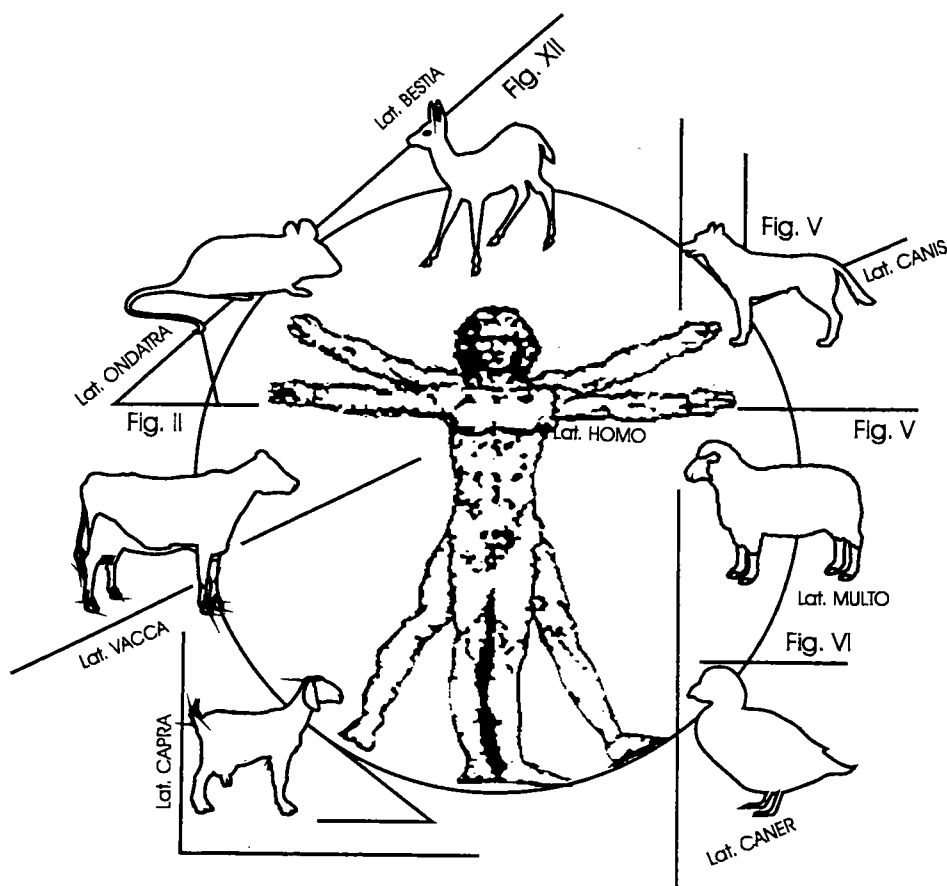


WHO MEETING ON COOPERATION ON ORAL IMMUNIZATION OF FOXES AGAINST RABIES IN 1995 (CENTRAL AND EASTERN EUROPE)

BUDAPEST, HUNGARY
31 MARCH TO 1 APRIL 1995



WORLD HEALTH ORGANIZATION
VETERINARY PUBLIC HEALTH UNIT

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1. INTRODUCTION

As rabid foxes do not respect national borders and, as areas to be vaccinated should be as large as possible, representatives of six countries (Belgium, France, Germany, Luxembourg, Netherlands, Switzerland) decided at a WHO Workshop on Oral Immunization of Wildlife against Rabies in Europe (INTORAL) in 1986 in Tübingen, Germany, to have a common strategy for oral vaccination (OV) of foxes against rabies along their borders. In the meantime these countries, calling their WHO supported initiative "European Border Cooperation - West", met eight times in annual meetings to present their results during the past year and plans for the forthcoming ones.

In 1990, a similar initiative was started for the "European Border Cooperation - East" with countries of middle and eastern Europe. Meetings have been held in Vienna, Prague and Pulawy (Poland).

Of 20 countries invited, 18 sent representatives.

The Honorable Minister of Agriculture in Hungary, Dr L. Lakos, opened the meeting. He was pleased that OV was having such a good impact on the control of rabies in Europe and that Hungary, in particular, had made some modest progress since it started in 1992.

The Minister elaborated on the difficult economic situation in eastern Europe compared to the west and expressed his wish that resources be found, by national authorities as well as from international organizations to enlarge the OV programmes in order to move ahead faster.

Dr W. Müller of the WHO Collaborating Centre for Rabies Surveillance and Research, Tübingen, welcomed the participants (list: Annex 1) and opened the

workshop on behalf of WHO, Veterinary Public Health unit, Geneva, and conveyed the greetings of Dr K. Stöhr who was unable to attend. He thanked the Honorable Minister and the Hungarian colleagues for organizing the meeting and welcomed Dr J. Caffrey of the European Union (EU).

The objectives of the meeting were to present reports of the participating countries on the progress of rabies control (Annex 2: planned OV for 1995) and on future plans:

- follow-up on the suggestions made during the previous meeting in Pulawy, Poland, January 1994 (strategy of vaccination campaigns, laboratory tests in relation to OV and administrative aspects of implementation); and
- discuss how to deal with fox populations when OV has failed in its effects.

Dr Caffrey explained the contribution of the EU in regard to the OV scheme (see details under 2.4) and elaborated on the new geographical situation of the EU in as much as more bordering countries could now participate in the scheme and profit from its support.

According to the objectives of the meeting, the above mentioned subjects were dealt with in presentations and through discussions in working groups which made recommendations on several aspects of OV of foxes against rabies.

2. CONCLUSIONS AND RECOMMENDATIONS

2.1 Continued cooperation for the planning of OV

OV was first field tested in 1978 in Switzerland and in 1983 in Germany. Due to its great success, 12 other European countries have since joined, the last being Poland in 1993. The impact on the rabies situation is obvious. While in 1989, 24 377

rabies cases were recorded in Europe, there were only 8 819 in 1994 (Rabies Bulletin Europe 4/94).

OV is influenced by many epidemiological and biological parameters. For **OV** campaigns to be successful, an effective vaccine-bait system, thorough and long-term planning of the campaigns, and a fastidious evaluation of the pre- and post-vaccination data are essential. There is a multitude of prerequisites that have to be fulfilled and conditions to be met in order to launch a thriving oral rabies vaccination programme.

Countries should prepare their plan of action for **OV** well in advance and ensure smooth intersectoral and inter-institutional cooperation between all sectors involved (e.g. veterinary, medical, hunting, forestry etc.) from the very beginning. Strategies have to be worked out according to geographical factors and neighbouring projects of **OV**.

Because of the many parameters having an effect on the **OV** and partly because of the difficult technology, it is the declared aim of WHO to bring people with different experience in the field together, to help establish the method in every country. As the countries started at different times, often under quite different conditions, the success rate is variable.

The challenge for the staff in countries involved in planning **OV** programmes is to obtain and maintain the interest and the continued support of local, national and international authorities, and also to find the best possible compromise between national priorities and international obligations.

International cooperation in the field of cross-border **OV** strategic planning continues. The majority of the participants suggested organizing the next cross-border cooperation meeting in Slovenia in 1996.

2.2 Exchange of data and dates of vaccination campaigns

Countries vaccinating at common borders collaborate on a bilateral basis. A monthly exchange of rabies data would be most suitable, at least on a district level, using maps. The authorities on both sides of the border would therefore be informed on the development of the rabies situation. There should also be coordination and exchange on the dates and areas where vaccination campaigns are carried out.

Once contact is made all sorts of help can be given on the organization, sample taking and surveillance during and after the campaigns.

Decision makers of the two countries should agree on the areas where vaccination should be carried out on both sides of the border and plan a common strategy of vaccination.

2.3 Information on recent research

From the beginning of the border cooperation meetings there has always been the objective to disseminate the most recent research on **OV**. Over the years there have been great improvements.

There has been a shift from the chicken head used as bait in the first field trials to industrially produced baits to fulfill the requirements of all interested countries.

While in the beginning there were one or two vaccines used in field trials, today there are approximately eight.

It was an improvement in the vaccine bait delivery system when small aircraft and helicopters were used. Today, aerial placement of vaccine baits is mainly used, combined with manual distribution in urban areas where aerial methods have their limits. Urban areas are often densely populated by large numbers of foxes and should by no means be missed out in **OV**.

A more recent problem has come up with the increasing fox population density in many parts of Europe. This new situation needs alternative strategies to immunize the appropriate number of foxes to interrupt the chain of infection.

Furthermore, there remain other questions of **OV** still to be answered such as: when should vaccination be discontinued after the last case in the area? What should be the minimum requirement on surveillance to really certify freedom from rabies? What measures are most effective and should be taken in the case where rabies reemerges in a free area?

2.4 EU scheme to support OV financially

The European Union supports **OV** of member countries for several years. 0.5 ECU per vaccine bait is paid and 50% of the costs incurred by the distribution of the vaccine baits is covered.

The above ruling has been extended to countries bordering with EU member countries for an area up to 150 km away from the member country's border.

Until 1 June 1996, a report on the plan of vaccination for 1996 has to be forwarded to the EC in Brussels. The member country files this report for itself and for the country bordering it.

Until 1 July 1997, the bills for the costs of the 1996 vaccination campaigns have to be submitted to Brussels.

2.5 Working group recommendations

At the last meeting on cooperation on **OV** in middle and eastern Europe, in Pulawy, Poland, three subjects were recommended to be emphasized at the next meeting. These subjects were discussed in working groups and recommendations formulated. The final phrasing of the recommendations were agreed upon in a plenary session.

Group 1:

Strategies of eradication programmes

Strategies to be applied in rabies eradication programmes by means of oral vaccination depend on the epidemiological situation and on the status of the vector species (e.g. red fox) population. Strategies to be used at the beginning of an oral vaccination programme (in a situation of low fox density) are well established and have been tested over the last 15 years; however, some adjustments are necessary. Strategies to cope with residual rabies foci or the re-infection of an area of high fox density need further evaluation. The following items are generally accepted or proposed as ideas to be followed:

1. Countries and borders versus geographical regions and natural barriers: the definition of the extension of an oral vaccination campaign should be defined by landscapes rather than by states and borders.
2. Commencement of **OV** (low fox density): the "classical" strategy applied in western Europe can work if particular attention is given to the following points:
 - large areas must be vaccinated (e.g. >6 000 km²);
 - sufficient bait (density e.g. =20 baits/km²) should be maintained from the very beginning;
 - vaccination campaigns must be secured for several years.
3. Continuous campaigns, re-infections (high fox densities). Alternative strategies are needed. None are established yet, but some were tested or proposed:
 - additional campaigns within the same year;
 - vaccination of juvenile foxes at the den in late spring;
 - several bait distribution

campaigns within a short time span;

- reduction of food sources of the red fox (decrease of the carrying capacity of the habitat).

4. If the epizootic comes to an end in an area, it is important to maintain the following tasks:

- continued vaccination for at least two years after the last rabies case
- fastidious surveillance for rabies in the reservoir animal species;
- monitoring of the red fox population dynamics;
- observation of the rabies development in neighbouring countries;

In case of reinfection of an area the rabies virus needs to be characterized.

5. Collaboration and Research:

- international cooperation and close contacts between neighbouring countries;
- close cooperation between scientists (veterinarians, biologists), public health services, wildlife managers, hunters etc.;
- detailed research programmes for specific questions in rabies epidemiology and fox biology.

Group 2:

Funding and administering OV campaigns

1. The EU Decision 89/445 supports OV. The EU pays member countries 0.5 ECU per vaccine bait used in the campaigns and 50% of their distribution cost. The same financial assistance is also provided to adjacent non-member countries for an area of up to 150 km away from the EU-border. It is recommended that the EU supports all areas of up to 150 km away from any point of a border of the EU, irrespective of whether the EU has common borders with these countries. In that way countries such as Croatia, for example, would qualify.
2. In view of the great initial expense to

get a country rabies free by OV one should look for additional finances to vaccinate areas as large as possible. It should therefore be considered that the often practised free vaccination of dogs and cats by the state becomes the responsibility of the owner to reduce costs to the state for OV.

3. Countries with experience in OV should assist countries without such experience for laboratory technology, organizing field campaigns, monitoring and evaluating data, etc.

Group 3:

Diagnostic tests in connection with OV

1. National reference laboratories for rabies should be established. These laboratories should cooperate with WHO reference centres. They should adhere to common standardized procedures.
2. FAT conjugates which are on the market, and those which are prepared locally, should be evaluated by WHO reference laboratories.
3. Training courses at national and international level should be arranged.

3. SUMMARY OF COUNTRY REPORTS

Austria

G. Gram

In Austria, canine rabies was eradicated in 1950. There was no sylvatic rabies from 1955 until 1966. A peak of the rabies epidemic occurred in 1978 with 4 044 animal cases reported. The first OV campaign was conducted in Austria in 1986. Since 1991, when the total infected areas of the country could be vaccinated, there has been a continued decline of cases to 254 in 1994. In 1994, two Federal Provinces bordering Slovenia, Kärnten and Steiermark, were reinfected.

The encouraging results of **OV** led to the issuing of a decree in September 1991, by the Ministry of Agriculture, to use **OV** as the best method of combating rabies.

Current infected areas in Austria are all in the immediate vicinity of the state border. Approximately 440 000 vaccine baits will be distributed in 1995, either by hand placement or by airplane.

From 1986 until 1994 the vaccine strain SAD B19 was used. The SAG 1 strain is going to be used for the first time in 1995.

Bulgaria

R. Valtchovski

From 1984 to 1994, 489 rabies cases in domestic and wild animals, and in one human were diagnosed in Bulgaria. Of the wild animals affected, more than 90% were in foxes and of the domestic animals more than 70% were in farm animals. Though more than half of the rabies cases occurred in domestic animals, the epidemiological pattern is that of fox-mediated rabies.

Rabies occurred throughout the country with a concentration of cases in the northern part of the country (provinces of Montana, Lovetch, Vidin, Vratza and Stara Zagora).

OV of foxes is contemplated. However, it is not envisaged to commence the campaign in 1995.

Meanwhile, preparatory research is being carried out and information in connection with **OV** is being gathered. In regard to density of the fox population it has been evaluated that, in certain places, 0.91 to 1.13 foxes per km² exist. In a limited laboratory trial some years ago, foxes were vaccinated with a SAD strain and challenged. Three out of four foxes survived the challenge.

For Bulgaria it can be concluded that:

1. The rabies situation is worsening with

most areas being affected and an increase in the total number of rabies cases in animals.

2. Features of the disease can now clearly be identified as sylvatic rabies.

Czech Republic

O. Matouch

Rabies was diagnosed in 13 095 animals from 1984 to 1994. The peak was recorded in 1984 with 2 232 cases, the lowest incidence was in 1994 with 221 cases. The incidence has decreased significantly since 1989 when **OV** was started.

Rabies in the Czech Republic occurs in the typical sylvatic form: 90.2% of all animals affected were foxes, 5.1% other wild animals and 4.7% domestic animals.

OV was started along the border to Germany, first towards Bavaria and later towards Saxony. Currently the entire infected area of the country is vaccinated. For 1994 this amounted to 45 500 km² in spring, and 46 900 km² in autumn.

From 1989 until 1992 the SAD B19 vaccine strain was used, since 1992 the SAD Bern strain has been applied. The vaccine baits are produced in the Czech Republic.

The control examinations of the two vaccination campaigns in 1994 gave the following results:

	Bait uptake	Tetracycline marking	Antibody formation
spring	83%	85%	53%
autumn	86%	82%	58%

In 1995 it is planned to revaccinate approximately the same area as in 1994. From 15-16 April 1995, 21 districts of North and East Bohemia are to be vaccinated; from 29-30 April 1995, 34 districts of North and South Moravia. All infected areas of the bordering countries Germany, Poland, Slovakia and Austria

are going to be vaccinated.

Besides the regular control examinations to accompany the **OV**, the National Rabies Reference Laboratory of the State Veterinary Institute in Liberec works on the improvement of the vaccine bait in use.

Croatia

M. Brstilo, A. Rajić and Z. Čač

The first case of sylvatic rabies occurred in 1977 in the north-east of Croatia. Until 1985, the disease covered virtually the entire country, except for the Adriatic islands. The disease clearly follows the pattern of fox-mediated rabies. In 1994, approximately 90% of all animals diagnosed as rabid were foxes.

A small trial of **OV** was first carried out in 1991 with 4 100 vaccine baits, but further trials were interrupted due to the war. In autumn 1994, 80 000 vaccine baits were placed in an area of 5 200 km² in the west of the country in the districts of Istria, Primorje and Gorski Kotar. The vaccine baits were hand-placed by 3 000 hunters.

The follow-up investigations are still underway, except for the bait up-take which resulted in a satisfying 79%.

A plan has been drawn up for future **OV** in the country until 1997, commencing with the areas bordering Slovenia and Hungary. For the 1995 spring campaign the same area will be vaccinated as in autumn 1994.

Estonia

A. Pärtel

Rabies is among the most important diseases in the country. Animal cases varied between 108 (1994) and 451 (1986) from 1980 to 1994. Human rabies cases varied between 8 (1947-1949), 16 (1950-1955) and 3 (1984-1986). The decline in rabies cases within the last six years is considered to be due to greater awareness of the disease by the public and the

vaccination of dogs (between 50 000 and 60 000 annually).

The disease follows a seasonal pattern as in sylvatic rabies. Nevertheless, contrary to most of the other European countries, the red fox and the raccoon dog are equally affected by the disease.

An **OV** would be desirable for the country but, because of financial constraints, a programme cannot be started.

Finland

B. Westerling

In April 1988, a foci of sylvatic rabies, possibly introduced by migrating wolves from the former USSR, was discovered in south-eastern Finland. Almost 30 years had passed since the last occurrence of rabies in animals. All earlier outbreaks had been dog mediated and were eradicated by vaccination of the local dog population.

From April 1988 to February 1989, a total number of 66 virologically verified cases were recorded within a geographical area of 1 700 km² (48 raccoon dogs, 12 foxes, 2 badgers, 2 domestic cats, 1 dog and 1 dairy bull). All virus isolates were identical on a monoclonal antibody panel and all reacted with the MAB P41, which indicates, that the virus was of an arctic strain. It also proved identical to the strain occurring on the Estonian mainland.

As a first measure, the local dog population, some 8 000 animals, was vaccinated against rabies at the expense of the state. In cooperation with the WHO Collaborating Centre for Rabies Surveillance and Research in Tübingen, Germany, a field campaign of oral vaccination of raccoon dogs and foxes was started in September 1988. During four distribution operations, the last one in autumn 1990, a total of 200 000 Tübingen vaccine baits were distributed over a total area of 12 725 km². The largest operation was carried out in spring of 1989 when

120 000 baits were distributed over an area of 8 000 km² covering the estimated infected area and a wide buffer zone.

In accordance with WHO standards, Finland was declared rabies free in March 1991. In spite of this, rabies vaccination of hunting dogs and show dogs remains compulsory.

In an attempt to prevent re-introduction of sylvatic rabies from the south-east, 80 000 vaccine baits are distributed (20/km²) every autumn by air over a 20-25 km deep and 250 km long zone along the south-eastern border with Russia at a yearly cost of about US\$ 0.15 million. In autumn 1995, the distribution will be repeated accordingly.

Germany

H. Schlüter

Rabies in Germany increased slowly in the 1950s and 1960s. In the 1970s the number of cases went down for a few years due to control efforts by gassing, trapping and intensive hunting. Only in the 1980s was there a substantial decrease due to **OV** efforts. Nevertheless, due to set-backs in **OV** programmes, an increase reoccurred in 1994.

OV was started in the Federal Republic of Germany in 1983 and in the former German Democratic Republic in 1989. While hand placing of vaccine baits prevailed in the initial years, aerial distribution of vaccine baits now far exceeds the manual placement of baits by hunters.

In west Germany, a strategy was followed by the different Federal States to vaccinate the widest possible areas according to funds available. However, certain areas were reinfected, or not vaccinated long enough therefore, residual foci caused new outbreaks. As the total area of the former German Democratic Republic could be vaccinated biannually, the results led to a continued decline of rabies cases.

In southern Germany persistently infected areas were treated with a third (or summer) vaccination. This policy was quite successful.

In 1995, all infected areas of Germany will be vaccinated plus some additional areas which, according to a declared policy, have not yet been free of rabies for more than two years.

The following research is carried out in connection with **OV**:

- mathematical models trying to explain deficiencies of the methods and policies,
- monitoring of different numbers of vaccine baits in field trials,
- new method of more than three annual vaccinations in a field trial,
- wildlife research in regard to population dynamics.

Hungary

T. Fehervari, B. Kerekes and I. Heltay

The entire country has been rabies infected for many years. Since 1978 the annual case data have shown little change.

There are signs that the fox population has been growing in recent years, in areas where **OV** is practiced as well as in other areas. More and more foxes are being observed in urban areas.

In Hungary **OV** was started in 1992 covering an area of 5 000 km² at the Austrian/Hungarian border. From the beginning, aerial distribution of vaccine baits was carried out and this technology is consistently being improved. Almost every vaccine bait can be accounted for. The latest advancement is the use of two aircrafts distributing vaccine baits simultaneously.

In follow-up examinations it was demonstrated that 81% of the foxes were positive for tetracycline.

In 1995, an area of about 6 000 km² at the Austrian/Hungarian border will be vaccinated twice.

Italy

F. Mutinelli and M. Tollis

Sylvatic rabies only occurred in the north of Italy in the alpine area from the provinces of Aosta to Trieste (west to east). The first epizootic lasted from 1977 to 1986. **OV** started in 1984 and was practised until 1987. The second epizootic lasted from 1988 to 1989, caused by an infection originating in Slovenia. Only two vaccination campaigns were carried out in 1989 to stop the latter outbreak. The third epizootic started in 1991, again at the Italian/Slovenian border and is still going on. From 1993 to 1994 the province of Bolzano was infected from Austria. Here 10 municipalities were involved.

Three imported dog rabies cases were reported in Italy - in Rome 1984 from Yugoslavia, in Milan 1989 from the Ivory Coast and in Brescia 1992 from Hungary.

In 1994, in four provinces - Udine, Gorizia, Trieste and Bolzano - 46 360 vaccine baits were distributed covering 90 municipalities.

Together with **OV**, an intensified reduction of foxes (hunting) is practised. Dogs and pasture animals at risk are parenterally vaccinated in areas of **OV**. Stray dogs are captured and waste sites (camping etc.) are controlled to reduce food availability to wild and domestic stray animals.

In 1995 it is planned to revaccinate the same areas as in 1994.

Latvia

J. Rimeicans, Z. Andersons and A. Zilvinskis

Until the early 1960s, canine rabies was predominant in Latvia, since then it has changed to sylvatic rabies. Of 2 407 rabies cases (from 126 to 306 annually) registered from 1985 to 1994, 42.5% were foxes, 22% raccoon dogs, 12% dogs, 10.3% cats, 7.9% cattle, 4.5% other wild animals, 0.8% other domestic animals. During this time 2 human cases occurred after exposure to rabies infected animals.

All 26 administrative units of the country were affected by the disease. The most affected districts were Saldus, Ogre, Liepaja, Riga, Talsi, Tukums and Cesis.

In Latvia a first field trial of **OV** was started in 1991 with a vaccine produced in Minsk, Byelorussia.

The trial was extended to 11 districts (22% of the territory of the country) in 1992. No vaccination campaigns were carried out in 1993 and 1994 due to difficulties in vaccine supply.

In 1995, a new type of vaccine is going to be tried in eight districts. The vaccine is going to be injected into meat or chicken heads. It is planned to vaccinate an area of 27 000 km².

The trial will be closely monitored - follow-up samples will be examined in the laboratory, surveillance data will be evaluated, a post-vaccination programme is to be established, etc.

Lithuania

K. Lukauskas and A. Dranseika

From 1985 to 1994, 1 191 rabies cases in animals were registered in Lithuania. There were between 62 (1991) and 173 (1987) cases annually. Of these, 886 were in domestic animals - 181 dogs, 137 cats, 551 cattle, 9 small ruminants, 8 horses. Of the 305 rabid wild animals, 170 were foxes, 92 raccoon dogs, 6 wolves, 22 martens, 6 polecats, 3 badgers, 1 red deer, 4 elks, 1 lynx. The animal distribution does not resemble the picture of sylvatic rabies, nevertheless wild animals appear to be the main rabies reservoir. An explanation as to why domestic animals are in the majority could be an under-representation of wildlife samples coming to the laboratory.

Rabies occur throughout the entire country with a cluster of cases in the north.

Between 1960 and 1994, eight human rabies cases were reported.

OV was practiced from 1983 onward using a Byelorussian vaccine, however this has been stopped during the last years due to financial restraints.

No OV has been planned for 1995.

Moldova

B. Demchenko, I. Stratan

In the last 10 years, 91 foci for rabies were found involving 110 animals,

including 39 foxes, 12 stray dogs and seven cats. In all cases diagnosis was established in the Republic Veterinary Centre (Tab 1).

Rabies foci are reported almost every year and particularly in the central part "codry", which covers five regions (Kalarash, Orgeev, Strasheny, Telensheny, Telenshty, Nisporeny).

Tab. 1: Rabies in Moldova

Year	Number of Foci	Rabies Cases in Animals			
		Total	Wild Animals	Dogs	Cats
1985	11	19	7	1	1
1986	8	8	4	-	1
1987	15	24	7	1	-
1988	4	4	2	-	-
1989	7	7	5	1	1
1990	13	13	7	1	-
1991	14	16	-	6	1
1992	7	7	2	2	1
1993	6	6	4	-	1
1994	6	6	1	-	2
Totals	91	110	39	12	7

Vaccination is the principal method for preventing rabies in towns and villages. About 500 000 dogs are vaccinated annually, and in rabies infected areas, 1 000 cats and livestock are also vaccinated.

To reduce the number of rabies foci during winter (December, January, February), rabies prophylaxis includes shooting, catching and destroying of stray dogs and cats, shooting of wild animals with subsequent laboratory examination.

For the first time oral immunization of wild carnivores took place in 1992 in six regions of the Republic (Edinetscii - 280 doses, New Anens - 350 doses, Orgeevscii

- 420 doses, Kriulensci - 280 doses, Kamenka - 420 doses, Hantchescht - 280 doses). An antirabies virus vaccine was used for oral immunization of wild carnivorous animals; the vaccine was produced by the Belarusian Institute of Experimental Veterinary Medicine.

A total of 2 030 doses of vaccine were used for immunization, injected into the bait (chicken heads, pieces of meat) and was placed during autumn and winter near burrows at a density of 10-15 baits per km². On days 2-3, the number of baits taken was calculated.

In December 1994 and January 1995 in five affected or threatened regions 2

100 doses of vaccine of Belarusian production were used including New Anens - 140 doses, Kalaraschii - 280 doses, Orgeevscii - 280 doses, Sangereiskii - 700 doses and Teleneshtscii - 700 doses.

Oral vaccination focused on the above regions as rabies was reported several times in wild fauna especially in Kalarashscii, Orgeevscii and Teleneshtscii regions; Sangereiskii region is adjacent to Teleneshatcii region where this disease is found. No cases of rabies had been reported in that region since October 1994, nor in the entire Republic since November.

For the autumn-winter period of this year in last year's foci it is planned to use 5 000 doses of vaccine of Belarusian production for oral vaccination.

Between 1983 and 1991 there was one human rabies case every year in the Republic, but none have been reported since 1991. Control activities are coordinated and veterinary and medical services exchange information.

All domestic animals which bite humans are registered. Incidence of rabies may be reduced by mass vaccination of wild animals, regulation of their quantity in forests and reserves (in the last three years that work has not been done due to lack of funding), catching stray dogs and cats, and preventive vaccination of working, guard and pet dogs.

Poland

J.F. Zmudzinski, M. Smreczak and
D. Skulmowska-Kryszkowska

Out of 11 278 rabies cases reported in the last five years in Poland, 81.9% occurred in wild animals. Of all wild animals affected 82.7% were foxes, followed by the raccoon dog with 8.7%. The fox is the rabies reservoir in the country.

The peak of rabies cases was experienced in 1992 with 3 074 cases

reported.

In 1992 the Polish Veterinary Administration decided to introduce **OV**. The first campaign was carried out in 1993 using 700 000 vaccine baits over an area of 39 916 km² in the west of the country along the German/Polish and the Czech/Polish border.

The area comprised six provinces (voivodeships): Szczecin, Gorzow, Zielona Gora, Legnica, Jelenia Gora and Walbrzych. Biannual campaigns were carried out. In 1994 the area to be vaccinated was extended to the neighbouring provinces of Koszalin, Pila and Slupsk. In this year 1 000 000 vaccine baits were distributed by aircraft in the first campaign and 900 000 vaccine baits during the second campaign.

The rabies situation after four vaccinations has much improved. Out of 122 blood samples from foxes in the vaccination area, 113 (92.6%) had rabies antibodies. Out of 240 bone samples tested, 188 (78.3%) were positive for tetracycline.

In 1995 it is planned to vaccinate twice again an area of approximately 60 000 km².

Romania

F. Sapuneanu and F. Badescu

During a 10 year period, the annual rabies incidence in animals varied between 33 (1989) and 107 (1993) cases. There were between 1 (1994) and 9 (1993) human rabies cases recorded.

Rabies is more often reported in domestic animals than wild animals. Among wild animals, foxes are the majority.

Until now there has been no **OV** in the country. Nevertheless, in autumn 1995 there are plans to start a first trial in areas along the Hungarian and Ukrainian border.

Russia

G.S. Poljuškina, D.K. Gruzdev

In the European part of Russia the main reservoirs of rabies are wild carnivores, especially foxes. Between 1986 and 1991, over 1 500 cases of rabies per annum were reported among agricultural, domestic and wild animals; in 1992 there were 950 cases and in 1993-1994, 740 cases per annum.

The worst areas for rabies were, and still are, the central Black Sea, Volga and Ural regions of Russia.

In the central Black Sea sub-region, the highest incidence in the last 10 years occurred in 1988, in 1987 for the Volga sub-region and in 1989 for the Urals sub-region. The number of animals confirmed as suffering from rabies ranged from 400 to 750. In 1993-1994 the number of animals with rabies in the central Black Sea sub-region fell to 90, while in the Volga and Ural sub-regions the level is still at 215 and 230 cases respectively. As a rule, foxes are the source of rabies in domestic and agricultural animals.

In Russia in 1988-1994, rabies was found in over 2 000 dogs and cats and 2 034 wild animals. Over the last four years there have been 150-180 cases of rabies per annum among dogs and 50 among cats.

The peak of incidence among wild animals was in 1988-1989, when rabies was diagnosed in 300-360 wild animals per annum. Incidence among wild animals then fell to 130-140 cases in 1991-1993, and 114 cases in 1994.

In 1994 in the worst regions such as the central Black Sea, there were 143 cases of rabies, of which 44 were in wild animals; in the Volga sub-region there were 215 cases, 28 in wild animals, in the Ural sub-region there were 231 cases, 20 of them in wild carnivores.

The situation in Baškiriya, part of the Ural region, is very complex. In 1993

there were 238 cases of rabies among animals, 33 of them wild animals. One human died of hydrophobia. Because of this they began using rabies vaccine for oral immunization of wild animals in Baškiriya. The vaccine used was RB-71 strain cultured, developed jointly by Russia and Belarus. One thousand doses of vaccine were used in Baškiriya in spring 1994, 5 000 doses in December 1994 and 5 000 more in March 1995. Chicken heads were used as bait, scattered by hand in the paths of foxes and near their lairs. The number of reported cases of rabies in Baškiriya in 1994 fell to 104, seven of them among wild animals.

On one reserve in Tver region occupying 1 150 km², where there are 600-700 foxes and raccoons, they began with 500 doses of vaccine in 1991, increasing to 500 doses twice a year since 1993.

In a number of sub-regions of Moscow region wild animals were vaccinated in autumn 1994 and spring 1995, using 3 000-3 500 doses per season.

Such quantities of vaccine are probably inadequate, since the territory of the region is host to 2 500 foxes, as well as about 30 wolves, 1300 raccoons and badgers, and 5 000 martens, stoats and marsh-otters. Meanwhile in one sub-region of the region, where a hunting reserve of 1 211 km² contains 400 wild animals including 80 foxes, where 500 doses of vaccine were used twice a year in 1993-1994, no cases of rabies have been reported.

In Penza region, where the vaccine was used twice - at the end of 1990 and at the beginning of 1992 - incidence has fallen from 61 cases to one. After a two-year interval the vaccination campaign was restarted, and 4 600 doses of the preparation were used in March 1995.

Vaccination will be continued in a number of affected regions of Russia.

Slovak Republic

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There were 565 cases of animal rabies diagnosed in 1994. The animals involved show the typical picture of sylvatic rabies. 75.7% of all rabid animals were foxes, 4.3% other wild animals and 20% domestic animals (64 dogs, 43 cats, 6 farm animals).

First **OV** trials using a locally developed vaccine (Vnukovo-32/107) were carried out in Slovakia in 1992 and 1993. The vaccine bait is commercially produced. It is planned to continue **OV** in 1995-1996 with biannual campaigns.

In spring 1994, 400 100 Czech-made vaccine baits were used and only 299 900 locally produced vaccine baits. In autumn 1994, 700 000 Slovakian vaccine baits were used.

There were the following results on the control examination:

- On days 3, 8 and 14 in certain areas the uptake of the vaccine baits was checked. 78.9% of all vaccine baits had disappeared in spring, and 81% in autumn.
- The tetracycline marker was discovered in bones in spring from 202 samples out of 298 (67.8%) and in autumn from 188 out of 388 (48.5%).
- For the rabies antibody titration an ELISA test and a neutralization test with mice were used. In spring, 76.2% of 21 samples were antibody positive with the ELISA test (80.9% for the mouse neutralization test), in autumn, 94.7% of 19 samples were positive for the ELISA test (89.4% for the mouse neutralization test).
- 75 rabies virus strains isolated in the field were characterized with monoclonal antibodies to be field virus.

In 1995, 49 035 km² are going to be vaccinated twice with the Vnukovo 32/107 strain. This is the total area of the

country.

Research is going to be carried out on the efficacy of the vaccine baits produced in Slovakia.

Republic of Slovenia

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In Slovenia **OV** was practised for the first time in 1988. In that year 805 rabies cases, a peak for the country, were registered. An improvement in the rabies situation was noticed only up to 1991 (188 cases). Afterwards regular vaccination could not be guaranteed for financial reasons. This strategy exemplifies that irregular vaccination programmes and vaccination of small areas only have a detrimental effect rather than improving the disease situation.

In 1994, 842 rabies cases were recorded with 723 cases in foxes (86% of the total).

In a spring campaign in 1994, an area of 2 800 km² was vaccinated in the north-west of the country. In autumn 1994 the area was extended to cover 11 000 km² (54% of total country).

There were the following control results: 2 weeks after placing the vaccine baits 56% of them had been taken by the wild animals, 45% of a first sample were positive for tetracycline and 38% of a first sample had rabies antibodies.

In 1995 it is planned to have two vaccination campaigns to cover the entire country.

Former Socialist Republic of Yugoslavia

M. Petrovic, M. Simic and S. Stankov

Former Yugoslavia had been treated from 1953 as a country with urban rabies in its southern part and occasional local sylvatic rabies epizootics in the north-eastern part of AR Vojvodina. In 1977 the European sylvatic rabies spread into Former Yugoslavia from Hungary and

arrived in 1982 in Bosnia, an area where classic urban rabies had earlier existed. Eleven rabies isolates from Bosnia and Herzegovina (4), Montenegro (2) and Vojvodina (5) were characterized by monoclonal antibodies by the Pasteur Institute, Paris, in 1987. The examination revealed that both classic urban and European sylvatic fox rabies virus strains were present in Bosnia. Unexpectedly, all strains from Vojvodina were characterized as urban strains, where epidemiologically a typical form of sylvatic rabies had previously existed.

However, the epidemiological evaluation of the rabies cases over the last 10 years gives the picture of sylvatic rabies. Of 849 rabies cases from 1985 to 1994, 655 (77.2% of total) were in foxes, 9 in other wild animals and 185 (21.8%) in domestic animals.

The cases are distributed mainly in the north of the country up to the north-eastern part of Serbia close to Romania and Bulgaria. There had been no registered cases in the last ten years in southern Serbia, Kosovo and Metohija where urban rabies had previously existed.

Plans for OV had been made a few years ago, but were subsequently not realized and there are no plans for 1995.

4. LECTURES

4.1 OV in Europe - Why does it take so long?

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(Please see also: Stöhr et al.: Vet. Rec. London, 1995, in press) Oral immunization of foxes has led to a substantial decrease in the number of rabies cases in Europe. Between 1989 and 1994 rabies prevalence in animals was reduced to less than 20% of the 1989 level in countries which had been conducting oral immunization campaigns since 1992 or before. Since 1978 more than 73.7

million baits have been spread over an area of 4.9 million km². However, large territories have not yet been declared rabies-free so that the economical and public health benefits stemming from reduced rabies prevalence has so far remained marginal. Setbacks have occurred which have slowed vaccination progress, particularly during the end of the 1980s and the first years of the 1990s. Reasons for territorial differences in progress are discussed and factors which have contributed to greater success in vaccination campaigns are identified. Regrettably the many positive and negative experiences with oral immunization of foxes in Europe have resulted in only a limited number of scientifically validated recommendations for strategy selection under the different European ecological and epidemiological conditions. Substantiated answers have not yet been found to questions such as duration of vaccination campaigns, level of surveillance during and after campaigns and appropriate countermeasures to compensate for the obvious increase in fox population density. In this context WHO will facilitate drawing up of guidelines to assist responsible officers at national and regional level to make strategic decision for field application of oral immunization with the ultimate aim of eliminating terrestrial rabies from Europe.

4.2 Strategy in vaccination campaigns- examples from Germany

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There are a number of very encouraging examples of how rabies was controlled in limited areas by OV. However, this development was sometimes paralleled by a number of setbacks in various countries.

Only the countries Austria, the Czech Republic and France show a record of continued improvement of the disease situation. Countries like Belgium, Germany, Slovenia and Switzerland

recorded a rabies incidence in 1994 higher than in the previous year. The reasons for the latter are reinfections of formerly rabies-free areas or outbreaks which develop from residual foci in areas not treated long enough by **OV**.

At present there are two obstacles in many countries in regard to **OV** which cause setbacks:

- lack of money and
- high fox populations.

Due to budget restraints it becomes difficult to guarantee even the regular bi-annual vaccinations using 15-20 vaccine baits per km², in a large enough area (minimum of 6 000 km²), long enough (two years after the last case). The high fox populations are a technical problem on the one hand, a strain on finances on the other.

The fox hunting bag is a measure for us, though not a very exact one, on the fox population density. Fig 1 gives the

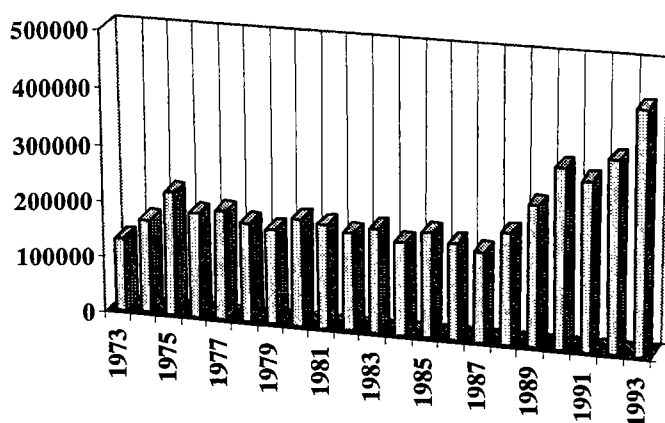


Fig 1: Fox hunting bag - Western-Germany

example how fox density has risen in Germany during the more recent years. Certain areas within the country, being excellent fox habitats, have an even more pronounced rise. Living conditions must have been favourable for the fox; like food abundance, mild winters, no rabies as a reducing factor and maybe a diminished hunting pressure.

In the beginning of **OV** it was considered that 15 vaccine baits per km²

would be used well in surplus of the fox population.

Nevertheless, this strategy was quietly changed over the years after the beginning of **OV** in Germany. First, 18-22 vaccine baits were used in the bi-annual vaccination campaigns. Later, in Bavaria and Baden-Württemberg a third vaccination in summer was introduced. With the latter vaccination especially the rabies susceptible young foxes of the spring were to be reached and the vaccination was to be solidified in the autumn campaign.

Today in Baden-Württemberg a method is used which makes use of three campaigns, for the very resistant rabies foci where high fox populations have been found. In each campaign the aircraft distributes twice approximately 15-18 vaccine baits in the same area at an interval of three to four weeks. The aircraft flies the first time from north to south and the second time from east to west. This increases the total number of baits to about 90-108 per km² distributed over a year.

The interim vaccination methods are not yet well established but they are no doubt a first alternative where the "classical" method has hitherto failed.

The following is a summary on the points above and adds others supplementing the strategy in vaccination campaigns:

1. Start **OV** big - with an area as large as possible, and not less than 6 000 km². Choose the area well by considering natural barriers and consider the vaccination programme of the neighbouring countries. Secure money for several years.
2. Decide on vaccine baits per km² and vaccination campaigns according to fox population; you may have to adjust in the course of your programme:
 - 15-20 vaccine baits and biannual vaccination

- Consider increase of vaccine baits or third vaccination in one year
 - In extreme situations the problem areas could be vaccinated twice in one campaign as practised in Baden-Württemberg. Reference is made as well to the method practised in Switzerland baiting the young foxes at the den in May/June.
3. Hold and defend your rabies-free area. Do not move into new areas when finances are not secured. Consider buffer zones.
 4. Organize intensive surveillance during vaccination campaigns and after elimination of rabies.
 - Practice follow-up examinations on efficacy of vaccination (TC, serology) and safety (MAB)
 - Initiate securance of sample supply on freedom from rabies (8 foxes per 100 km² and year). The rabies-free status prevails when 2 years after the last case in the country (or during the programme for an area) no further case occurs.
 - Use computer programmes in the laboratory for easy evaluation of data.
 5. Form a committee of scientists and

decision makers of veterinary, medical, hunting departments to accompany OV. Meet at least once a year.

6. Practice close bilateral border cooperation.

4.3 Rabies persistence and fox population dynamics in Switzerland

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The current rabies epidemic reached Switzerland in 1967 and spread over most of the country in subsequent years. Campaigns to orally vaccinate red foxes (*Vulpes vulpes*) against rabies began in 1978 (Kappeler et al. 1988), and resulted in a decline in the number of diagnosed cases from a maximum of 1 738 in 1976 to 25 cases in 1990. After a re-infection in the Jura Mountains (north-western Switzerland), the number of cases started to increase again and reached 225 in 1994, in spite of the continued vaccination campaigns in spring and autumn and the rather high bait uptake rate (78% in 1992, 68% in 1993). In the following, we summarize our hypotheses and results of a field trial presented in detail by Breitenmoser et al. (1995) and Kaphegyi (1995).

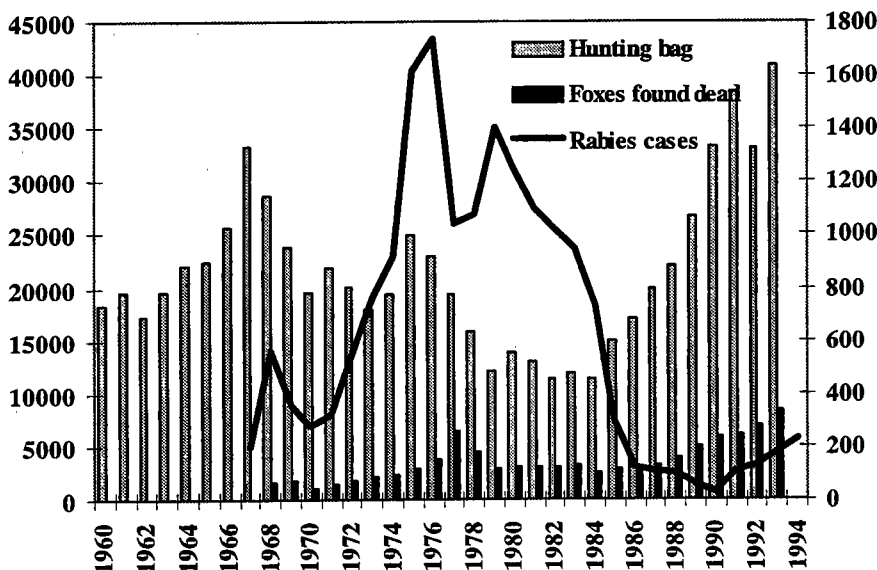


Fig 2: Switzerland (Breitenmoser et al. 1995)

The renaissance of rabies in Switzerland occurred parallel to an increase of the fox population. The hunting bag and the number of road kills indicate a rapid growth of the fox population in recent years (Fig 2). With the increase of the fox density, we observed a shift of rabies from adult to juvenile foxes. While juvenile and sub-adult foxes were

under-represented in samples of rabid foxes until the mid-eighties when, compared with hunting figures or road kills, the proportion has increased considerably in the past years. In 1993, 74% of all foxes found positive for rabies were less than one year old. Young foxes are difficult to reach with the conventional spring vaccination campaigns because most of them still live in the dens and do not yet take solid food.

The data suggest that today, the high population density and especially the high number of young foxes with no or weak protection are mainly responsible for the persistence of rabies in Switzerland. To improve this situation, additional methods to either reduce the number of young foxes or to increase the proportion of vaccinated juveniles were tested in the Canton of Solothurn in northwestern Switzerland. The study area of 600 km², representing 48 hunting grounds, was split into three sub-areas. These three areas were treated as follows:

(1) additional vaccination of juvenile foxes by placing vaccine baits near the den; (2) reduction of juvenile foxes by trapping or shooting at the den; (3) control area. The activities at the dens took place in the months of May and June 1994, when the young foxes start to leave the den and to accept solid food.

Reduction of the juvenile foxes at the den was planned in 13 hunting grounds. Only eight of those hunting associations returned the data forms, and of those, only six reported the culling of young foxes. The hunters shot or trapped at a total of 80 dens. They trapped 31 fox cubs, and shot 14, with a total time input of 650 hours. Of the 45 cubs killed, 21 were trapped in one hunting ground only; but the hunters needed more than 350 hours to do it. The additional bait distribution at the den was carried out in 23 hunting grounds. Each den was treated twice with a time difference of seven days. The hunters placed 6-10 baits around the entrances of the dens. They distributed a total of 6 000 baits at 344 dens. To determine the efficacy of the action, we

examined foxes shot in the study area for the bait-marker tetracycline. Up to the autumn vaccination campaign (9 September 1994), the percentage of juvenile foxes showing tetracycline marker was significantly higher in the den-vaccination area, compared to the control and culling area (80% versus 40% respectively; $\chi^2 = 16.82$, $p = 0.001$). The percentage of the tetracycline-positive adult foxes increased as well, but the difference was not significant (89% versus 85%, respectively).

The experiments carried out in the canton of Solothurn showed that the culling of juvenile foxes at the den at present is not an efficient method to reduce the number of foxes. Even if experienced hunters perform the shooting or trapping, the time investment is quite high. In one hunting ground, 22 juvenile foxes were killed, but the time needed was 410 hours. In contrast, the additional delivery of vaccine baits to the dens can improve the immunization of juvenile foxes. The distribution of the baits was not very time consuming, and the effect was significant. The bait uptake rate in the areas additionally treated at the dens was doubled. This will reduce significantly the density of susceptible young foxes in the critical period between their dispersal from the parental territories up to the vaccination campaign in autumn.

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4.4 Diagnostic methods in regard to OV

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Rabies diagnosis

The most important laboratory task is to identify rabies infected animals. This is important for the surveillance of the disease as well as evaluating the success of OV.

The three tests today most commonly used are:

- the fluorescent antibody test (FAT)
- the mouse inoculation test (MIT)
- the cell culture inoculation test (CCIT)

All these tests can be considered as approximately equally sensitive in spite of a very different technology. Quite often they are combined, especially when human exposure is involved.

The FAT is probably most widely used. It replaced the histopathological tests though the latter might still be in use occasionally. The FAT results are greatly influenced by the conjugate and the quality of the microscope (exciter and barrier filter must be chosen for optimal fluorescence), among others.

The rabies group of our laboratory has experience with three different conjugates.

1. Bioveta Ivanovice na Hane in the Czech Republic

The conjugate is lyophilized; it contains a specific globulin labelled with fluorescein in isothiocyanate. The contents of the vial is to be diluted with 20% normal mouse brain suspension. The working dilution is 1:7.

The conjugate sometimes gives non-specific reactions and has a high background fluorescence.

2. Centocor

It is provided lyophilized for reconstitution in 5 ml distilled water. It is relatively expensive and recommended to be used undiluted. We use it though to stain brain impressions diluted 1:10 or 1:20. It has virtually no background fluorescence. When

Bioveta, Centocor and MIT were compared in our institute in 1992, 13 out of 36 brain samples treated with Centocor gave negative results, and four doubtful results, while they were strongly positive with the Bioveta conjugate and the MIT. There were similar results from the literature (P. Stöhr *et. al* in Tierärztl Umschau 47, 813-818, 1992) when Centocor was compared with two German-produced conjugates. Centocor showed to a certain extent negative or doubtful results, with certain strains of rabies field virus. The producers of Centocor have now changed the content of mAB of the product with the effect that all field virus in Europe are recognized accordingly.

Centocor gives very good results in the RFFIT (and can be highly diluted because of the cell culture technique) when the CVS is used.

3. Diagnostic Pasteur, 3 Bd. Raymond Poincaré-BP3, 92430 Marnes-la-Coquette, France.

The lyophilized product contains IgG immunoglobulin obtained by immunization of rabbits with purified nucleocapsid from the Pasteur strain. This conjugate has been used by us over the last three years for brain impression smears, RFFIT and CCIT. It has almost no background fluorescence.

The MIT has long been in use. Even small amounts of virus in the tissue can be proved due to the intracerebral inoculation. The disadvantage is that the results can only be read approx. 7-10 days after inoculation.

The CCIT is usually practiced with murine neuroblastoma cells. It is, under well-defined circumstances, at least as efficient as the MIT, but needs more skilful personnel and a well equipped laboratory.

Control tests in connection with OV

Specific tests to check on the efficacy and safety of OV are the following:

- a test to determine the rate of bait up-take by identifying a marker (usually tetracycline) which is added to the bait;
- a serum neutralization test. Most commonly in use is the RFFIT (the rapid fluorescence

focus inhibition test) to determine the seroconversion rate;

- a monoclonal antibody (MAB) test as safety measure to distinguish field and vaccine virus (if modified live rabies virus is used).

Four weeks after the placing of vaccine baits (by hand or by aircraft), foxes are shot by the hunters, forwarded to the next veterinary investigation centre where the samples for the above tests are taken. The running of the above mentioned tests are usually carried out by specialized laboratories. The techniques can be acquired in WHO reference laboratories.

It is recommended that after each

vaccination campaign at least eight foxes per 100 km² are investigated for an appropriate evaluation of **OV** in the area.

Monitoring data

The scientists of the laboratory should make an effort to evaluate the collected data by making use of the modern computer technology. Software programmes are available which combine the assessing of the routine surveillance and the results of the follow-up examinations of **OV**.

Oral immunization of foxes in Europe - computer software: *Weekly Epidemiological Record*, 69, No. 21, 159 (1994).

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ANNEX 2: MAP OF EUROPE - PLANNED OV FOR 1995

