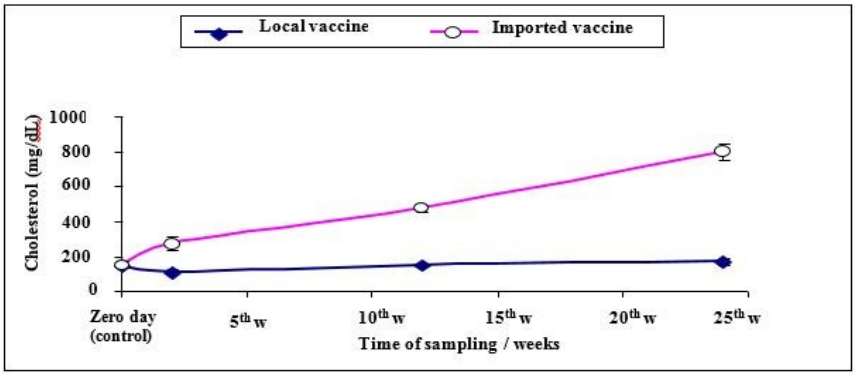

Fig.6. Cholesterol level of vaccinated cattle with Local ((SERVAC CAPRI-C®)) and imported (BOVIVAX LSD®) vaccines against LSD.

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تقييم الاستجابة المناعية للقاحات المحلية والمستوردة لفيروس مرض الجلد العقدي في الأبقار كخطوة وقائية

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الخلاصة

يعتبر التحصين ضد مرض التهاب الجلد العقدي هو الطريقة الفعالة للوقاية منه ومكافحته في المناطق الاكثر انتشارا، حيث ان الأجسام المناعية لها دورًا مهمًا في الحماية طويلة المدى بعد التحصين. تم تجميع 260 عينة سيرم من 20 تيران المحصنة، تراوحت أعمارها بين 9 إلى 12 شهراً. تم تقسيم هذه الثيران إلى مجموعتين (كل مجموعة تحتوي على 10 ثيران) في مزارع الأبقار في منشأة، القناطر أبوغالب، محافظة الجيزة، مصر. أجريت هذه الدراسة في الفترة من نوفمبر 2022 إلى مايو 2023. تم تحصين إحدى هذه المجموعات باللقاح المحلي و تحصين الاخري باللقاح المستورد، تم أخذ عينات السيرم قبل التحصين وبعد التحصين لمدة 6 أشهر مع فاصل زمني اسبوعين. هدفت دراستنا إلى تحديد مدى فعالية كلا من اللقاحين في مناعة الأبقار لتقييم الاستجابة المناعية للأبقار المحصنة ضد لقاح سلالة نيثلينغ. أظهرت النتائج زيادة معنوية ($P<0.05$) في مستوي الاجسام المناعيه طوال فترة التجربه للقاح المستورد مقارنه باللقاح المحلي الذي اظهرانخفاضًا ملحوظًا في مستوي الأجسام المناعيه بعد 24 أسبوعًا من التحصين. ، عملية التحصين قد يكون لها تأثير طفيف جدا على بعض انزيمات الكبد مثل اختبار ناقلة أمين الألانين ،في الختام فإن اللقاح المستورد له قدرة كبيرة على زيادة مستوى الأجسام المناعية لفترة طويله المدي لذلك يعتبر أفضل اختيار للوقاية والسيطرة على مرض التهاب الجلد العقدي ويمكن تطبيقه عمليا في مجال الطب البيطري.

الكلمات الدالة: مرض التهاب الجلد العقدي، سلالة نيثلينغ، اللقاح، المناعة، الوقاية.