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Rabies-free status of the Czech Republic after 15 years of oral vaccination

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Summary
Rabies in foxes was widespread in the Czech Republic after World War II, reaching its highest incidence in the 1980s. Applied control measures had only a limited effect and rabies incidence in wildlife continuously endangered both domestic animals and human beings. A significant improvement was noticed after the introduction of oral vaccination of foxes in 1989. The original manual distribution of vaccine baits has been replaced by aerial distribution, leading to the total elimination of rabies throughout the country. The last case of rabies was diagnosed in a fox in the district of Trutnov in April 2002. Since that time no case of rabies has been registered in the Czech Republic, and it therefore fulfils the requirements for the status of a 'rabies-free country'. Effective epidemiological surveillance and preventive oral vaccination of foxes will be necessary to maintain this status, especially in the endangered border regions.

Keywords
Control strategy – Czech Republic – Epidemiology – Oral vaccination – Rabies – Red fox.

Introduction

Rabies is undoubtedly one of the most serious and deadly zoonoses that threaten human populations. According to global figures from the World Health Organization (WHO), there are between 30,000 and 50,000 human victims every year. Various species of carnivores cause its spread and persistence. One report on rabies vaccination reported that 'Given its distribution, veterinary and public health consequences, and the highest case fatality rate of any infectious disease, rabies remains a significant global problem' (10). The typical pattern of sylvatic rabies has been present for a long time in the European continent and the red fox (*Vulpes vulpes)* has been the principal animal reservoir for decades (8). Oral vaccination of red foxes against rabies was a significant factor in the elimination of rabies; it started at the beginning of the 1980s in Switzerland and Germany and then expanded into other European countries (11, 12, 13).

Rabies in Europe and in the Czech Republic

The sylvatic rabies infection wave among red foxes developed during World War II. During the 1950s and 1960s rabies struck many European countries and it is still present in some of them today (15). The epidemiology of rabies has been studied systematically and in detail since 1977, when the WHO Collaborating Centre for Rabies Surveillance and Research was established in Tübingen in Germany. The Centre (now based in Wusterhausen) publishes *Rabies Bulletin Europe*, which contains statistical data concerning rabies occurrence within animal and human populations. This publication provides a highly reliable and functional system of collecting and processing information concerning rabies outbreaks. It reliably tracks the currently observed incidence and geographical localisation of rabies cases. Also, it tracks the progress of

contagion in the European continent, and helps to coordinate protective measures. The evolution of rabies epidemiology and the development of oral vaccination techniques in 1998 have been described (3), and are reiterated here with an emphasis on the culmination of the process, resulting in the elimination of rabies in the Czech Republic.

In the Czech Republic, the first rabies-positive red fox was diagnosed in the district of Broumov at the Czech-Polish border in March 1947 (9). A significant increase in rabies incidence was recorded in the following year, when 146 cases were recorded in total, 106 (74%) of which were in red foxes (9). Rabies was at its most widespread in the 1980s. At that time, with the exception of a few districts, rabies was spread over the entire territory of the Czech Republic. The greatest occurrence was recorded in 1984, with 2,232 cases, 2,052 of which were in red foxes. In spite of a decrease of rabies cases in the following years, the situation remained unfavourable. Neither incentives for red fox hunting (established in 1969) nor gassing of dens brought improvement. The unfavourable disease situation and permanent disease reservoir in wild animals continued to raise rabies incidence among domestic animals.

Rabies control by reducing the red fox population

In line with the trends in neighbouring countries, the control measures used in the Czech Republic in the 1960s were designed to reduce the red fox population (14). The starting point for this approach was a Danish investigation, which showed that a reduction of the red fox population density to the optimal one red fox per 500 ha, could interrupt the continual spread of rabies (7). For this reason, in 1969, the government began to offer 100 Czech Crowns for every hunted red fox handed over to the Veterinary Services. The idea behind this action was to increase an interest in hunting, ensure samples for rabies monitoring among the red fox population, discover new outbreaks and pinpoint the topography and dynamics of the disease. After 1977 the incentive was increased to 150 Czech Crowns and at the same time a programme of den fumigation was implemented. In 1979 fumigation began to be replaced by gassing by means of preparations that released phosphorus hydrogen, but this proved to be no more successful than encouraging hunting (5).

Moreover, poisoning fox cubs (*Vulpes vulpes*) with gas was not in compliance with the ethics of traditional hunting. It had widely varying effects locally, and did not lead to a reduction in rabies incidence anywhere in the country. Paradoxically, the reduction of population density in certain areas encouraged increased migration of red foxes from surrounding areas, leading to a rapid return to high numbers of foxes in these areas. Also, provisionally permitted poisoning by strychnine baits proved ineffective; moreover, it was unselective and threatened other free-ranging animals and even humans. Despite the introduction of these measures to reduce the fox population, disease prevalence remained high, reaching a peak in the 1980s.

Oral vaccination

The introduction of oral immunisation for foxes (1) was a considerable step forward in rabies control in wildlife and significantly contributed to the improvement of the disease situation in many European countries (2, 12, 13). The programme of oral vaccination of foxes against rabies was launched in the Czech Republic in 1989.

Vaccine

The vaccine of German origin (Tübingen), prepared from the attenuated strain SAD-B19 (11) was used in the first seven campaigns of the rabies elimination programme. In 1991, cooperation with the Swiss Rabies Centre of the Institute of Veterinary Virology at the University of Bern was established. The vaccine strain SAD Berne, an appropriate cell culture (BHK-21 cells) and a machine for producing and filling plastic containers were obtained from this department. This meant that specialists from the Czech Veterinary Services were then equipped with all the facilities, information and know-how that made the oral vaccination programme in Switzerland such a success (12, 13). In 1992, the veterinary pharmaceutical company Bioveta Ivanovice na Hané successsfully launched the production of its own live virus vaccine based on the SAD Berne strain. The vaccine baits consisted of a mixture of fat, fish, meat, and bone meal. The Czech vaccine Lysvulpen was financially advantageous and after a field trial in the autumn of 1992 this vaccine completely replaced the one originally imported.

Distribution and baiting density

When oral vaccination was first introduced, only the Bavarian model of bait distribution was used (11). This involved members of voluntary hunting organisations distributing baits twice a year. Spring (April) and autumn (October) were chosen as the most suitable periods for vaccination due to the weather conditions. Originally, 15 baits were laid per km², then the number of baits manually distributed gradually increased, especially in regions where the number of disease cases was particularly high. Aerial distribution of baits was introduced in limited

areas in 1996 and the number of districts treated in this way gradually increased over the next six years. In the autumn campaign of 1996, 89,600 baits were dropped on an area of 3,520 km². The effectiveness of both manual and aerial distribution was determined using a tetracycline marker (TTC). In five districts where only manual distribution was used during 11 campaigns between 1997 and 2004, 79.54% of 1,982 foxes tested TTC positive. In five other districts that were treated primarily aerially during the same time period, 86.53% of 2,808 foxes tested TTC positive. The tetracycline rates recorded after respective campaigns are shown in Figure 1.

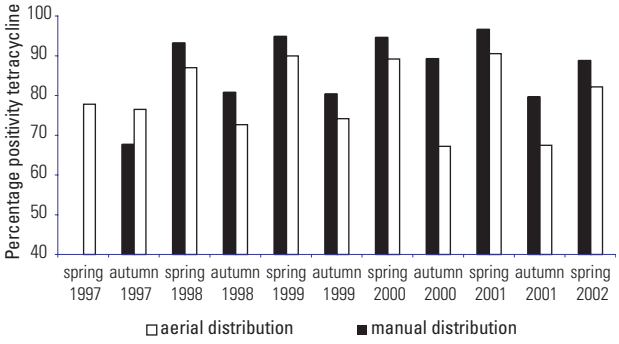

Fig. 1
Tetracycline marking after manual and aerial distribution of baits, 1997 to 2002

Aerial distribution has proven to be more effective than manual distribution and its use has been extended to the entire treated area. Almost 25 million baits were used from the spring of 1989 to the autumn of 2004. Fifteen to twenty baits per km² were used for manual distribution and 25 to 31 baits per km² for airplane distribution. Details are presented in Table I. The annual figures presented include data for both the spring and autumn campaigns.

Treated territory and vaccination strategy

The extension of the vaccinated area over several selected years is presented in Fig. 2. The first application of Tübingen oral vaccine was carried out in the spring of 1989 in the districts of Klatovy, Domazlice and Tachov, which are adjacent to the German border (6). In the course of subsequent campaigns, the treated area was extended, and by the autumn of 1992 it covered 44 districts. In the autumn of 1993 the whole territory of the Czech Republic, with the exception of the rabies-free districts bordering Germany, was included. Over subsequent years the strategy of oral vaccination was based on two basic principles:

a) intensive treatment in districts affected with rabies

b) continuation of oral immunisation for at least two years after the last outbreak.

Table I
Oral vaccination in the Czech Republic, 1989 to 2004: manual and aerial distribution of vaccine baits

Year	Size of vaccinated area (km²)		Total number of baits		Baits per km²		Type of vaccine
	Manual distribution	Aerial distribution	Manual distribution	Aerial distribution	Manual distribution	Aerial distribution	
1989	5,440		81,600		15.0		Tübingen
1990	29,040		435,600		15.0		Tübingen
1991	37,820		567,800		15.0		Tübingen
1992	82,000		1,235,000		15.1		Tübingen/Lysvulpen
1993	94,750		1,541,600		16.3		Lysvulpen
1994	92,350		1,489,400		16.1		Lysvulpen
1995	95,800		1,649,600		17.2		Lysvulpen
1996	93,180	3,520	1,597,800	89,600	17.1	25.5	Lysvulpen
1997	90,280	7,020	1,570,000	220,000	17.4	31.3	Lysvulpen
1998	53,950	8,550	1,105,000	225,000	20.5	26.3	Lysvulpen
1999	56,468	14,872	1,085,000	371,800	19.2	25.0	Lysvulpen
2000	53,090	29,980	956,600	749,600	18.0	25.0	Lysvulpen
2001	60,592	30,816	1,090,800	768,900	18.0	25.0	Lysvulpen
2002	52,166	56,228	937,800	1,410,500	18.0	25.1	Lysvulpen
2003	866	108,612	15,600	2,725,100	18.0	25.1	Lysvulpen
2004	866	113,650	15,600	2,847,800	18.0	25.1	Lysvulpen
Total	**898,658**	**373,248**	**15,374,800**	**9,408,300**	**17.1 (average)**	**25.2 (average)**	

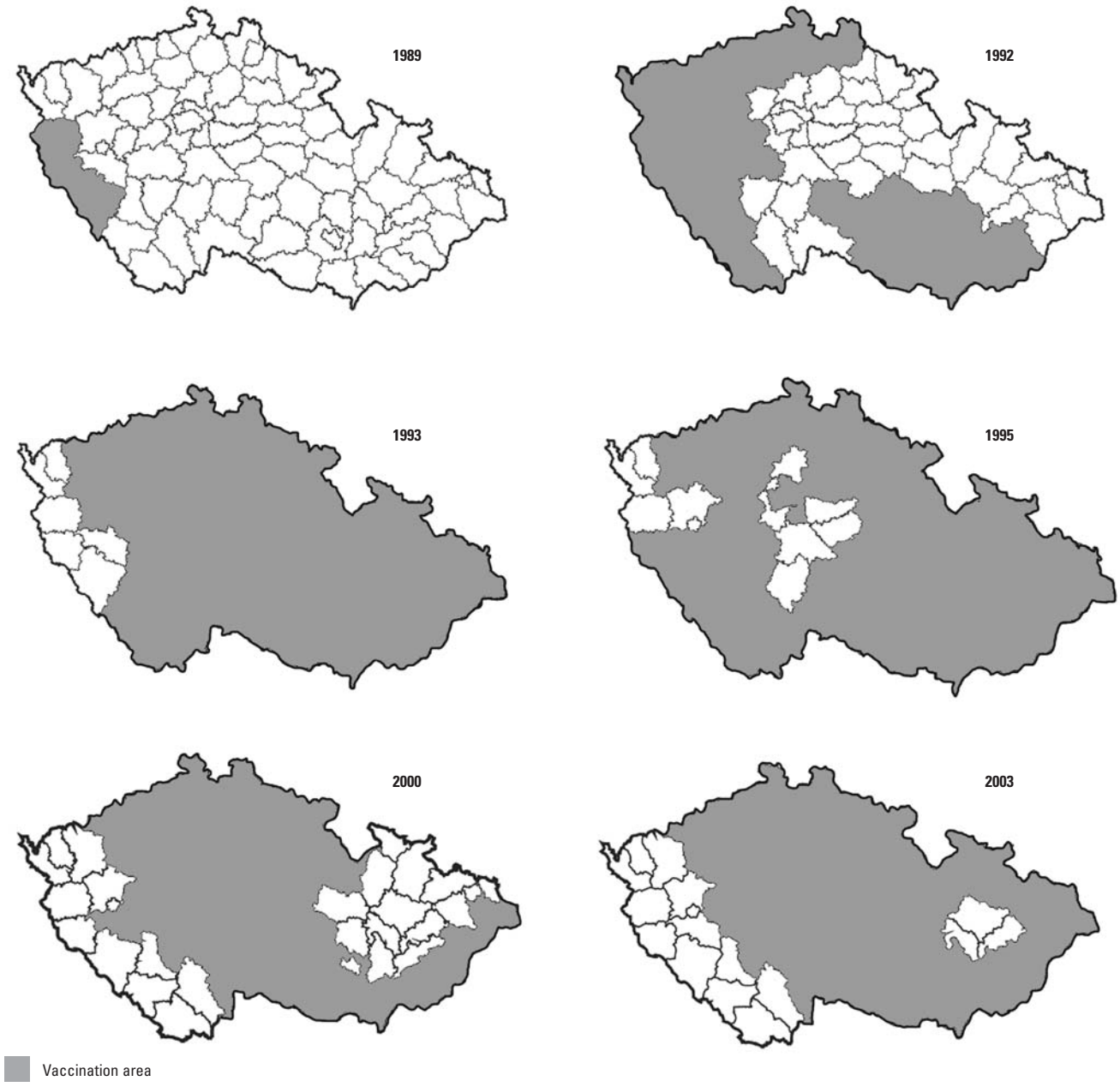

Fig. 2
Oral vaccination of foxes in the Czech Republic
Over the years the vaccination area was extended and vaccination campaigns increased

The oral vaccination strategy used was not always optimal, especially in the period between 1995 and 2001. Due to limited financial resources vaccination was implemented in only a few administrative units (districts); in other districts oral vaccination was stopped prematurely and untreated holes were created in the middle of vaccinated areas. These gaps sometimes became sources of new outbreaks and reoccurrence of disease and consequently it became necessary to re-launch vaccination campaigns in these areas. Owing to this fact, the whole process of rabies elimination was complicated, delayed and, therefore, increasingly expensive. The reduced epidemiological awareness and ineffective monitoring in originally rabies-free areas outside vaccination zones often led to the reoccurrence of the disease. A high population density of unvaccinated red foxes facilitated this reoccurrence, leading to an adverse disease situation near the border and in the territory of neighbouring countries. A typical example was rabies reoccurrence in the previously rabies-free area located in the vicinity of the Polish border in 2000 (4). Subsequently, animals in other areas of contiguous territory were vaccinated and a consistent

strategy applied. Only those districts rabies free for more than two years were omitted, as can be seen in the map for 2003 in Figure 2.

Monitoring the effectiveness of oral vaccination

Follow-up investigations were organised by the National Reference Laboratory for Rabies and were directed towards bait uptake, tetracycline marking, antibody formation, rabies diagnosis and characterisation of isolated strains by monoclonal antibodies.

Bait uptake was assessed by hunters and varied between 55% and 88.8%, with an average of 82.3% during all campaigns. In addition, as an indirect but more precise measurement of bait uptake, fox bones were examined for tetracycline incorporation. Tetracycline was found in 65% to 80% of examined samples, with an average of 78%, with considerable differences in particular areas. Antibody formation in the body fluids of foxes was tested by rapid fluorescent focus inhibition test (RFFIT). Samples of body fluids were taken from the chest cavities of fox cadavers, and were composed of blood and transudate. Rabies antibodies were detected in 50% to 78% of examined samples.

As recommended by the WHO, after each campaign, wildlife specimens were collected within vaccination areas for examination. Over 50,000 animals were examined and a total of 3,500 cases of rabies were found over the course of the 32 campaigns. All viruses isolated were characterised as street virus strains by monoclonal antibodies.

Table II and Figure 3 show that since the launch of the vaccination programme in 1989, the incidence of rabies in the Czech Republic has exhibited a pronounced tendency to decline. A series of maps in Fig. 4 shows the location of rabies cases and the gradual reduction in their numbers during the course of the vaccination campaigns. By 1995, the overall number of positive cases had decreased by 88% since the beginning of the campaign. This positive trend has continued, with minor variations in subsequent years. In 2001 only 35 rabies cases were recorded and in 2002 this number had dropped to three. The last rabies case was found in April of 2002 in a red fox in the district of Trutnov. Since that time there has not been a single case of rabies diagnosed. The Czech Republic thereby fulfilled the criteria for recognition as a rabies-free state and enlarged the list of rabies-free countries which obtained this status using oral vaccination programmes (2). This event was documented in the on-line publication of the World Organisation for Animal Health (OIE) *'Disease Information'* No. 30, 23 July 2004. The Czech Republic has remained rabies-free ever since. This undisputed success is a great incentive to continue fulfilling the strict criteria for maintaining this status. Proper surveillance in all areas is part of the criteria, focusing on any suspicious cases and the identification of new infection foci. A territory can be declared rabies-free if no incident of rabies is found within two years of the last incident. A continual rabies-free status must be documented by proper monitoring. The WHO requires the annual examination of at least eight foxes per 100 km². The disease situation can be objectively evaluated only after this required number of examinations have taken

Table II
Rabies cases per species in the Czech Republic, 1989 to 2004

Year	Number of cases per species				Total
	Dog	Cat	Fox	Others	
1989	10	45	1,369	77	1,501
1990	9	34	1,046	68	1,157
1991	8	30	1,044	72	1,154
1992	7	14	526	23	570
1993	2	19	359	42	422
1994	6	5	191	19	221
1995	2	5	157	14	178
1996	0	3	223	11	237
1997	0	6	224	8	238
1998	1	3	77	4	85
1999	1	3	192	18	214
2000	2	3	142	18	165
2001	0	2	29	4	35
2002	0	0	3	0	3
2003	0	0	0	0	0
2004	0	0	0	0	0
Total	**48**	**172**	**5,582**	**378**	**6,180**
Percentage	**0.8**	**2.8**	**90.3**	**6.1**	**100**

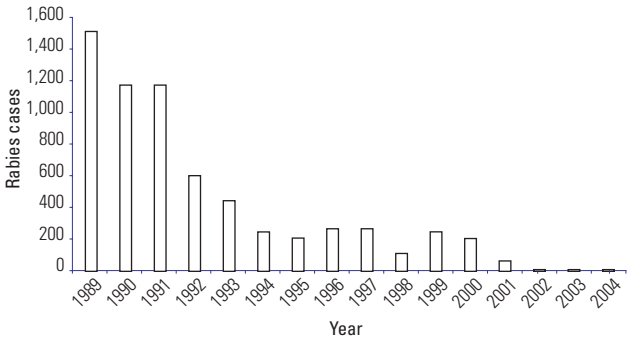

Fig. 3
Rabies cases in the Czech Republic, 1989 to 2004
From 1989 there was a significant decline in rabies cases and the country became rabies-free in 2003

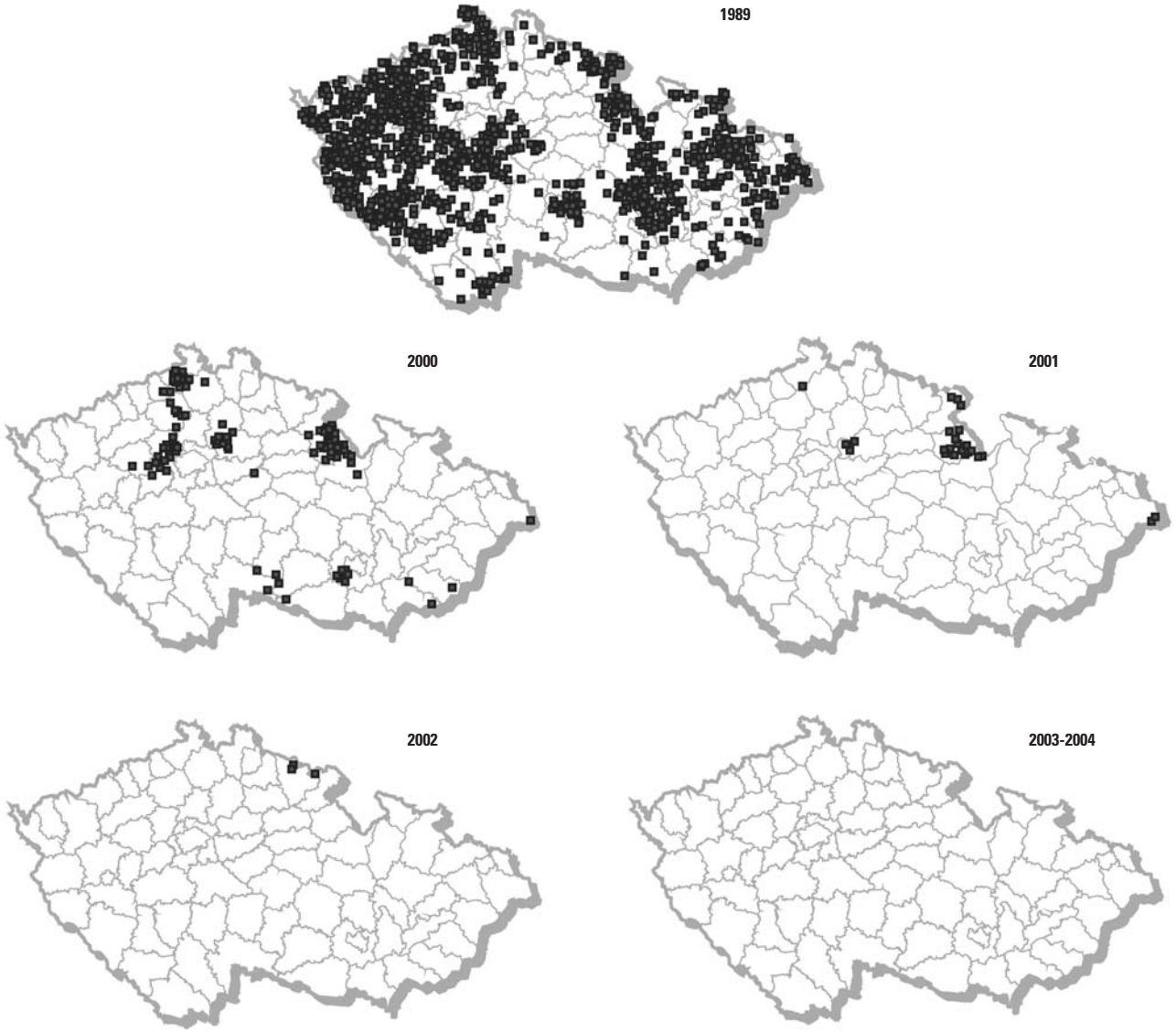

Fig. 4
Geografical distribution of rabies cases in the Czech Republic, 1989 to 2004
The number of rabies cases fell from 1,501 in 1989 to 3 in 2002

place in the region (the minimum area that can be declared rabies-free is 5,000 km²). The number of control samples must be distributed equally over the entire area. Examining areas adjoining those infected, e.g. the northern and eastern borders of the Czech Republic, is most important. The number of samples taken each year in the Czech Republic exceeds requirements, and typically surpasses ten foxes per 100 km², as documented in Table III.

Table III
Number of foxes examined annually in the Czech Republic, 1999 to 2004

Year	Total number of foxes examined	Number of foxes examined per 100 km²
1999	6,411	10.6
2000	5,281	8.8
2001	6,607	11.0
2002	5,812	9.7
2003	6,248	10.4
2004	7,164	11.9

Conclusion

The elimination of rabies in the Czech Republic is undoubtedly significant progress towards fulfilling the

ultimate goal of lowering, or rather eliminating, rabies risk to animal and human populations throughout Europe. The number of post-exposure vaccinations and treatments should decrease in the future, and greater ease of animal shipment should simplify international commerce. On the other hand, the current status requires continuous rabies prevention. The significantly increasing fox population density in rabies-free areas provides good conditions for potential re-infection and renewed contagion. So, it is expected that oral vaccination of foxes will continue for several years, at least in areas bordering on countries to the north and east, where there is still an unacceptably infectious rabies situation. Furthermore, there must be continued co-operation with hunters, since fox hunting remains the primary population density regulation mechanism, and an important source of laboratory samples. In order to declare the absence of rabies, about six thousand foxes must be examined every year in the Czech Republic. It is important to enlist the co-operation of all parties interested in maintaining the country's rabies-free status in future years.

La République tchèque indemne de rage après 15 ans de vaccination orale

O. Matouch, J. Vitasek, Z. Semerad & M. Malena

Résumé
Depuis la deuxième guerre mondiale, la rage vulpine était très répandue en République tchèque, le pic d'incidence se situant dans les années 1980. Les premières mesures appliquées pour lutter contre la rage n'ont pas eu l'effet escompté et l'incidence de la maladie chez les animaux sauvages a continué de menacer les animaux domestiques et l'homme. Une amélioration significative a été constatée à partir de 1989, date à laquelle la vaccination orale des renards a commencé à être pratiquée. La distribution des appâts vaccinaux, au sol dans un premier temps, s'est faite ensuite par avion, ce qui a permis d'éliminer totalement la rage de l'ensemble du territoire national. Le dernier cas de rage a été diagnostiqué dans le district de Trutnov en avril 2002. Aucun cas de rage n'ayant été observé en République tchèque depuis cette date, le pays remplit les conditions prescrites pour être reconnu indemne de rage. Il sera nécessaire de poursuivre une surveillance épidémiologique appropriée ainsi qu'une vaccination orale préventive des renards pour maintenir ce statut indemne, en particulier dans les régions frontalières vulnérables.

Mots-clés
Épidémiologie – Rage – Renard roux – République tchèque – Stratégie de lutte – Vaccination orale.

Tras 15 años de vacunación oral, la República Checa libre de rabia

O. Matouch, J. Vitasek, Z. Semerad & M. Malena

Resumen
En los años 80, la rabia del zorro, que estaba muy extendida en la República Checa después de la Segunda Guerra Mundial, alcanzó su incidencia máxima. Los efectos de las medidas de control eran limitados y la incidencia de la enfermedad en la fauna salvaje amenazaba constantemente a animales domésticos y seres humanos. En 1989, tras la introducción de la vacunación oral

de los zorros, la situación experimentó una mejora significativa. En un inicio, se diseminaba los cebos con la vacuna en forma manual. Posteriormente, se procedió a lanzarlos desde el aire, lográndose la eliminación total de la rabia en todo el país. El último caso se diagnosticó, en abril de 2002, en un zorro del distrito de Trutnov. No volvió a registrarse caso alguno de la enfermedad desde esa fecha. Por consiguiente, la República Checa cumple los requisitos para que se la reconozca "país libre de rabia". A fin de mantener ese estatus, será preciso aplicar una vigilancia epidemiológica eficaz y administrar vacunas orales a los zorros con carácter preventivo, en particular, en las peligrosas regiones fronterizas.

Palabras clave
Epidemiología – Estrategia de control – Rabia – República Checa – Vacunación oral – Zorro común.

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