An outbreak of rabies in West Malaysia in 1970 with unusual laboratory observations

by Dora S. K. Tan

Virus Research Officer,
Institute for Medical Research,
Kuala Lumpur.

Abdul Wahab b. Mohamed Ariff

Director of Development and Research,
Ministry of Health,
Kuala Lumpur.

Mohamed Noordin Keling

Director,
Training Branch,
Veterinary Division,
Ministry of Agriculture and Lands,
Kuala Lumpur.

and George W. Beran

World Health Organisation Consultant on Rabies,
Western Pacific Regional Office,
Manila,
Philippines.

Historical Review

ALTHOUGH RABIES has been known in Malaysia since 1884, it was only since 1924 that records of human cases have been kept. Most of the cases have consistently been in states bordering Thailand which is highly endemic for rabies; sporadic outbreaks in other states have usually been quickly suppressed.

One major interior outbreak occurred in Province Wellesley and Perak state in 1945. Epidemiological evidence indicated that Allied troops arriving from India and Burma introduced the disease into these areas through their dogs. Although only sporadic cases continued to be reported from Province Wellesley, rabies became highly endemic in Perak and was not brought under control until 1953. In 1952, an extension southward resulted in

Four laboratory methods were promptly employed at IMR in the diagnosis of rabies.

Mouse inoculation tests (World Health Organisation, 1966 b)

Samples of hippocampal gyrus, cerebrum and cerebellar cortex were weighed and ground in sterile mortars with pestles. Physiological saline with 10% inactivated filtered rabbit serum was slowly added to prepare 10% tissue emulsions. Following centrifugation at 1000 rpm for 5 minutes, 0.5 ml. of supernatant fluids were mixed with 0.5 ml. of antibiotic solution containing 100 units of penicillin and 4 mg. of streptomycin per ml. The mixtures were incubated at least 30 minutes at 4°C. Six mice (10-15 grams) were lightly ether anaesthetised and injected intracerebrally with 0.03 ml. each. Mice showing signs of rabies after 5 days were harvested in a moribund state or following death and the diagnoses confirmed by microscopic tests. Mice remaining normal for 30 days were usually discarded. In some cases, serial passages were made of brain tissues from mice dying or remaining normal.

Seller's stained smear examinations (World Health Organisation, 1966 b)

Rubbed smears of the inner surfaces of the hypocampal gyrus (relatively free of glycerol-saline) were immersed while still wet in Seller's stain for 1-5 seconds, rinsed in running water and air dried. Stained smears were examined microscopically for Negri bodies.

Histopathological examinations

Zenker-acetate fixed tissues were paraffin blocked, sectioned and stained by Fast Green-Acid Safranin stain (Smith, 1953) by the Pathology Division of the IMR. Microscopically, Negri bodies were stained intensely bluish green with symmetrically arranged innerkorperchen corpuscles and vacuoles. Erythrocytes stained uniformly green, microglial and nerve cell nuclei and nucleoli stained bright red, and cytoplasm and general background stained pale pink or grey.

Fluorescent antibody (FA) microscopic examinations

The Communicable Disease Center (CDC), Atlanta, Georgia, USA (Cherry, et al., 1960) technique was followed except that commercial antirabies conjugate was diluted 1:40 to reduce non-specific staining while retaining specific sensitivity as measured by the positive controls.

Laboratory findings

During 1970, 50 brain specimens received from all over Malaysia were examined by the 4 diagnostic techniques. In all of the 6 positive findings (Tables 1 and 2), the mouse inoculation test was the only initial procedure yielding evidence of infection. In all of these, the inoculated mice were confirmed as positive by each of the 3 microscopic tests. Reisolation attempts on the field specimens following storage in 50% glycerol saline at −20°C for 7-9 months were positive only on the cat brain tissue. The negative microscopic examinations-positive mouse inoculation tests encountered during the 1970 outbreak were unusual; this pattern had not been observed during the 1954 and 1963 outbreaks.

Discussion

The control of rabies in Malaysia is based on coordinated functions of dog vaccination and stray dog control by the veterinary services, of rabies diagnosis by the IMR, and of post-exposure prophylactic vaccination of exposed persons by the medical services. An immune belt, in which compulsory dog vaccination and stray control are continuously carried out, is maintained along the Malaysia-Thailand border and similar zones are instituted wherever outbreaks occur in the rest of Malaysia. Although this programme cannot be expected to achieve eradication of rabies from Malaysia, it has enabled the country to control the disease with an expenditure that is economically feasible. During the 1970 outbreak in Perlis and Kedah states, the vaccination of more than 3000 dogs and destruction of over 8,000 dogs and cats must be credited with limiting the principal part of the epidemic to 3 months. However, it must be stressed that rabies spread rapidly throughout Perlis and Kedah following its introduction, and that 5 persons died of the disease before it was again brought under control.

The fact that 3 of the dogs and the single cat which were laboratory confirmed as rabid were pets, and that only 1 of these had been vaccinated, indicated that dog vaccination campaigns should be intensified, if possible on a house-to-house basis, in the border areas. The one confirmed case of canine vaccination failure emphasised the need for proper handling and use of these vaccines and the importance of confinement and observation of all dogs, whether vaccinated or not, which have bitten persons. The fact that all 5 human deaths, 1 of which was laboratory-confirmed, followed bites by stray dogs emphasised the need for continuous thorough control of stray and unvaccinated dogs in the immune belt.

Although adequate facilities for prompt diagnosis of rabies are maintained at the IMR, their

usefulness is dependent upon the receipt of specimens from field cases. Of the 10 dogs and cats proven to be involved in this outbreak, diagnostic specimens of only 5 were received and of 5 human deaths, tissues collected at necropsy were submitted from only one.

The 5 human cases were all clinically characteristic of rabies, with incubation periods of 1-3 months, clinical syndromes with hydrophobia and encephalitis, and death ensuing within 1-5 days. It is pertinent that 4 of the patients reported pruritis at the site of exposure; in one patient an intense pruritis extended along the arm and shoulder to the spine. The lack of Negri bodies and FA detectable antigens in the brain of the one patient examined at necropsy after a clinical period of 4 days could not be explained; the mouse inoculation test was strongly positive.

The dogs and cat diagnosed as rabid in Perlis and Kedah states all exhibited clinical illnesses compatible with rabies. The 4 dogs were all killed promptly after biting human victims; all were negative for Negri bodies, which may be explicable on the basis of their destruction early in the clinical course of the disease. The FA test should still have been positive; it is possible, however, that the FA procedure used at the time was not sensitive enough to detect only limited amounts of rabies antigen which might have been present. The brain tissues of each of these dogs yielded rabies virus by mouse inoculation tests. The cat died in confinement on the first day of observation. Negri bodies or FA detectable antigens were not found, although rabies virus was demonstrated by mouse inoculation tests on 2 occasions, 6 months apart.

The dog from Kuala Lumpur posed unique clinical and diagnostic problems. The source of exposure could not be determined although an extensive investigation was conducted. The clinical illness in the dog was compatible with rabies in the initial acute phase but the disease progressed to a chronic phase with divergent complications and the animal was ultimately euthanised. Although rabies virus was recovered from the brain of the dog, it is evident that it was present in very limited quantity. The brain was negative for Negri bodies, as was found in the cases of rabies in Perlis and Kedah. Following inoculation of 6 mice, 1 died on each of days 8 and 16 and 1 was killed in a moribund state on day 13. The first mouse was not further studied, the last two were positive for both Negri bodies and fluorescing antigens. One of the 3 surviving mice was sacrificed on day 20 for blind passage but proved negative for virus; the other 2 survived to day 30 and were disposed of. One of us (GWD), sent to Malaysia as a WHO consultant at the request of the IMR, found the laboratory capability in rabies diagnosis at the IMR to be reliable. While certain procedural modifications were recommended to increase the sensitivity, especially of the fluorescent antibody test, the lack of Negri bodies in the human and animal brains submitted to the laboratory in this epidemic remains an unusual observation. The findings in this incident again emphasise the importance of combining mouse inoculation tests with direct tissue examinations in the laboratory diagnosis of rabies.

Another of us (DSKT) has suggested that the rabies virus strain involved in this epidemic may have possessed some atypical characteristics, possibly having emerged as a variant strain through the continual co-existence during the past 2 decades of 2 live rabies viruses, the Flury LEP strain on the Malaysian side of the border and street rabies virus on the Thai side. It must be recognised that the coexistence of these 2 types of rabies virus in dogs is a unique situation prevalent only along the Malaysia-Thailand border.

Summary

Rabies is a public health problem in Malaysia only along the Malaysia-Thailand border where the disease has been periodically introduced through dogs crossing over from Thailand. There is no evidence of maintaining reservoirs in the wildlife of Malaysia. Rabies control is based on regulation of importation of dogs from other countries and upon the maintenance of a canine rabies immune belt 30-50 miles in depth along the Thai border to prevent the extension of the disease from the north. Dog movement between the immune belt and the rest of Malaysia is strictly regulated. Continual surveillance is maintained throughout Malaysia and prompt measures are taken to suppress any outbreaks of rabies.

During the first half of 1970, a rabies outbreak occurred in Perlis and Kedah states along the Thai border, presumably due to relaxation of rabies control measures in the immune belt because of increased communist terrorist activities in the area. A single case of canine rabies was found in Selangor state during the same period. Among 11 persons exposed to rabies during the outbreak, 7 were given anti-rabies vaccine (prepared at IMR) before the appearance of any clinical manifestations of rabies. No post-vaccinal reactions were recorded, and only 1 of these patients subsequently developed

Table 1

DETAILS OF REPORTED HUMAN RABIES CASES, KEDAH AND PERLIS, 1970.

Name	State (locality)	Age (years)	Sex	Race	Site of Bite	Date of Bite	Treatment (Date started)	Particulars of Patient			Particulars of Biting Animal			
								Date hospitalized	Date of death	Lab. Confirmation	Species	Stray or pet	Outcome	Lab. Diagnosis
Cheong Kee Song	Perlis (Bkt. Ketri)	9	M	Chinese	Right popliteal fossa	Jan., 1970	Nil	24.4.70	26.4.70	+	Dog	Stray	Escaped	N.D
Ku Ah bte Ku Ahmad	Kedah (Kodiang)	30	F	Malay	Right thigh and leg	22.2.70	2ml.x14 (23.2.70)	19.5.70	20.5.70	N.D	Dog	Stray	Killed	N.D
Ibrahim bin M. Hassan	Kedah (Kodiang)	61	M	Malay	Right foot	21.4.70	2ml.x 6 (21.5.70)	21.5.70	26.5.70	N.D	Dog	Stray	Killed	N.D
Elias bin Che Mohd.	Perlis (Bkt. Ketri)	10	M	Malay	Right popliteal fossa	Apr., 1970	Nil	16.6.70	18.6.70	N.D	Dog	Stray	Escaped	N.D
Ng Doi Kuan	Perlis (Kaki Bukit)	8	M	Chinese	Unknown	Unknown	Nil	11.10.70	11.10.70	N.D	Dog	Stray	Escaped	N.D

Note: N.D = Not Done; i.e. not submitted for laboratory study.

in June after 2 days hospitalisation in Kangar.

An 8-year-old Chinese boy from Bukit Tasoh Kaki, 2 miles from the Thailand border, died in the Kangar Hospital in October with clinical diagnosis of rabies. His parents confirmed that he had been bitten some time earlier by a stray dog but the time or site of exposure could not be ascertained and he had been given no ARV. In none of these 5 human rabies cases were the exposing dogs submitted for laboratory diagnosis and only the first patient was examined at necropsy. (See Table 1)

Animal rabies cases

Aside from the 5 stray dogs in which rabies was confirmed by resultant human cases, 4 of 7 animal brains submitted to IMR from Perlis and Kedah during the first half of 1970 were confirmed as rabid. All of the 4 positive animals were submitted during June and all were from Kedah. One of the positive dogs, a stray, entered a village in Kodiang at about 10 one evening and viciously bit 4 goats, 5 cattle, 2 geese and a 7-year-old Chinese girl before it was killed at about 7 the next morning. Two other positive dogs, one from Changloon and the other from Tunjang, 5 and 16 miles respectively from the Thailand border, were pets which were killed after biting their masters. One of these had been vaccinated with Flury LEP strain vaccine 16 months previously. Three other dog brains submitted were negative for rabies. One cat in Jitra, about 20 miles from the Thailand border, was placed under veterinary observation after biting its master; it died during the following night and was confirmed as rabid upon examination at IMR.

A single case of canine rabies with many unusual features was diagnosed in Kuala Lumpur, Selangor state in May. A 3-year-old male Shetland sheep dog, one of a pair from a dam originally imported from Australia to Kuantan in 1968, had been kept in Kuala Lumpur since September, 1969. The owner reported that the 2 siblings had been kept in his neighbourhood since their arrival and had only limited contact with other local dogs. No other pets were kept on his premises. From 14 April, the male dog suffered a protracted febrile illness with anorexia, champing of the jaws and profuse salivation. Following initial treatment by a private veterinarian, the dog was apparently improving by 17 April, but relapsed on 26 April. It developed a goose-stepping gait, apparent blindness in the right eye and circling to the left. Further therapy was non-productive and the dog was euthanised on 8 May. Laboratory studies confirmed the presence of rabies infection. The source of exposure of this dog to rabies could not be ascertained. (See Table 2)

Treatment of persons exposed to rabies

Evaluation of all animal bite victims and administration of ARV are performed only at government hospitals. District and state veterinary offices are charged with epidemiological investigations of biting animals, confinement of such animals for a 10-day observation period where possible, and shipment of brain specimens to IMR for analyses. Post-exposure immunisation is administered promptly, or deferred pending clinical and laboratory findings, in accordance with the recommendations of the W.H.O. Expert Committee on Rabies (1966a)

Table 2

DETAILS OF HUMAN EXPOSURES TO PROVEN RABID ANIMALS, 1970

Name	State (locality)	Age (years)	Sex	Race	Site of Bite	Date of Bite	Treatment (Date started)	Particulars of Biting Animals				Remarks
								Species	Stray or pet	Outcome	Lab Diagnosis	
Ng Hoo Lum	Kedah (Kodiang)	60	M	Chinese	Right foot	21.4.70	2ml.x15 (21.5.70)	Dog	Stray	Killed	N.D	Diagnosis in dog based on fatal case in Ibrahim bin M Hassan bitten on same day by same dog
Shanta Kumari	Selangor (Kuala Lumpur)	22	F	Indian	Scratch on finger	10.5.70	2ml.x15 (10.6.70)	Dog	Pet	Killed	+	
LEE LAY TIN	Kedah (Kodiang)	7	F	Chinese	Right ear	2.6.70	1ml.x14 (12.6.70)	Dog	Stray	Killed	+	
Lim Pook Nyan	Kedah (Changloon)	14	M	Chinese	Scratch on right palm	10.6.70	2ml.x15 (24.6.70)	Dog	Pet	Killed	+	
Tan Kim Kooy	Kedah (Tunjang)	74	M	Chinese	Right thumb	22.6.70	2ml.x15 (8.7.70)	Dog	Pet (vaccinated)	Killed	+	
M. Radzi bin A.Rani	Kedah (Jitra)	2	M	Malay	Left leg	30.6.70	1ml.x15 (13.7.70)	Cat	Pet	Died	+	

Note: N.D = Not Done, i.e. not submitted for laboratory study

and the IMR Bulletin Number 8 which has been distributed to relevant authorities and clinicians.

Semple-type phenolised sheep brain tissue origin vaccine is prepared and distributed by the IMR to medical officers throughout Malaysia. This vaccine is given in 14 daily subcutaneous injections of 2 ml. each, to persons 14 years and older, usually in the anterior abdominal wall. The dosage is reduced in children under 1 year to 0.25 ml., 1-3 years to 0.5 ml., 4-6 years to 0.75 ml.; 7-9 years to 1.0 ml., and 10-13 years to 1.5 ml. per injection. The administration of anti-rabies hyperimmune serum is recommended in accord with W.H.O. recommendations, but is available only by importation.

Of the 11 persons bitten by animals which were promptly or subsequently (through human death following exposure) proven rabid, administration of AVR was begun the day following exposure in one, within 2 weeks in an additional 3, within 1 month in an additional 3, on the day of onset of clinical rabies in one, and not at all in 3 animal bite victims. The one person given ARV beginning the day after being bitten on the thigh and leg died of rabies after an incubation period of 3 months. The one rabies patient administered ARV during the clinical phase of the disease was not protected. This latter patient, and the other 3 who received no vaccine, all died of rabies. The 5 victims who were given ARV following laboratory confirmation of rabies in the exposing animals have remained normal 5-7 months since their exposures. No post-vaccinal reactions have occurred in the patients given ARV. (See Table 3.)

Table 3

The Outcome Of Animal Bite Victims Given Or Not Given Anti-Rabies Vaccine Following Bites By Confirmed Or Unconfirmed Rabid Animals.

Anti Rabies Vaccination	Patient Outcome	Rabies Confirmed	Rabies Not Confirmed	Total Patients
Given*	Died	0	1	1
	Survived	6**	0	6
Not Given	Died	0	4	4
	Survived	0	0	0
Totals	-	6	5	11

* Vaccine given prior to onset of clinical rabies.

** One canine case confirmed on basis of rabies death in another person bitten by the same dog.

Laboratory Diagnostic Procedures

Initial processing

Brains were removed from cadavers (dogs, 1 cat, 1 human patient) at respective veterinary centres or hospitals. Excised brains were cut in two halves, anterior-posteriorly. One-half was placed in 50% glycerol saline for impression smear and virus isolation studies. The other half was placed in Zenker-acetate fixative for histological studies. The specimens were sent by courier, railway or air express or other rapid means to IMR.

a major epidemic in Selangor state. The severity of this outbreak prompted a national programme of compulsory vaccination of all dogs in rabies-infected states and a rigorous programme of destruction of unowned and unvaccinated dogs. The success of the control programme was such that in April, 1954, Malaya (as the country was known before 1963) was declared rabies-free. An immune belt was established in the states bordering Thailand and, except for this area, free movement of dogs was permitted throughout Malaysia.

In August 1963, small outbreaks of rabies occurred in Perlis, the most north-west state, and subsequently in Selangor. Epidemiological investigations indicated that rabies entered Perlis through dogs from Thailand and was carried to Selangor by army dogs air-transported from Perlis. Rapid laboratory diagnosis prompted swift and effective control measures against canine rabies, and with mass dog vaccination and elimination of unvaccinated dogs, the outbreaks were quickly suppressed before they could spread beyond the two states.

In 1965, sporadic cases of canine rabies were diagnosed in Kelantan, another state which borders Thailand. Intensification of control efforts quickly halted the spread of the disease. In 1970, an outbreak involving both dogs and man occurred in the Kedah and Perlis states along the Thai border (Fig. 1) and a single canine rabies case was recorded in Selangor. This outbreak is described in detail in this communication.

The Control of Rabies in West Malaysia

Reservoirs of rabies

In all outbreaks, dogs have been the principal animals involved. Elimination of dogs as susceptible reservoirs has led to the disappearance of the disease. No wild animal reservoirs have been incriminated in any part of the country. A survey of 478 bats of 12 species live netted in 17 localities in West Malaysia yielded no evidence of rabies upon laboratory analyses (Tan et al, 1969).

The national rabies control programme of Malaysia is based on continuous compulsory vaccination of all dogs in an immune belt along the Thai border and in any rabies-infected areas, continuous destruction of unvaccinated and stray dogs in the immune belt and in any rabies-infected areas, legislative regulation of dog movement, dog registration and public education on rabies control.

Compulsory vaccination

Triennial vaccination with Flury LEP strain rabies vaccine and annual registration of all dogs 3 months of age and over are compulsory in the

immune belt. Mass vaccination and registration of dogs south of the immune belt are mandatory only during outbreaks of rabies. When the presence of rabies is confirmed in previously rabies-free areas, the areas, states or adjoining states, if outbreaks occur near contiguous borders, will be declared rabies-infected and vaccination of all dogs within the declared zones will be mandatory. Dogs in such zones must also be maintained confined within enclosed areas, tied securely, or led on strong chains, or leather or cord leashes as long as rabies remains present.

Prior to 1952, Semple type brain tissue vaccines were used for immunisation of dogs. During 1952-1953, field trials proved Flury LEP strain attenuated live virus vaccine more effective if properly handled. During this extensive outbreak, the vaccination of 80% of the dog population with Flury LEP strain vaccine successfully removed rabies from the campaign areas.

Destruction of unvaccinated and stray dogs

Destruction of all unvaccinated dogs within the immune belt and infected zones is compulsory. In uninfected areas, efforts are made to destroy stray or unowned dogs. Even a slight relaxation in stray control, especially along the Malaysia-Thailand border, can lead to rapid development of a potentially dangerous stray dog population.

Rabies control legislation

The Animal Ordinance, 1953, of the Federation of Malaya prescribes the declaration of rabies-infected areas, including an immune belt along the Malaysia-Thailand border, and the compulsory vaccination and licensing of dogs within infected areas to suppress the disease. The legislative acts are designed to prevent the entrance of rabies into Malaysia from other countries by land, sea or air routes; to remove the population of stray and unowned dogs; to build up the population of immune dogs above the level to prevent rabies transmission; and to limit spread of any focal outbreaks of the disease.

Public education on rabies control

Press releases, radio, television and rediffusion features and announcements, cinema slides, brochures for public distribution and posters are used to give advance publicity and provide public information on rabies control programmes. It is not difficult to stimulate public interest in rabies control measures during outbreaks but continual efforts are necessary to maintain a sustained public response.

The immune belt

The entire state of Perlis and the districts of Kedah, Perak and Kelantan states which border Thailand are maintained as a canine rabies immune belt 30-50 miles in depth. Continual compulsory vaccination and registration of dogs and destruction of stray and unlicensed dogs are maintained to prevent the spread of rabies introduced through dogs infiltrating from Thailand. Movement of dogs out of the immune belt into the rest of Malaysia requires certification of rabies vaccination performed at least 30 days but not over 3 years previously. Dogs may be brought into the immune belt only upon permission of the state veterinary officer of the state being entered; immediate vaccination upon arrival, if not performed earlier, is mandatory.

The effective maintenance of the immune belt is integral to rabies control in Malaysia. Although this effort has been historically successful, recent communist activities and the necessity for greater security in the border area have hampered the effective maintenance of stray dog control. Dog shooting has had to be curtailed, especially during the night, to avoid alarming the public which might confuse any shooting as terrorist activity.

The Rabies Outbreak of 1970

Epidemiology

Epidemiological investigations of rabies in people and dogs in northern West Malaysia during 1970 provided evidence that stray dogs entering from Thailand, but not destroyed after crossing the border, introduced rabies in several areas of the immune belt. Dog vaccination and destruction records from Perlis and Kedah showed a reduction in numbers of dogs vaccinated and killed from 1967 until the outbreak, explained by the absence of the disease and by heightened unrest in the border area. Unfortunately, no records were available to determine whether there was an increased incidence of rabies in southern Thailand at the time of, or preceding, the outbreak in northern West Malaysia. During the first half of 1970, rabies spread rapidly throughout Perlis and Kedah, and 5 persons, all of whom were bitten by stray dogs, died of the disease before it was again brought under control.

Human rabies cases

The first victim, a 9-year-old boy living in Bukit Ketri, Perlis, 5 miles from the Thailand border, was exposed during January but received post-exposure immunisation. He was admitted at the State Hospital in Kangar in April with fever, backache, restlessness and inability to swallow for 3 days. At admission his temperature was 100°F, he was markedly dehydrated, and experienced spasmodic throat contractions upon sight of water. Only by retracting his neck could he reduce the spasms of the muscles of deglutition enough to permit him to swallow. The patient died the same day. Necropsy was performed and the brain sent to the Institute for Medical Research (IMR) for analysis. It was eventually confirmed positive for rabies.

A 30-year-old Malay woman in Kodiang, 10 miles from the Thailand border, Kedah, was bitten by a stray dog during February. The dog was promptly killed and buried and the woman was given 14 doses of anti-rabies vaccine (ARV). Approximately 3 months after exposure, she was admitted at the Kangar Hospital with clinical diagnosis of rabies; she died 2 days later.

A 61-year-old Malay man and his 60-year-old Chinese friend were bitten in the same district during April by a stray dog which was also promptly kiled and buried. Neither received any prompt anti-rabies prophylactic vaccination. The Malay man was admitted at the Kangar Hospital one month later with clinical diagnosis of rabies. He was started on ARV at admission but expired 6 days later. His companion was started on ARV at that same time and received a full course of 14 daily injections plus 1 booster dose 30 days later; he is still normal at the time of this writing.

A 10-year-old Malay boy in Bukit Ketri, Perlis, was bitten during the same months by a stray dog which escaped. He was not given ARV and died

rabies. Among the 4 animal bite victims known to these authors and not given anti-rabies vaccine (1 was given vaccine only after symptoms of rabies were already manifest), all died of the disease.

A total of 4 dogs, 1 cat and 1 human brains was confirmed in the laboratory as rabies positive. All of these were negative for Negri bodies and fluorescing antigens but were unequivocally positive by mouse inoculation tests. These perplexing laboratory observations are discussed in this communication and have led to a suggestion by one of the authors (DSKT) that the rabies strain involved in this epidemic may have been a variant strain with low Negri body forming characteristics in the field cases.

Acknowledgements

Grateful appreciation is expressed to Mr. Mohamed Omar and Mr. Ali Amin for their technical assistance, and to medical and veterinary officers involved, for assistance in collection of necropsy specimens and epidemiological data.

Addendum

In January 1972, a dog was reported to have bitten a person and a buffalo in Jitra, Kedah. The dog was beaten to death and the brain sent to the IMR Virus Research Laboratory for investigation.

The preliminary findings were similar to those reported in this paper. The original specimen again did not show any typical Negri bodies or antigen by the FA test but the mouse inoculation results were positive. The brains of the infected mice all showed clear-cut Negri bodies in the stained smears and in the histological sections and were markedly positive in the FA test.

Samples of the original brain were sent to GWB (one of us) for more detailed study. Work is still in progress.

References

Cherry, W. B., Goldman, M. and Carski, T. R. (1960) Fluorescent Antibody Techniques in the Diagnosis of Communicable Diseases, Atlanta, Communicable Disease Center.

Smith, C. E. G. (1953) A red-green contrast stain for the routine demonstration of Negri bodies in brain-tissue sections. *Jour. Path. Bact.* 66, 309-310.

Tan, D. S. K., Beck, A. J. and Omar, M. (1969) The importance of Malaysian bats in the transmission of viral disease. *Med. Jour. Malaya* 24, 32-35.

World Health Organisation (1966a) Expert Committee on Rabies. Fifth Report, Geneva, W.H.O. 40pp.

World Health Organisation (1966b) Laboratory Techniques in Rabies. Second Edition, Geneva, W.H.O. 178 pp.