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LATEX ANTIGEN FOR SEROLOGIC DIAGNOSIS OF EMPHYSEMATOUS CARBUNCLE IN ANIMALS

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Annotation. Studies in latex agglutination reaction (LAR) using *Clostridium chauvoei* antigen showed the specificity and suitability of the method for the detection of specific antibodies in animals vaccinated against this disease and suspected to have emphysematous carbuncle disease.

It was found that a serum LAR titer of 1:30 -1:40 in immunized animals 30 days after vaccination can be considered as sufficient, and titers of 1:80 and 1:160 as a high level of immune response.

When studying the dynamics of postvaccinal antibody titers in immunized animals under production conditions, it was found that the average antibody titer in LAR 30 days after vaccination was equal to 1:33. In animals immunized against emkar the sufficient level of antibodies was preserved only up to 90 days, on day 150 the average titers of postvaccinal antibodies decreased twice in comparison with titers established on day 30, and in 180 days in 4 times. It should be noted that in studies 180 days after vaccination, antibodies were not detected in 30% of animals. The results of the conducted studies should be taken into account when organizing and conducting prophylactic immunization of animals against emphysematous carbuncle.

Keywords: emphysematous carbuncle, *Clostridium chauvoei*, vaccine, latex antigen, latex-agglutination reaction.

Introduction. Emphysematous carbuncle is an acute non-contagious disease that occurs enzootically in unfavorable areas, manifested mainly by severe focal lesions of musculature in the form of crepitating necroses and serous hemorrhagic infiltration of adjacent subcutaneous tissue. Cattle are susceptible to emphysematous carbuncle and usually become ill between three months and four years of age; buffalo are more likely to become ill at 1-2 years of age; sheep, goats, and elk are much less likely to become ill [1].

The causative agent is *Clostridium chauvoei*, a strict anaerobe that is a polymorphic spore-containing bacillus. The source of the pathogen is animals with emphysematous carbuncle, whose corpses produce spores, which then infect soil, feed, and water [2-4]. Emphysematous carbuncle usually occurs in pastures, more often in hot, dry summers. In these conditions, animals, eating dry grass, capture simultaneously particles of soil and

together with it spores of the causative agent of the disease. The disease is characterized by stationarity, which is due to the prolonged persistence of the pathogen in the external environment (soil, water) [5].

Emphysematous carbuncle is widespread in all countries of the world regardless of geographical location and soil and climatic conditions. In unfavorable farms, it causes great damage due to animal deaths and costs of anti-epizootic measures [6-10]. In CIS countries, the disease is registered in all regions [11], including Kazakhstan.

Our earlier studies have established that the disease occupies one of the first places by the number of epizootic foci of acute infectious diseases of animals in Kazakhstan. In the period from 2010 to 2020, the territory of 10 regions comprising 71.4% of the territory of the Republic of Kazakhstan were unfavorable for this disease.

One of the main directions of prevention and control measures against emphysematous carbuncle is immunization of animals [12, 13]. In the CIS, "inactivated concentrated aluminum hydroxide vaccine against emphysematous carbuncle of cattle and sheep", which creates immunity lasting up to 6 months, is used to prevent the disease.

Analysis of the practice of using this vaccine has shown that in some farms of the Republic of Kazakhstan there are cases of this nosology among vaccinated animals 4-6 months after vaccination, which may be associated with the quality of the vaccine or deficiencies of veterinary practitioners in the organization and conduct of vaccination. These facts require studying the state of immune response in vaccinated animals during the post-vaccination period.

However, the guidelines for the use of vaccines against emphysematous carbuncle do not provide for the determination of the level of immunity by determining postvaccine antibody titers. One of the main reasons for this is the lack of effective serologic methods for diagnosing emcarus. In the specialized literature, there are some reports on the development of methods for the detection of specific anti-emkar antibodies in RA, RNHA, and RIF [14].

However, these methods remained only within the framework of laboratory studies and were not accepted for practical use. Nevertheless, the need to develop serologic methods of diagnostics of this disease is obvious, since this method can be used to detect specific antibodies in the blood serum of vaccinated animals, as well as in those who have been infected or suspected of being infected. Post-vaccination serological tests can be used to monitor the vaccination and the titers of specific antibodies in the blood serum of immunized animals, thereby determining the quality of the vaccine used.

Therefore, the search for alternative methods of diagnostics of emphysematous carbuncle with sensitivity and specificity, ease of handling and accessibility is highly relevant.

In this aspect, there have been a number of recent reports on the prospects of new test systems based on polystyrene carriers of immunosorbents - polystyrene latexes, which are used in the latex-agglutination test [15, 16]. In view of the above, the aim of the present work was to develop and test an antigen for the latex agglutination reaction for the serologic diagnosis of this disease.

Materials and methods. The animals officially regulated for the diagnosis of emphysematous carbuncle and research methods according to GOST 26503-85 were used in the research work.

For epizootological monitoring of the disease, research methods described in the relevant manuals were used.

In order to study the epizootological manifestation of the disease, the following were used and analyzed: statistical reviews and official reports of the Committee for Veterinary Control and Supervision of the Ministry of Agriculture of the Republic of Kazakhstan (CVCS of MoA of RK), and Republican Veterinary Laboratory (RVL) on veterinary welfare of emphysematous carbuncle of animals; materials of own clinical and epizootological surveys

of epizootic foci of this nosology and assessments of the epizootic situation in different regions of the Republic.

207 blood samples of cattle immunized against emphysematous carbuncle or suspected of being infected with this disease, delivered from different regions of the republic, were subjected to serological study.

For comparative serologic study of the tested blood sera we used agglutination reaction (RA) according to the method described in the work of Odarenko K.I. and Gryazin V.I. As antigen in RA we used 10 billionth suspension of *Clostridium chauvoei* culture inactivated by formalin [41]. The reaction was carried out by the classical method in serological tubes in a total volume of 1 ml, at 370C in the thermostat, the results of the reaction were taken into account after 16-18 hours.

The formation of a precipitate of agglutinate flakes at the bottom of the tube was considered positive; in negative cases, the contents of the tube remained homogeneously turbid without precipitate formation.

The proposed diagnosticum for the latex-agglutination reaction was prepared according to the following procedure: microbial mass of *Clostridium chauvoei* in distilled water at a concentration of 10 bln. m.s.c. in 1.0 ml was inactivated with formalin at a final concentration of 0.7% in a thermostat at 37°C for 3 days, then ultrasonic disintegration was performed. in 1.0 ml was inactivated with formalin at a final concentration of 0.7% in the thermostat at 37°C for 3 days, then ultrasonic disintegration of *Clostridium chauvoei* microbial cells was carried out three times, 20 minutes each, with an interval of 30 minutes at a current of 0.6 ampere and a frequency of 22 kHz, followed by centrifugation at 5000 rpm for 10-15 minutes, then the precipitate was resuspended to the original volume with phosphate-salt buffer pH 7.2. Further sensitization of 1% polymer suspension of latex particles was carried out with an equal volume of ultrasound-disintegrated emphysematous carbuncle antigen containing 5, 15 and 30 mg/ml, using phosphate-salt buffer pH - 7.2 as a dissolving liquid of latex and antigen, followed by incubation for 2 hours in the thermostat at 37°C.

The adsorption products were washed by centrifugation at 3000 rpm for 10-15 minutes three times and resuspended to the original volume with phosphate-salt buffer pH 7.2. Sodium azide solution was used to preserve the prepared antigen to a final concentration of 0.05%.

Thus, prepared variants of diagnostics with different concentrations of antigen were used for the latex-agglutination reaction (LAR) in dilutions of tested animal sera 1:10, 1:20, 1:40, 1:80, 1:160, 1:320. The reaction was carried out in polystyrene microplates, at room temperature for 5-6 hours. A positive result was considered to be the formation of clear agglutination at the bottom of the wells of the plate, in the form of large or small flakes and clarification of the liquid.

If the reaction results are negative, the latex suspension remains homogeneously turbid, with no agglutinate clumps or areas of luminescence.

Positive serum obtained by hyperimmunization of rabbits with the emkar pathogen and negative serum obtained from healthy rabbits in the same dilutions were used as a reaction control.

To assess the specificity of the diagnosticum, blood sera from healthy calves were used, as well as those vaccinated against pasteurellosis, anthrax and brucellosis.

207 blood samples of cattle immunized against emkar or suspected of being infected with this disease, delivered from different regions of the republic, were subjected to serological method of research.

Results. At the initial stage of research to determine the optimal concentration of antigen in the diagnosticum, the latex-agglutination reaction was performed with negative and positive serum for emphysematous carbuncle using diagnosticums containing different

concentrations of antigen of this nosology.

The results are summarized in Table 1.

Table 1 – LAR results at different concentrations of antigen in the diagnosticum

№	Sera studied	Antigen concentration to 1 volume of latex suspension					
		5 mg/mL		15 mg/mL		30 mg/mL	
		Positive in exploration	Antigen control	Positive in exploration	Antigen control	Positive in exploration	Antigen control
Animals containing antibodies to *Cl. Chauvoei* immunized against emphysematous carbuncle							
1		1:20	-	1:80	-	1:80	s/a
2		1:10	-	1:40	-	1:40	-
3		1:10	-	1:80	-	1:40	-
4		1:20	-	1:40	-	1:80	s/a
5		1:20	-	1:80	-	1:80	s/a
Non-containing antibodies to *Cl.Chauvoei* (healthy animals)							
1		-	-	-	-	-	-
2		-	-	-	-	-	-
3		-	-	-	-	-	-

Примечание: s/a – self-agglutination.

As shown in Table 1, the diagnosticum with antigen content of 5 mg/mL showed weak agglutination on 2 crosses in the first two dilutions (1:10 and 1:20) of positive serum, and increasing the antigen load per unit of carrier up to 30 mg/mL resulted in spontaneous agglutination in controls or gives indistinctly outlined discs of agglutination on a turbid background. The optimal concentration of antigen was 15 mg/mL to 1 volume of latex suspension, which shows higher antibody titers (1:40 and 1:80) and clear pictures of controls.

Further, using the diagnosticum containing 15 mg/mL antigen to 1 volume of latex suspension as the most optimal and previously known RA method, 3 samples of blood sera from cattle suspected of disease incidence and 10 samples of blood sera from animals vaccinated against emphysematous carbuncle were investigated. In parallel, blood serum samples from healthy animals and those vaccinated against pasteurellosis, anthrax and brucellosis were examined. The results are summarized in Table 2.

Table 2 – Results of RA and LAR in blood serum studies of cattle with different immunologic statuses

№	Characteristics of the studied animals	Test results, antibody titers					Diagnostic control
		1:10	1:20	1:40	1:80	1:160	
		RA / LAR	RA / LAR	RA / LAR	RA / LAR	RA / LAR	
1	**2**	**3**	**4**	**5**	**6**	**7**	**8**
1	Suspects of emcarbative disease	- / -	- / -	- / -	- / -	- / -	- / -
2	-//-//-//-//-//-	- / -	- / -	- / -	- / -	- / -	- / -
3	-//-//-//-//-//-	++++/++++	++++/++++	- / ++++	- / -	- / -	- / -
	30 days after vaccination						
1	against emcar	++++ /++++	++++ /++++	++++ /++++	- / +++	- / -	- / -
2	-//-//-//-//-//-	++++ /++++	++++ /++++	++++ /++++	- / +++	- / -	- / -

1	2	3	4	5	6	7	8
3	-//-//-//-//-//-	++++ /++++	++++ /++++	++ /++++	- / -	- / -	- / -
4	-//-//-//-//-//-	++++ /++++	++++ /++++	++++ /++++	+++ / ++++	- / +++	- / -
5	-//-//-//-//-//-	++++ /++++	++++ /++++	++++ /++++	- / +++	- / -	- / -
6	-//-//-//-//-//-	++++ /++++	++++ /++++	- / +++	- / -	- / -	- / -
7	-//-//-//-//-//-	++++ /++++	++++ /++++	++++ /++++	- / +++	- / -	- / -
8	-//-//-//-//-//-	++++ /++++	++++ /++++	++++ /++++	+++ / ++++	- / +++	- / -
9	-//-//-//-//-//-	++++ /++++	++++ /++++	++ /++++	- / -	- / -	- / -
10	-//-//-//-//-//-	++++ /++++	++++ /++++	- / +++	- / -	- / -	- / -
1	against pasteurellosis	-	-	-	-	-	-
2		-	-	-	-	-	-
3		-	-	-	-	-	-
1	against anthrax	-	-	-	-	-	-
2		-	-	-	-	-	-
3		-	-	-	-	-	-
1	healthy animals	-	-	-	-	-	-
2		-	-	-	-	-	-
3		-	-	-	-	-	-

As can be seen from Table 2, comparative serologic studies of the tested sera in RA and LAR showed similar results of both methods in serum dilutions of 1:10 and 1:20. In serum dilution 1:40 against 11 positive LAR reactions, only 6 samples (54.5%) reacted positively in RA and 2 samples (18.2%) reacted as doubtful. Positive LAR results in titer 1:80 was obtained in 6 samples, when RA was positive in only 2 cases (33.3%). In dilution of tested blood sera 1:160 by LAR, positive results were registered in 2 samples, while RA results were negative. In studies of healthy and immunized animals with heterologous vaccines the same negative results were obtained, which testifies to the specificity of the compared methods. In terms of sensitivity (33.3 - 54.5%) RA was much inferior to LAR. Taking into account these data, LAR with the developed antigen was used in further studies to detect specific antibodies to emphysematous carbuncle in the blood serum of animals.

From the same table it is seen that, in studies of animals vaccinated against emkar in LAR, 30 days after vaccination, 100% of animals reacted positively in titer 1:40, 60% in titer 1:80 and 20% in titer 1:160. It follows that the serum titer in LAR 1:40 in vaccinated animals 30 days after vaccination can be considered as sufficient, and titers of 1:80 and 1:160 as a high level of immune response. When examining the blood sera of 3 animals suspected of the disease, one of them had a positive result with a titer of 1:40, which indicates the presence of antibodies specific to the causative agent of emphysematous carbuncle in the animal's body.

Thus, the developed latex antigenic diagnosticum proved to be suitable for LAR and allows to detect specific antibodies in bovine blood serum to the causative agent of emphysematous carbuncle in animals suspected of the disease and vaccinated against this infection. Negative results of LAR in healthy and immunized animals with heterologous vaccines and in the reaction control testify to the specificity of the manufactured diagnosticum.

At the next stage of work, postvaccinal studies were carried out on 167 samples of blood sera of animals immunized against emphysematous carbuncle delivered from different regions of the Republic (northern, southern, eastern, western and central), which were tested in latex-agglutination reaction using the developed antigen (Table 3).

As shown in Table 3, 23 days after vaccination, the mean antibody titers in animals in LAR were 1:45; after 30 and 35 days, respectively, 1:34 and 1:37; after 150 days, postvaccine antibodies were detected in 76.6% of animals, with mean titers of 1:11, indicating a very low level of immune response of the organism. In immunized animals, there was no significant difference in antibody titers after vaccination depending on the area of their location and the type of vaccine.

Table 3 – Results of blood serum of animals inoculated against emkar at LAR

№	Region	Name of area from which the sample is taken	Name of vaccine used	Study time after vaccination (days)	Quantity			Average antibody titer
					tested samples	Positive results /%	Negative results /%	
1	eastern	East Kazakhstan	cont.GOA vaccine, Stavr.b/f	23	30	30/100	0/0	1:45
2	western	West Kazakhstan	cont.GOA vaccine, Stavr.b/f	30	30	30/100	0/0	1:34
3	central	Karaganda	cont.GOA vaccine, Stavr.b/f	30	30	30/100	0/0	1:34
4	южный	Almaty	cont.GOA vaccine, Armavir.b/f	35	47	47/100	0/0	1: 37
5	northern	Kostanay	cont.GOA vaccine, Stavr.b/f	150	30	23/76,6	7/23,3	1:11

Note: Concentrated GOA vaccine - concentrated allium hydroxide vaccine. Stavr.b/f - Stavrapol biofactory; Armavir.b/f - Armavir biofactory; Pos.res - positive result; Neg.res - negative result.

Thus, the conducted studies have shown the possibility of determining the level of antibodies in the serum of animals vaccinated against emcarus, using the developed emcarus antigen. At the next stage of work, in farm "Syrymbet" of Talgar district of Almaty region, the dynamics of titer of postvaccinal antibodies in cattle vaccinated against emphysematous carbuncle was studied during 180 days (6 months).

Ten experimental animals were injected with Emkara vaccine produced in Armavir biofactory in accordance with the instructions for use. To study the dynamics of postvaccinal antibodies, immunized animals were serologically tested after 30,90,150 and 180 days. The results of the study are presented in Table 4.

As can be seen from Table 4, the average antibody titer in animals 30 days after vaccination was 1:33. In the following terms of the study (90, 150 and 180 days), the average antibody titer in animals of this group was 1:29, 1:16 and 1:8, respectively. It should be noted that in the studies in 180 days after vaccination in 3 animals (30%) antibodies were not detected at all. The obtained data shows that in animals immunized with vaccine against emphysematous carbuncle sufficient level of antibodies were preserved only up to 90 days, on day 150 the average titers of postvaccinal antibodies decreased twice in comparison with titers

established on day 30 and amounted to 1:16, and after 180 days the level of antibodies decreased 4 times, i.e. up to 1:8.

Table 4 – Dynamics of postvaccinal antibody titers of cattle immunized against emphysematous carbuncle

№	Individual No. of animals	Timing (days) of the study and antibody titer after vaccination			
		30	90	150	180
1	00331387	1:40	1:30	1:20	1:20
2	58188906	1:40	1:40	1:20	1:10
3	96760014	1:30	1:30	1:20	1:10
4	60237761	1:30	1:30	1:10	-
5	60237760	1:40	1:40	1:20	1:10
6	61278185	1:20	1:20	1:10	-
7	60636700	1:40	1:30	1:20	1:10
8	62056139	1:20	1:20	1:10	-
9	59685365	1:40	1:30	1:20	1:10
10	60237768	1:30	1:20	1:10	1:10
Average titer		1:33,0	1:29,0	1:16,0	1:8,0

These data indicate that in this farm sufficient level of immunity in animals immunized against emphysematous carbuncle were preserved only up to 90 days, and in the following terms sharply decreased from 2 to 4 times. The obtained data should be taken into account when organizing and conducting prophylactic vaccination of animals against emphysematous carbuncle of animals.

Discussion. Emphysematous carbuncle is an acute non-contagious toxic-infectious disease of cattle characterized by the formation of rapidly enlarging crepitating swellings in the body muscles and lameness.

Cattle of improved, cultured, especially beef breeds (with large muscle mass) and fatter individuals are more susceptible to the disease. Animals brought to the unfavorable zone from other farms or imported animals often fall ill. Cattle of any age are affected, but young animals aged 3 months to 3-4 years are most susceptible [17-19].

The causative agent of emphysematous carbuncle *Clostridium chauvoei* is an anaerobe; it is a straight or slightly curved bacillus with rounded ends, arranged singly, in pairs, less often in short chains; in young cultures it is Gram-positive. Spores of the pathogen are very stable: they remain viable in soil for several years, in rotting muscles, manure - up to 6 months, at the bottom of water bodies - over 10 years, under appropriate conditions in soil the pathogen can vegetate and multiply. Biological properties of the emphysema pathogen have been studied by many researchers.

The causative agent of emphysematous carbuncle synthesizes and secretes exotoxin. Hemotoxic and necrotizing components are found in the composition of the toxin. Another diagnostic factor of this microbe is its ability to produce aggresins [20].

Emphysematous carbuncle of cattle is widespread in many countries of the world, including the Republic of Kazakhstan (RK). By the number of registered epizootic foci of acute infectious diseases of animals in Kazakhstan, emphysematous carbuncle occupies one of the first places. In the period from 2010 to 2020, the territory of 10 regions comprising 71.4% of the country's territory were unfavorable for this disease.

One of the main directions of prevention and control measures against emphysematous carbuncle in Kazakhstan is immunization of animals with "inactivated concentrated aluminum hydroxide vaccine against emphysematous carbuncle of cattle and sheep", which creates immunity lasting up to 6 months.

In the practice of using vaccines against other particularly dangerous diseases (foot and mouth disease, nodular dermatitis, brucellosis, etc.), the level of immune response in immunized animals is revealed by serological examination of vaccinated animals after a certain time and determination of the titer of postvaccinal antibodies in blood serum. Based on the results of these studies, the level of immune restructuring of the organism is judged [21].

However, the guidelines for the use of emkar vaccines do not provide for the determination of the level of immune response by determining postvaccine antibody titers. One of the main reasons for this is the lack of effective serologic methods for diagnosing emkar. Serologic methods of diagnostics of this disease have not been developed, because blood serum of an animal diseased with emphysematous carbuncle does not contain specific antibodies due to the rapid course of the disease and animal death and is unsuitable for diagnostics.

The reports available in the scientific literature on the diagnosis of emphysematous carbuncle using modern methods concern the indication of the causative agent of emphysematous carbuncle from pathological material and animal products and its molecular genetic analysis [21].

Some previously published reports on the detection of antibodies in animals inoculated against emphysematous carbuncle by various serologic reactions (RA, long precipitation reaction, indirect immune fluorescence reaction) remained only within the framework of laboratory studies and were not accepted for practical us.

However, the need for the development of serological methods for the diagnosis of emphysematous carbuncle is obvious, since this method can be used to detect specific antibodies in the blood serum of animals that have been infected, suspected of being infected or vaccinated against emphysematous carbuncle.

In view of the above, the aim of the present work was to develop and test an antigen for setting up a latex agglutination reaction for serologic diagnosis of this disease.

As a result of the conducted studies, the researchers developed an inactivated emphysematous carbuncle antigen obtained by disintegration of the pathogen *Clostridium chauvoei* under the influence of ultrasound with its subsequent sensitization to latex particles.

Studies in the latex agglutination reaction (LAR) using this antigen have shown the specificity and suitability of the method for the detection of specific antibodies in disease suspects and in animals vaccinated against the disease.

Comparative serologic studies of tested blood sera in LAR in comparison with the known RA method showed that RA was much inferior to LAR in terms of sensitivity (33.3 - 54.5%). In studies of healthy and immunized animals with heterologous vaccines, the same negative results of both reactions were obtained, which testify to the specificity of the compared methods, LAR compares favorably with RA in terms of reaction time (RA is carried out within 16-18 hours, LAR within 5-6 hours), as well as the pronounced manifestation of the agglutination phenomenon when reading the reaction results. LAR does not require complex laboratory equipment (thermostats, water baths, etc.) and can be performed in the field.

Taking this into account, in further studies, LAR with the developed antigen was used to detect specific anti-emetic antibodies in the blood serum of animals.

Postvaccinal studies of animals immunized against emphysematous carbuncle under production conditions, delivered from different regions of the country, were carried out. It was found that in 23 days after vaccination the average antibody titers in animals in LAR were 1:45; in 30 and 35 days, respectively, 1:34 and 1:37; in 150 days postvaccinal antibodies were detected in 76.6% of animals, the average titers of which were equal to 1:11, indicating a very low level of immune response of the organism.

To study the dynamics of postvaccinal antibodies, 10 experimental animals were

injected with emphysematous carbuncle vaccine according to the instructions for use. Immunized animals were subjected to serological tests after 30,90,150 and 180 days.

The average antibody titer in animals 30 days after vaccination was found to be 1:33. In the following terms of the study (90, 150 and 180 days) the average antibody titer in animals was 1:29, 1:16 and 1:8, respectively. It should be noted that in the studies in 180 days after vaccination in 3 animals (30%) antibodies were not detected at all. The obtained data shows that in this farm a satisfactory level of immune response in vaccinated animals was maintained only up to 90 days, and in terms of 150 and 180 days after vaccination titers of post-vaccination antibodies decreased by 2 and 4 times, respectively. These data indicate a sharp decrease in immunity to the causative agent of emphysematous carbuncle and the need to take measures to correct the immune state of animals.

Conclusion. Taking into account the absence of effective serological methods of diagnostics of emphysematous carbuncle allowing to determine the state of immune response in vaccinated animals, the authors developed an inactivated antigen obtained by disintegration of the pathogen *Clostridium chauvoei* under the influence of ultrasound with its subsequent sensitization to latex particles.

LAR with the use of antigenic latex diagnosticum allows to detect specific antibodies in the blood serum of cattle to the causative agent of emkar in animals suspected of the disease and vaccinated against this infection. Using this method, the dynamics and level of postvaccinal antibody titers in the blood serum of cattle were analyzed. It was found that a sufficient level of postvaccinal antibody titers in immunized animals remained only up to 90 days, and on 150 and 180 days after vaccination the titers of postvaccinal antibodies decreased by 2 and 4 times, respectively. These data indicate the need to take measures to correct the immune state of animals for emphysematous carbuncle. The results of the conducted studies should be taken into account when organizing and conducting prophylactic immunization of animals against animal emphysematous carbuncle.

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ЖАНУАРЛАРДАҒЫ ЭМФИЗЕМАТОЗДЫ КАРБУНГКУЛДЫҢ СЕРОЛОГИЯЛЫҚ ДИАГНОСТИКАСЫНА АРНАЛҒАН ЛАТЕКС АНТИГЕНІ

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Аңдатпа. *Clostridium chauvoei* антигенін пайдалана отырып, латекс агглютинация

реакциясын (ЛАР) зерттеу осы ауруға қарсы вакцина егілген және эмфизематозды қара аяғы бар деп күдіктенген жануарларда спецификалық антиденелерді анықтау әдісінің ерекшелігі мен жарамдылығын көрсетті.

Вакцинациядан кейін 30 күннен кейін иммунизацияланған жануарларда қан сарысуындағы 1:30 -1:40 қан сарысуының титрі жеткілікті, ал 1:80 және 1:160 титрлері иммундық жауаптың жоғары деңгейі ретінде қарастырылуы мүмкін екендігі анықталды.

Өндірістік жағдайда иммунизацияланған жануарларда вакцинациядан кейінгі антиденелер титрінің динамикасын зерттеу кезінде вакцинациядан кейінгі 30 күннен кейін ЛАР-дағы антиденелердің орташа титрінің 1:33-ке тең екендігі анықталды. Эмкарға қарсы иммунизацияланған жануарларда антиденелердің жеткілікті деңгейі тек 90 күнге дейін сақталды; 150-ші күні вакцинациядан кейінгі антиденелердің орташа титрлері 30-шы күні белгіленген титрлермен салыстырғанда екі есеге, ал 180 күннен кейін 4 есеге төмендеді. Бұл деректер вакцинациядан кейін 150 күннен бастап вакцинацияланған жануарларда иммундық жауап деңгейінің төмендеуін және эмфизематозды карбункулы бар жануарлардың иммундық жағдайын түзету бойынша шаралар қабылдау қажеттігін көрсетеді. Жануарларды эмфизематозды карбункулға қарсы профилактикалық иммунизациялауды ұйымдастыру және жүргізу кезінде жүргізілген зерттеулердің нәтижелерін ескеру қажет.

Тірек сөздер: вакцина, *Clostridium chauvoei*, латексті антиген, латекс-агглютинация реакциясы, эмфизематозды карбункул.

ЛАТЕКС АНТИГЕН ДЛЯ СЕРОЛОГИЧЕСКОЙ ДИАГНОСТИКИ ЭМФИЗЕМАТОЗНОГО КАРБУНКУЛА ЖИВОТНЫХ

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Аннотация. Исследования в реакции латекс-агглютинации (РЛА) с использованием антигена *Clostridium chauvoei* показала специфичность и пригодность метода для выявления специфических антител у вакцинированных против этой болезни и подозреваемых в заболеваний эмфизематозным карбункулом животных.

Установлено, что титр сыворотки в РЛА 1:30 -1:40 у иммунизированных животных через 30 дней после вакцинации можно считать как достаточный, а титр 1:80 и 1:160 как высокий уровень иммунного ответа.

При изучении динамики поствакцинальных титров антител у иммунизированных животных в производственных услових установлено, что средний титр антител в РЛА через 30 дней после вакцинации был равен 1:33. У иммунизированных против эмкара животных достаточный уровень антител сохранялся только до 90 дней, на 150 день средние титры поствакцинальных антител снизились в два раза по сравнению с титрами установленные на 30 день, а через 180 дней в 4 раза. Эти данные указывают на снижение уровня иммунного ответа у привитых животных начиная с 150 дней после вакцинации и о необходимости принятия мер по коррекции иммунного состояния животных по эмфизематозному карбункулу. Результаты проведенных исследований необходимо учесть при организации и проведении профилактической иммунизации животных против эмфизематозного карбункула.

Ключевые слова: вакцина, *Clostridium chauvoei*, латексный антиген, реакция латекс-агглютинации, эмфизематозный карбункул.