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AVIAN ARBOVIRUSES OF THE WITLESS BAY SEABIRD SANCTUARY, NEWFOUNDLAND, CANADA[1]

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Abstract: A virologic and serologic survey of arbovirus infections among seabirds and seabird ticks, *Ixodes uriae*, on Great Island, Witless Bay, Newfoundland, Canada, was conducted during 1971 and 1972. Kemerovo (Great Island, Bauline) and Sakhalin (Avalon) group viruses previously reported from birds and/or ticks on Great Island were prevalent among avian populations, while conclusive evidence of known nonindigenous serotypes was lacking. Circumstantial evidence—hemagglutination inhibiting antibody—of an unidentified member of the group B tick-borne encephalitis complex transmitted among marine birds of North America by *I. uriae* is reported. No evidence of human infections with any of these viruses was detected in a small number of biologists doing research on Great Island.

INTRODUCTION

The Witless Bay Seabird Sanctuary consists of three small islands—Great Island, Gull Island, and Green Island—located off the coast of the Avalon Peninsula in Newfoundland, Canada. Large, relatively stable populations of marine birds nest here each year. Large breeding colonies of Leach's storm petrels (*Oceanodroma leuchorhoa*), common murres (*Uria aalge*), common puffins (*Fratercula arctica*), black-legged kittiwakes (*Rissa tridactyla*) and herring gulls (*Larus argentatus*), with smaller numbers of great black-backed gulls (*Larus marinus*), razorbills (*Alca torda*), black guillemots (*Cepphus grylle*), thick-billed murres (*Uria lomvia*), and northern fulmars (*Fulmaris glacialis*) inhabit these islands from early spring until fall.

Because of several ecological conditions ideal for the enzootic transmission of various microorganisms [i.e. a large, annually renewed supply of susceptible, gregarious, ground-nesting vertebrates interacting with populations of vector species of ticks (*Ixodes uriae*) and mosquitoes (*Aedes* spp.)], Great Island, the southernmost of these three rookeries (47° 11′ N; 42° 46′ W) was selected for a study of the arthropod-borne viruses occurring in the avian populations of the North Atlantic. The isolation and characterization of newly recognized viruses—Great Island, Bauline, and Avalon—from this island were previously reported;[17,18] the current paper includes additional isolates of these viruses from ticks and evidence of past infection by these and other viruses in the birds.

METHODS AND PROCEDURES

The topography, vegetation and avifauna of Great Island are given in detail by Nettleship.[21]

I. uriae were collected from birds, their nests and burrows, as well as from the ground on Great Island, during July, 1971 and 1972. These ticks were transported alive in stoppered glass tubes, containing a moistened plaster-of-Paris and

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charcoal mixture, to the Yale Arbovirus Research Unit in New Haven, Connecticut; they were then triturated in 0.75% bovine albumin in phosphate-buffered (pH 7.2) saline, centrifuged at 2500 RPM (1600 X g), the supernatant fluid decanted, and inoculated intracerebrally (IC) into 1-to 4-day old Swiss mice. Virus isolates were identified by standard serologic techniques.[17]

In July, 1972, blood samples were drawn by cardiac puncture from several species of birds captured by hand or in Japanese mist nets on the island. Samples for virus isolation attempts obtained from unfledged chicks were sealed in glass ampoules in the field and frozen in liquid nitrogen. These bloods, like the tick suspensions, were screened for virus by IC inoculation of suckling mice. Blood samples from juvenile or adult birds to be tested for antibody were centrifuged and the sera held in screw-capped glass tubes on wet ice until they reached the laboratory where they were stored at —20C.[18]

Blood samples were drawn by venipuncture from a small number of biologists involved in ornithological studies on these islands; these samples were handled in the same manner as the avian sera.

Hemagglutination (HA) and hemagglutination-inhibition (HI) tests were done by micro-techniques with sucrose-acetone extracted suckling mouse brain antigens and acetone extracted sera.[3] The HI test included 0.025 ml of 4 to 8 HA units of antigen in 0.4% bovine albumin in borate saline (pH 9.0), 0.025 ml of two-fold serial dilutions of sera starting at 1:10, and 0.05 ml of goose erythrocytes in the appropriate pH adjusting diluent (1:24) added after 18 hr incubation at 4 C. The antigens used in these tests were: eastern equine encephalomyelitis (EEE) (Ten Broeck strain from the eastern USA), western equine encephalomyelitis (WEE) (USA 72666 from Connecticut), Sindbis (SIN) (EgAr 339 from Egypt), St. Louis encephalitis (SLE) (Parton strain from the western USA), Japanese B encephalitis (JBE) (Nakayama strain from Japan), West Nile (WN) (UgB 956 from Uganda), tick-borne encephalitis Russian spring-summer (RSSE) (Sophy strain from eastern USSR), louping ill (LI) (Moredun strain from Scotland), Powassan (POW) (Byers strain from Ontario), and Tyuleniy (TYU) (USSR LEIV 6c from the eastern USSR).

Neutralization (Nt) antibody was detected by incubating 100 (final = 50) LD_{50} of virus with an equal volume of untreated, undiluted sera for one hour at 37 C; the mixture was then inoculated IC into suckling mice. Virus stocks used in the Nt tests included: LI (Moredun strain) passage 13; TYU (USSR LEIV 6c) passage 8, Great Island (GI) (CanAr 41 from Newfoundland) passages 3, 5, and 7; Bauline (BAU) (CanAr 14 from Newfoundland) passages 5 and 7; Yaquina Head (YH) (USA 56298-15) from western US) passages 8, 11, and 12; Cape Wrath (CW) (ScotAr 20 from Scotland) passage 3; Nugget (NUG) (AusMI 14847 from Australia) passage 6; an unnamed Kemerovo group virus (DenAr 3 from the Faeroe Islands) passage 3; Avalon (AVA) (CanAr 173 from Newfoundland) passage 8; Clo Mor (CM) (ScotAr 7 from Scotland) passages 8 and 10; Sakhalin (SAK) (USSR LEIV 71c from eastern USSR) passage 8. Immune ascitic fluids were drawn from adult female mice following 4 weekly intraperitoneal injections of virus and Freund's complete adjuvant.[17]

RESULTS

Virus was recovered from 16 of 143 pools of *I. uriae* collected in 1971 and 1972 (Tables 1, 2). Thirteen of these isolates were Kemerovo group viruses—seven identified as GI and six as BAU by Nt tests (Table 3). Kemerovo group isolates were predominantly GI in 1971 and BAU in 1972 (Tables 1, 2). Both GI and BAU were recovered from nymphs and adult female ticks; BAU was also isolated from adult males suggesting transstadial transmission since the ticks do not feed in this stage. No isolates were made from eggs or larvae.

Three strains were identified as AVA, a new Sakhalin group virus.[16] These isolates were made from nymphs and adult males collected in 1971 and from adult female ticks in 1972 (Tables 1, 2).

TABLE 1. Virus isolation attempts from **Ixodes uriae** collected in Witless Bay, Newfoundland, Canada during 1971 and 1972.

Stage/Sex	Number Tested	Number of pools	Great Island Virus Isolations	Great Island Virus MIR*	Bauline Virus Isolations	Bauline Virus MIR*	Avalon Virus Isolations	Avalon Virus MIR*
1971								
Eggs	227	3	0		0		0	
Larvae	46	5	0		0		0	
Nymphs	193	22	4	2.1	1	0.5	1	0.5
Adult Males	94	11	0		0		1	1.1
Adult Females	57	18	2	3.5	0		0	
subtotals	617	59	6	1.0	1	0.2	2	0.3
1972								
Eggs	0	0	0		0		0	
Larvae	2	1	0		0		0	
Nymphs	286	38	0		1	0.3	0	
Adult Males	11	3	0		1	9.1	0	
Adult Females	299	42	1	0.3	3	1.0	1	0.3
subtotals	598	84	1	0.2	5	0.8	1	0.2
TOTALS	1215	143	7	0.6	6	0.5	3	0.2

* minimum infection rates as per cent

TABLE 2. Virus isolations from ticks (**Ixodes uriae**) and birds (**Larus argentatus**) on Great Island, Witless Bay, Newfoundland, Canada during 1971 and 1972.

Strain	Source	Microhabitat	Date	Reisolated	Serogroup	Virus
*CanAr 14	10 engorged nymphs	Puffin Burrows	7/23/71	—	Kemorovo	Bauline
CanAr 15	6 engorged nymphs	Puffin Burrows	7/23/71	+	Sakhalin	Avalon
CanAr 32	1 engorged female	Puffin Burrows	7/23/71	—	Kemerovo	Great Island
CanAr 40	10 engorged nymphs	Puffin Burrows	7/27/71	—	Kemerovo	Great Island
*CanAr 41	10 engorged nymphs	Puffin Burrows	7/27/71	+	Kemerovo	Great Island
CanAr 42	10 engorged nymphs	Puffin Burrows	7/27/71	—	Kemerovo	Great Island
CanAr 45	2 unengorged nymphs	Puffin Burrows	7/27/71	+	Kemerovo	Great Island
CanAr 46B	9 unengorged nymphs	Puffin Burrows	7/27/71	+	Sakhalin	Avalon
CanAr 49	10 engorged nymphs	*L. argentatus*	7/27/71	—	Kemerovo	Great Island
CanAr 63	6 unengorged males	Rocks, Burrows	7/12/72	—	Kemerovo	Bauline
CanAr 128	11 engorged females	*L. argentatus*	7/20/72	—	Kemerovo	Bauline
CanAr 133	9 engorged nymphs	*L. argentatus*	7/20/72	—	Kemerovo	Bauline
CanAr 172	10 engorged females	*L. argentatus*	7/31/72	+	Kemerovo	Bauline
*CanAr 173	10 engorged females	*L. argentatus*	7/31/72	+	Sakhalin	Avalon
CanAr 174	6 engorged females	*L. argentatus*	7/31/72	+	Kemerovo	Bauline
CanAr 176	8 engorged females	*L. argentatus*	7/31/72	+	Kemerovo	Great Island
CanAn 476	Blood (*L. argentatus*)		7/31/72	+	Sakhalin	Avalon

* prototype strains of the viruses

TABLE 3. Neutralization tests comparing Kemerovo group virus isolations from **Ixodes uriae** collected in Oregon, U.S.A. (USA-15), and Newfoundland, Canada (CanAr 41; CanAr 14).

	Virus			A.F.**		
	Great Island (CanAr 41)	Bauline (CanAr 14)	Yaquina Head (USA 15)	Great Island (CanAr 41)	Bauline (CanAr 14)	Yaquina Head (USA 15)
CanAr 41		0.4/3.4*	0.6/3.4		0.2/2.7	0.6/3.4
CanAr 32	1.3/3.2	0.3/1.8	0.6/1.8	1.8/3.4	0.0/3.0	0.2/3.7
CanAr 40	1.5/2.4	0.0/3.7	0.3/3.7	3.3/3.4	0.1/3.0	0.0/3.7
CanAr 42	2.4/3.0	0.3/3.0	0.3/3.0	2.3/3.4	0.0/3.0	0.6/3.7
CanAr 45	3.7/2.8	0.1/2.8	0.0/2.8	3.5/3.4	0.7/3.0	0.6/3.7
CanAr 49	3.2/2.1	0.0/4.0	0.3/4.0	3.5/3.4	0.9/3.0	≦0.8/3.7
CanAr 176 A.F. A	0.0/2.3	0.4/2.3	0.4/2.3	1.4/2.8	0.9/3.0	0.3/3.7
A.F. B	0.0/1.7	0.0/1.7				
CanAr 14	0.2/2.7		0.7/2.7	0.4/3.4		0.3/3.9
CanAr 63	0.0/2.7	2.5/2.8	0.7/2.8	0.7/2.8	2.9/3.0	0.3/3.7
CanAr 128 A.F. A	0.0/2.2	0.3/2.2	0.9/2.2	1.6/2.8	2.4/3.0	0.8/3.7
A.F. B	0.5/1.9	0.0/1.9				
CanAr 133 A.F. A	0.0/≧1.8	0.4/≧1.8	0.9/≧1.8	1.5/2.8	2.0/3.0	0.9/3.7
A.F. B	0.5/2.8	0.9/2.8				
A.F. C	0.9/3.2	0.5/3.2				
CanAr 172	0.0/≧2.8	3.3/≧2.8	1.0/≧2.8	2.0/2.8	2.5/3.0	0.2/3.7
CanAr 174	0.0/2.2	1.8/2.2	1.1/2.2	1.5/2.8	2.0/3.0	0.9/3.7
USA 15	0.3/2.9	0.3/3.7		0.6/3.4	0.7/2.7	

* $\log_{10}$ of heterologous neutralization index/$\log_{10}$ of homologous neutralization index; results are a composite of several tests.

**mouse hyperimmune (4 injections) ascitic fluids.

AVA was also recovered from the blood of 1 of 84 (1.2%) herring gull chicks (Tables 2, 4). This was the only virus isolated from 124 blood samples from five species of birds (Table 4). The viremic chick did not show any obvious signs of illness when captured.

No HI antibody was detected in bird sera tested with antigens of mosquito-borne alphaviruses (EEE, WEE, SIN) or flaviviruses (SLE, JBE, WN) (Table 5). However, several sera did react to titers of 1:10 to 1:40 with tick-borne flaviviruses: RSSE (21.7%), LI (10.0%), TYU (4.1%), and POW (0.8%) Tables 5, 6). The positive reactions were not confirmed by Nt tests with LI or TYUL (Table 7) and were not attempted with RSSE or POW.

Nt antibody was detected in avian sera with GI (19.8%) and BAU (18.3%) (Tables 7, 8). A small number of sera also neutralized DenAr 3 (6.1%) and CW (0.6%); these may represent cross-neutralization by a heterologous antibody since each of the samples also neutralized GI and/or BAU (Table 9). No YH antibody was found (Table 7).

AVA virus was neutralized by 27.6% of the avian sera (Table 7). Virus or antibody was demonstrated in three unfledged gull chicks indicating active transmission on Great Island. CM Nt antibody was not detected although two samples (4.0%) neutralized SAK virus (Table 7); one of the two samples also neutralized AVA and the quantity of the second was insufficient to test further (Table 9).

DISCUSSION

Mosquito-borne togaviruses:

There was no evidence (serological or virological) of alphavirus or mosquito-borne flavivirus activity among any of the seabirds tested in the current survey. Thus our findings lend no support to a hypothesis for the interepidemic maintenance of the North American mosquito-borne encephalitides (EEE, WEE, SLE) which states that active "silent" foci occur in large, stable populations of birds and/or mammals in undeveloped regions north of the epidemic foci, and that virus is disseminated southward.[8]

TABLE 4. Blood samples collected from birds captured on Great Island, Newfoundland during July 1972 and tested for virus or antibody.

Species	Age	Heparinized Blood (Virus)	Serum (Antibody)
herring gulls (*Larus argentatus*)	chicks	84*	28
black-backed gulls (*Larus marinus*)	chicks	2	2
black-legged kittiwakes (*Rissa tridactyla*)	chicks	15	
common puffins (*Fratercula arctica*)	chicks	20	
	adults		135
common murres (*Uria aalge*)	chicks	3	
Leach's storm petrels (*Oceanodroma leucorhoa*)	adults		236
TOTALS		124*	401

*One virus isolation—Avalon—from a herring gull chick

TABLE 5. Hemagglutination-inhibiting antibody in human and avian sera collected on Great Island during July 1972.

Antigens	Atlantic Puffins (adults)	Leach's Petrels (adults)	Herring Gulls (chicks)	Herring Gulls (chicks)	Human (adults)
Eastern Equine Encephalomyelitis	0/103*	0/114			0/8
Western Equine Encephalomyelitis	0/72	0/84			0/8
Sindbis	0/93	0/110			0/8
Japanese B Encephalitis	0/90	0/210	0/2	0/26	0/8
West Nile	0/102	0/108			0/8
St. Louis Encephalitis	0/41	0/107			0/1
Russian Spring-Summer Encephalitis	39/108 (35.8)	36/210 (17.1)	0/2	0/26	0/9
Louping III**	20/90 (22.2)	13/210 (6.2)	0/2	0/26	0/8
Powassan	2/86 (2.3)	0/130	0/2	0/26	0/8
Tyuleniy**	14/108 (13.0)	0/210	0/2	0/21	0/9

* number positive/number tested (percent positive in parentheses)

**not confirmed by neutralization (see Table 7)

Tick-borne togaviruses:

Viruses of the tick-borne encephalitis (TBE) complex were isolated from common murres (*U. aalge*) and seabird ticks (*I. uriae*) in eastern Murmansk on the Barents Sea.[2] Earlier, circumstantial evidence based on serosurveys of thick-billed murres (*U. lomvia*) and black-legged kittiwakes (*R. tridactyla*) suggested the presence of *I. uriae*-transmitted TBE virus.[6] The present study indicates a similar focus among seabirds in the western North Atlantic. Group B tick-borne encephalitis viruses have not yet been isolated from birds in North America.

TYU, another group B virus recovered from *I. uriae* in eastern USSR[13,14] and western USA[4] in the North Pacific, was not encountered by virus isolation or serologic evidence on Great Island.

Orbiviruses:

At least six serotypes of Kemerovo group viruses have been reported from *I. uriae* throughout the world; GI, BAU, and CW from the North Atlantic,[17,19] YH and Okhotskiy from the North Pacific[10,26] and NUG from the South Pacific.[5] Serologic evidence of the presence of the two indigenous strains—GI and BAU—was demonstrated among puffins and petrels on Great Island, with little or no indications of the other serotypes. Although transoceanic flights—St. Kilda, Scotland to Newfoundland, Canada—of two banded puffins have been reported,[11,25] there is little evidence to indicate that alcids of different rookeries interact in colonies.[12,24,25] The integrity of sub-specific races[22] tends to support this hypothesis. The antigenic identity of the serotypes may, therefore, be maintained by the isolation of the primary hosts of the vector. It has been reported that *I. uriae* does not attach to birds late in the nesting season, when birds begin moving out to sea.[10] The occasional dispersal of the viruses and/or ticks by primary (alcids) or secondary (gulls, terns, etc.) hosts may account for the widespread distribution of the tick species[7] and the serogroup. If this hypothesis is correct, the immune status may be a useful marker that can be found as a supplement to banding in studies on the movements of alcids.

TABLE 6. Results of hemagglutination-inhibition tests on avian sera with both Russian spring-summer encephalitis (RSSE) and louping ill antigens.

Puffin

		Louping ill		
		positive	negative	Total
RSSE	positive	10 (15, 19)*	27 (10, 0)	37
	negative	10 (0, 12)	43 (0, 0)	53
	Total	20	70	90

Petrels

		Louping ill		
		positive	negative	Total
RSSE	positive	9 (18, 19)	27 (13, 0)	37
	negative	5 (0, 22)	169 (0, 0)	174
	Total	14	196	210

* Reciprocal of mean titers of Russian spring-summer encephalitis and louping ill, respectively, in parentheses.

TABLE 7. Neutralizing antibody in human and avian sera collected on Great Island during July 1972.

Virus	Atlantic Puffins (adults)	Leach's Petrels (adults)	Black-backed Gulls (chicks)	Herring Gulls (chicks)	Human (adults)
Kemerovo Group Viruses					
Great Island (CanAr 41)	44/119 (37.0)*	6/104 (5.8)	0/2	0/28	0/9
Bauline (CanAr 14)	47/126 (37.3)	5/128 (3.9)	0/2	0/28	0/9
Cape Wrath (ScotAr 20)	0/66	1/64 (1.6)	0/2	0/25	0/8
———— (DenAr 3)	4/11 (36.4)	0/31	0/1	0/23	0/1
Yaquina Head (USA 15)	0/63	0/35	0/2	0/23	0/9
Nugget (AusMI-14847)	0/40	1/14 (7.1)	0/2	0/24	0/5
Sakhalin Group Viruses					
Avalon (CanAr 173)	18/29 (62.1)	12/63 (19.0)	0/1	2/23 (8.7)	0/6
Clo Mor (ScotAr 7)	0/38	0/78	0/2	0/25	0/6
Sakhalin (USSR LEIV 71c)	2/50 (4.0)				0/9
Group B					
Tyuleniy (USSR LEIV 6c)	0/49				0/9
Louping Ill (Moredun)	0/45	0/27	0/2	0/2	

* Number positive/number tested (percent positive in parentheses)

TABLE 8. Results of neutralization tests on avian sera with both Great Island and Bauline viruses.

Puffin

		Great Island		
		positive	negative	Total
Bauline	positive	23	21	44
	negative	21	51	72
	Total	44	72	116

Petrels

		Great Island		
		positive	negative	Total
Bauline	positive	3	0	3
	negative	3	90	93
	Total	6	90	96

TABLE 9. Serological profiles of avian sera which neutralized viruses not known to be indigenous on Great Island.

	VIRUS								
SERA	Great Island (CanAr 41)	Bauline (CanAr 14)	Cape Wrath (ScotAr 20)	N (DnnAr 3)	Yaquina Head (USA 15)	Nugget (AusMI-14847)	Avalon (CanAr 173)	Clo Mor (ScotAr 7)	Sakhalin (LEIV 71c)
Puffin									
# 306	+	—	—	+	0	—	+	—	0
# 310	—	+	—	+	0	—	+	—	0
# 329	+	+	—	+	—	—	—	—	0
# 330	+	—	—	+	0	—	+	—	0
# 121	+	+	0	0	—	0	0	0	+
# 143	0	+	0	0	0	0	+	0	+
Petrels									
# 22	+	—	+	0	0	+	+	0	0

— = negative
+ = positive
0 = not tested

Sakhalin group viruses:

Sakhalin group viruses have been reported from the North Atlantic,[18] North Pacific[15,23] and South Pacific.[5] AVA was recovered from both ticks and birds on Great Island. Serologic evidence suggests that this agent may be more prevalent than virus isolations indicate. Resistance of suckling mice to infection by AVA may be responsible for low rates of virus isolations. There was no conclusive evidence of serotypes from the eastern Atlantic (CM) and the North Pacific (SAK) among the birds on Great Island.

Differences in prevalence (antibody) rates of AVA, as well as GI, BAU, and RSSE, in puffins and petrels probably reflect, in part at least, the relative rates of attack by *I. uriae* due to habitat differences of the two avian species. The majority of petrels nest in burrows beneath stunted conifers—balsam fir (*Abies balsamea*) and black spruce (*Picea mariana*)—toward the interior of the island, while puffins prefer the peripheral grass slopes;[21] some overlap occurs at the interface. The restricted habitat requirements for *I. uriae*[9,39] were most frequently encountered on or near the puffin territories.

The relative susceptibility of puffins and petrels may also be a factor. Acquired immunity among the birds indicate survivors, but little is known about the effects of infection on birds, especially secondary or aberrant hosts or on man. Although there was no evidence of infection in the few biologists sampled, *I. uriae* will engorge on man[1,12] and antibody against one of these viruses (TYU) or a related virus was reported in residents of the Commodore Islands.[14]

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LITERATURE CITED

1. ARTHUR, D. R. 1963. *British Ticks*. Butterworths, London 213 p.
2. BERKLESHOVA, A. Yu., I. I. TERSKIKH, E. N. BYCHKOVA and V. A. SMIRNOV. 1969. A natural focus of tick-borne encephalitis and ornithosis in the extreme north. V-th symposium on the study of the role of migrating birds in the distribution of arboviruses (Abstract of Papers). p. 100. Academy of Sciences of the USSR. Siberian Branch. Novosibirsk.
3. CLARKE, D. and J. CASALS. 1958. Techniques for hemagglutination and hemagglutination-inhibition with arthropod-borne viruses. Am. J. trop. Med. Hyg. 7: 561-573.
4. CLIFFORD, C. M., C. E. YUNKER, L. A. THOMAS, E. R. EASTON and D. CORWIN. 1971. Isolation of a group B arbovirus from *Ixodes uriae* collected on Three Arch Rock National Wildlife Refuge, Oregon. Am. J. trop. Med. Hyg. 20: 461-468.
5. DOHERTY, R. L., J. G. CARLEY, M. D. MURRAY, A. J. MAIN, B. H. KAY and R. DOMROW. 1975. Isolation of arboviruses (Kemerovo group, Sakhalin group) from *Ixodes uriae* collected at Macquarie Island, Southern Ocean. Am. J. trop. Med. Hyg. 24: 521-526.
6. FLINT, V. E., V. A. SMIRNOV and A. A. PCHELKINA. 1965. Serological investigations of bird colonies in northeast Murmansk. Mater. 4 Vres. Ornit. Konf. (Alma Ata, 1-7 Sept. 1965), p. 395-397. (In Russian) (In English: NAMRU 3-T418).

7. HOOGSTRAAL, H. 1973. Viruses and Ticks. Ch. 18 In: *Viruses and Invertebrates,* A. J. Gibbs (ed.). Frontiers of Biology, Vol. 31 North Holland Publishing Company, Amsterdam.

8. JOHNSON, H. N. 1960. Encephalitis and Research. Proc. and Papers of 28th Annual Conf. Calif. Mosq. Cont. Assoc. p. 11-16.

9. KARPOVICH, V. N. 1970. (Properties of *Ceratixodes putus* Pick-Camb, parasitism on birds). Parazitologiya, Leningrad, 4: 345-351 (English summary).

10. KARPOVICH, V. N. 1973. (Life cycle of *Ceratixodes putus* Pick-Camb. in Murmansk conditions). Parazitologiya, Leningrad 7: 128-133.

11. LEACH, E. P. 1940. Puffin ringed at St. Kilda reported in Newfoundland. British Birds 33: 281.

12. LOCKLEY, R. M. 1953. *Puffins.* Devin-Adair Company. New York. 186 pp.

13. LVOV, D. K., A. A. TIMOFEEVA, V. I. CHERVONSKI, V. L. GROMASHEVSKI, G. A. KLISENKO, G. V. GOSTINSHCHIKOVA and I. N. KOSTRYKO. 1971. Tuleniy virus—A new group B arbovirus isolated from *Ixodes* (*Ceratixodes*) *putus* Pick-Camb. 1878 collected on Tuleniy Island, Sea of Okhotsk. Am. J. trop. Med. Hyg. 20: 456-460.

14. LVOV, D. K., A. A. TIMOFEEVA, V. L. GROMASHEVSKI, A. G. POGREBENDO, V. I. CHERVONSKI, A. I. GROMOV, Yu.M. TSYRKIN, A. S. ZEMIT, G. V. GOSTINSHCHIKOVA, G. N. LEONOVA, A. A. SAZONOV, V. ZHEZMER, V. M. POLIVANOV, T. I. CHUNAKHINA, N. P. BELIKOVA, Yu.V. SHABAEV, V. I. SHESTAKOV, A. G. YUDAKOV and B. I. LEBEDEV. 1972. On ecology of Tyuleniy and Sakhalin virus in the Far East. (In Russian, English summary). Mater. Simp. Itogi 6. Simp. Izuch. Virus Ekol. Svyazan. Ptits. (Omsk, 7-9 Dec. 1971), pp. 46-52.

15. LVOV, D. K., A. A. TIMOFEEVA, V. L. GROMASHEVSKI, Y. M. TSYRKIN, A. A. SAZNOV, A. G. POGREBENKO, G. A. SIDOROVA, G. V. GOSTINSHCHIKOVA, N. V. KHUTORETSKAYA, V. I. GUSHCHA, K. B. FOMINA, V. I. CHERVONSKI, V. A. ARISTOVA, G. N. LEONOVA, V. Y. ZHEZMER, Y. V. SHIBAEV, A. G. YUDAKOV, F. P. FILATOV and V. M. CHUMAKOV. 1974. The ecology of Sakhalin virus in the north of the Far East of the USSR. J. Hyg., Epid., Microbiol., Immunol. 18: 87-95.

16. LVOV, D. K., A. A. TIMOFEEVA, V. L. GROMASHEVSKI, Y. M. TSYRKIN, O. V. VESELOVSKAYA, G. V. GOSTINSHCHIKOVA, N. V. KLUTORETSKAYA, A. G. POGREBENKO, V. A. ARISTOVA, A. A. SAZONOV, V. I. CHERVONSKI, G. A. SIDOROVA, K. B. FOMINA and V. Y. ZHEZMER. 1973. "Okhotskiy" virus, a new arbovirus of the Kemerovo group isolated from *Ixodes* (*Ceratixodes*) *putus* Pick-Camb. 1878 in the Far East. Arch. ges. Virusforsch. 41: 160-164.

17. MAIN, A. J., W. G. DOWNS, R. E. SHOPE and R. C. WALLIS. 1973. Great Island and Bauline: Two new Kemerovo group orbiviruses from *Ixodes uriae* in eastern Canada. J. Med. Ent. 10: 229-235.

18. MAIN, A. J., W. G. DOWNS, R. E. SHOPE and R. C. WALLIS. 1976. Avalon and Clo Mor: Two new Sakhalin group viruses from the North Atlantic. J. Med. Ent. 12: *in press.*

19. MAIN, A. J., R. E. SHOPE and R. C. WALLIS. 1976. Cape Wrath: A new Kemerovo group orbivirus from *Ixodes uriae* in Scotland. J. Med. Ent. 12: *in press.*

20. MURRAY, M. D. and W. J. M. VESTJENS. 1967. Studies on the ectoparasites of seals and penguins. III. The distribution of the tick *Ixodes uriae* White and the flea *Parapsyllus magellanicus heardi* deMeillon on Macquarie Island. Austral. J. Zool. 15: 715-725.

21. NETTLESHIP, D. N. 1972. Breeding success of the common puffin (*Fratercula arctica* L.) on different habitats at Great Island, Newfoundland. Ecological Monographs 42: 239-268.

22. SALOMONSEN, F. 1944. The Atlantic Alcidae. The seasonal and geographical variation of the auks inhabiting the Atlantic Ocean and the adjacent waters. Sjatte foljder, Ser. B., Vol. 3, No. 5 pages 1-138.

23. THOMAS, L. A., C. M. CLIFFORD, C. E. YUNKER, J. E. KIERANS, E. R. PATZER, G. E. MONK and E. R. EASTON. 1973. Tickborne viruses in western North America. I. Viruses isolated from *Ixodes uriae* in coastal Oregon in 1970. J. Med. Ent. 10: 165-168.

24. TUCK, L. M. 1961. The Murres. Canadian Wildlife Series 1, 260 p.

25. TUCK, L. M. 1971. The occurrence of Greenland and European birds in Newfoundland. Bird-Banding 42: 184-209.

26. YUNKER, C. E., C. M. CLIFFORD, J. E. KEIRANS, L. A. THOMAS and J. CORY. 1973. Tickborne viruses in western North America. II. Yaquina Head, a new arbovirus of the Kemerovo group isolated from *Ixodes uriae*. J. Med. Ent. 10: 264-269.

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