Kyasanur Forest Disease.

By

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Formerly with the Indian Council of Medical Research, first as Entomologist at Virus Research Centre, Poona, later as Senior Scientist with WHO/ICMR project, and retired as Director of Vector Control Research Centre, Pondicherry (October 2017) Many vector borne diseases occur suddenly in epidemic form when they are actually recognized, sometimes sporadic, and, some times these were not even reported.

But No body ever looks for the disease agent in the ecosystem, where it may occur in animals in a ENZOOTIC form.

The investigator should be:

A Mr. KNOW ALL Required:

He should know the people, the environment, be familiar with agricultural practices and forest, climate, human and animal population data, their movements, the vectors and all kinds of animals prevalent in the area, he should be a Ecologist, Entomologist/parasitologist /microbiologist and what not, and, above, be someone who satisfies everyone. Can we hope for someone like the late K.F.Meyer, an authority on Zoonoses, my role model ?

Here is a presentation about

investigations conducted on a disease, with unknown etiology and which had broken out in an epidemic form in many villages adjoining forests in Sagar and Sorab Taluks of Shimoga District.(1957-1970).The disease was named

KYASANUR FOREST DISEASE

since the first virus isolation was made from a dead monkey from Kyasanur Forest..



Early investigations..... Panicky Reaction

- Since Yellow Fever as a disease, transmitted by a mosquito, Aedes aegypti, was not reported in India, there was considerable excitement in both scientific and Health circles. The Karnataka Government at that time ordered an emergency, declared a TYPHOID epidemic, patient care intensified, and free distribution of antibiotics initiated.
- The Virus Research Centre started investigations by looking for Aedes aegypti in the affected area !!!

Mosquito studies

- Showed there was no Ae. aegypti, no distinct canopy biting and ground biting mosquito Fauna;
- Also no virus was isolated from many pools of biting mosquitoes tested.
- Being dry season, there were not many mosquitoes even!



Before sending the workers to the field, all were vaccinated with **17-D strain of Yellow Fever** vaccine, to protect the staff. As a routine procedure, prevaccination and second blood samples were taken from all field personnel working and tested for Virus.

Results of blood test.

- Firstly, Group B viral antibodies were found in some of the blood samples.
- Most significantly, a strain of virus was isolated from blood samples collected from 4 insect collectors. This strain was identified as a virus belonging to Russian Spring Summer Encephalitis group.

Virus isolations were made from

• Febrile blood of Insect collectors of VRC,

 From Blood from sick monkeys, man, and from tissues of dead monkeys, even remnants of tissues from almost dried carcasses of monkeys, initially. All the virus strains isolated were the same, belonged to RSSE group, and was named after the forest from where the virus isolated, namely, KYASANUR FOREST adjoining Barige village, Sorab Taluk, Shimoga district, Mysore State

Since RSSE is a tick borne virus

- Immediately all mosquito studies were given up, and studies started on TICKS.
- This is the first comprehensive study anywhere in the world, where a group of Young scientists and workers started the
- Investigation of A disease with unknown etiology

PKR Dissecting the first monkey found dead in the forest, and the organs harvested





A dead monkey (Langur) being autopsied in *Situ* in the forest. Over 1,000 monkey deaths have been investigated and over 130 isolations of KFD virus have been made from dead monkeys. Intensive studies started to collect ticks and identify the tick vectors involved.

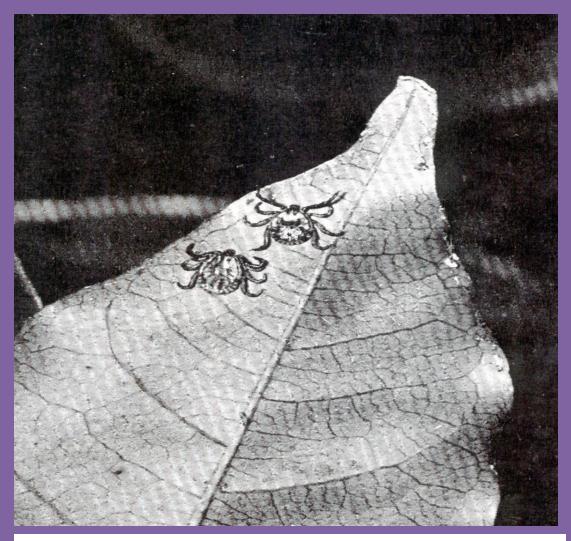
- Massive numbers of ticks were collected.
 Virus was isolated from nymphs of
 Haemaphysalis spinigera; and H.turturis, initially.
- Subsequently several other species of ticks were also found positive for virus.



A flag being dragged on the forest floor to collect ticks. Tick, which are questing for prey, cling to the flag. PKR



Ticks collected on the flag being picked off.



Ticks (*H. papuana*) on the underside of a leaf on a bush in the forest. From such a situation they await the arrival of a suitable host to cling on to (H. R. Bhat).

There are many species of ticks involved.....

- In the natural cycle of KFD, some species with wide host adaptability; some with restricted hosts. Even different stages of ticks show different host adaptability.
- Most of the ticks are three host ticks, meaning each of the three different stage, larva, nymph and adult needing a different host.
- This is of great epidemiological significance.

Haemaphysalis sp.

The vector of KFD is an arachnid, and transmits infection to man. Picture on the left is a male, and on the right is ventral side of a female tick which shows the genital aperture on the anterior side, and the anal aperture on posterior side.





Ventral side of head of Haemaphysalis tick.

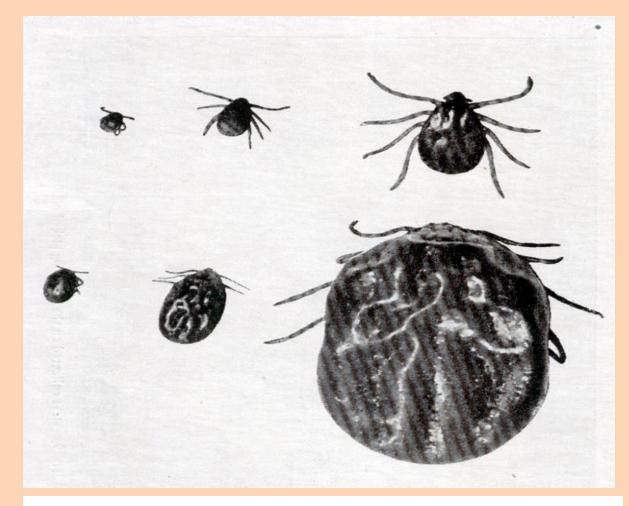


A median hypostome which has rows of backwardly directed teeth; a pair of chelicerae lying close together immediately above the hypostome.

Once the mouthpart is inside the host, the BACWARDLY DIRECTED teeth will prevent easy detachment form the host, unless the tick feeds to depletion !

A female Ixodid tick laying eggs





Haemaphysalis spinigera, the tick vector of KFD. Six other species of Haemaphysalis and one species of Ixodes area also known as vectors but *H. spinigera* is the tick which transmits the disease to man. In the top row are the unfed and in the bottom row are the fed ticks. The disease is more similar to OMSK hemorrhagic fever and the natural vectors are *Ixodid* ticks. Since the first isolation of KFD virus was from a dead monkey, monkey deaths were regarded as the first sign

of an approaching human infection in a given area.

KFD episode...

During the period 1957-2004, a total of 24721 human cases and 6475 monkey deaths were recorded.

More than 80% human cases and 64% of monkey deaths recorded were from Malnad belt of Western Ghats and the remaining were from other areas.

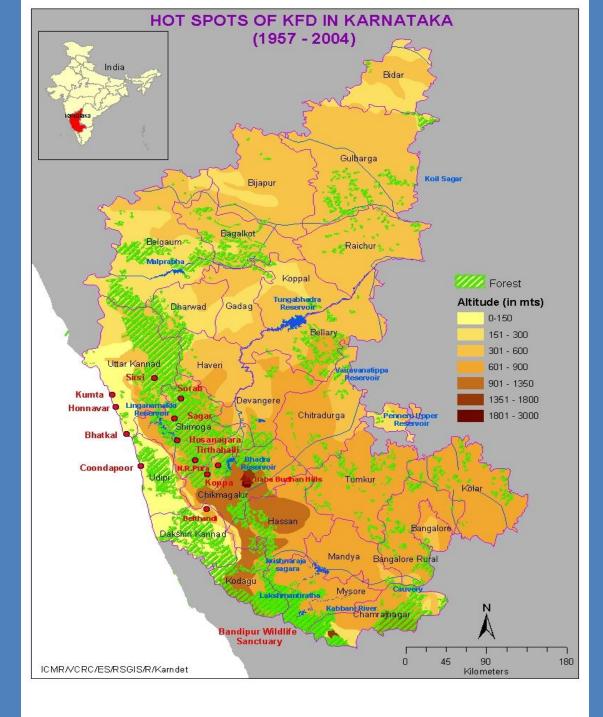
All these may be underestimates since there is a distinct possibility of many human cases not being reported and monkey mortality not being recognized.

Human cases & monkey deaths during 1957-2004

Taluk	Human Cases	Monkey deaths
Sorab	2761	2321
Sagar	3512	319
Hosnagar	3683	592
Thirthahalli	6786	399
Sirsi	239	64
N.R. Pura	1050	58
Корра	772	68
Belthangdi	1092	324
Other areas	4826	2330
Total	24721	6475

Since 1957, epidemics of KFD occured every year.

- The number of cases averaged 400-500 a year, with a low incidence in 1961 and a high (more than 1000 cases) in 1976, 77 and 83.
- For about a decade after its discovery the disease was restricted to a small part of the forests of Shimoga District.
- But in more recent years it has been found in parts of
 North and South Kanara districts. The incidence in
 Belthangady taluk of South Kanara has been one of the
 most severe so far.



PKR



A village in the KFD affected area in Sagar Taluk, Mysore State. These villages are situated in clearings in a belt of evergreen forest.



Presbytis entellus (the Langur): The monkey species most susceptible to Kyasanur Forest Disease.



Macaca radiata (the Bonnet monkey), which together with the Langur compose the monkey population of Sagar-Sorab area. This species is also highly susceptible to Kyasanur Forest Disease.

Man is a dead end host and there is no man to man transmission through any vector

- Virus infects man, monkeys, several species of small and large animals which are infected by ticks
- In the transmission cycle, many species of animals play a minor or secondary role in either the transmission or in maintaining the natural cycle

Isolation of KFD virus from ticks collected from forest floor and vegetation, by months (1961 – 64)

Month s	H. spinigera		H. turturis		Other Sp.	Total No.	Average rainfall in	
	Ν	Ad.	N	Ad.	N+Ad.	Isolates	mm	
Jan	10	0	1	0	4	15	0	
Feb	78	0	31	0	22	131	3.55	
Mar	24	0	88	0	19	131	23.11	
Apr	33	0	84	0	10	127	44.7	
Мау	13	0	39	1	17	70	179.52	
June	6	6	19	14	18	63	233.4	
July	0	62	1	42	7	112	961.4	
Aug	0	38	0	31	4	73	718.0	
Sept	0	19	0	29	4	52	158	
Oct	0	17	0	12	1	30	158	
Nov	0	8	0	6	0	14	34	
Dec	0	1	0	7	1	9	13.5	

KFD virus isolation from questing ticks collected by handpicking and dragging (1961-1972)

		Number of ticks			Number of isolations	
Species	Larvae	Nymphs	Adults	Larvae	Nymphs	Adults
H. spinigera	19	253946	88624	0	479	359
H. turturis	41	123738	53645	1*	431	265
H. kinneari	81	12597	26646	2*	26	49
H. kysanurensis	21	14712	629	1*	41	4
H. wellingtoni	0	11754	401	0	17	1
H.bispinosa	1	2907	702	0	11	0
H.minuta	0	2113	104	0	6	0
H.cusppidata	0	1517	509	0	11	2
H.aculeata	0	220	10	0	0	0
H.intermedia	0	0	181	0	0	0
H.cornigera	0	0	136	0	0	0
H.mixed sp.	18108	13386	928	0	30	0
l.petauristae	342939	4536	29	0	0	0
I.ceylonensis	8	62	10	0	0	0
D.auratus	15	10	1	1*	1*	0
Amblyomma sp.	0	62	0	0	0	0
Rhipicephalus sp.	0	4	0	0	0	0
Mixed genera	0	8	0	0	0	0
Total	361233	441572	172555	5	1053	680

• Disease in man occurs mainly during the dry months.

- Disease and death in monkeys also follow the similar course.
- The season coincides with the season of abundance of nymphs of two main species of ticks, Haemaphysalis spinigera, and H.turturis

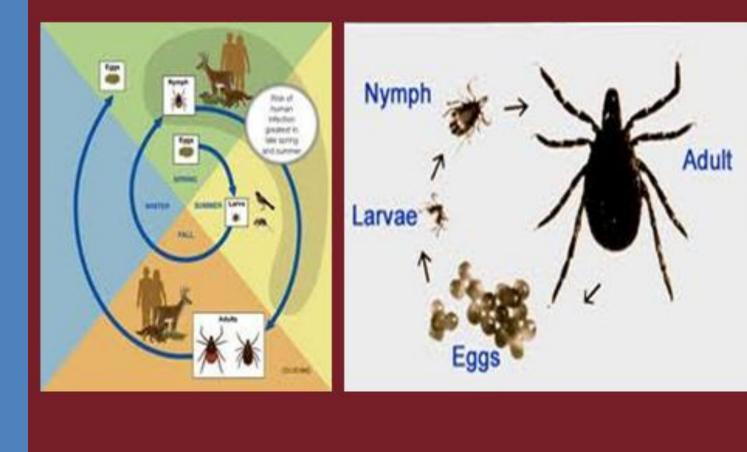
Seasonal cycles.....

- Adults of tick species) occur mainly during the rainy season. They have relatively limited host range.
- They attach on larger animals, mate, engorge, and drop to the gound where they lie dormant till the onset of drier months. The engorged adult females, lays eggs which moult into larvae.
- Fed adults are deposited in the rainy season in the forest floor and eggs laid in clusters.

Life cycle of tick in brief

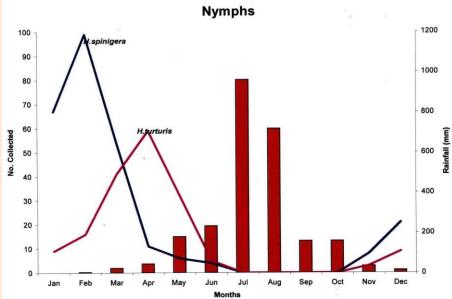
- The eggs hatch into thousands of larvae. They have limited mobility. They are picked up by moving small and large mammals and birds. All in the dry season.
- The fed larvae drop to the ground, moult into nymphs and then seek hosts, mostly small mammals and birds and MAN. The fed nymphs again drop to the ground, moult into adults and which attach to new hosts, mostly large animals.

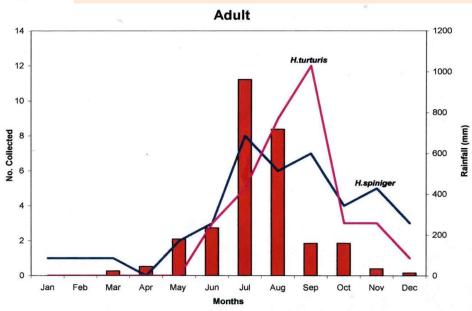
Life cycle of an Ixodid tick - summarized



They occur mostly in the dry season, from November to May, with peaks in February and March The disease in man and death of monkeys also follow similar course.

The season coincides with the season of prevalence of nymphs of Haemaphysalis spinigera, the only species of hard-tick found to infest man in any numbers. Adults of Haemaphysalis ticks occur during the rainy season from June to October





- Fed larvae moult into nymphs and drop to the ground. They have wide host range as larvae.
- The questing nymphs got infected as larvae (transtadial) - then attack other hosts like man and monkeys, and also several other species of small mammals.
- The process of infection continues for several months as broods of nymphs emerge from the fed larvae which drop to the ground throughout the season.

It has now been shown convincingly that the small mammals themselves become infected by the progeny of a few infected adults and nymphs of the previous season which had TRANSGRESSED into the new season.

- This was an important link for the survival of
- the Virus from season to season

 Man and monkeys get infected mostly by the nymphs of *H. spinigera* and *H. turturis* and the peak season of transmission is from January to March when questing nymphs are abundant. The small mammals like rats, mice, squirrels, shrews and bats were also found infested by ticks. The large animals such as cattle, deer etc. are infested mostly by adult ticks. A few hungry nymphs persist through the monsoon season and may be one of factors responsible for triggering a new cycle of infection in the next season

- Cattle play a very important role in KFD epidemiology, though the virus does not circulate to any significant levels in them. But they are the main multipliers and distributors of vector population.
- In the monsoon months the adult tick parasitizes cattle and the females dropped in the forest floor deposit enormous numbers of eggs, which remain dormant in the soil till the monsoon ends.
- The cattle roam about all over the forests and perhaps constitute the biggest means of tick production and DISTRIBUTION.

• The human disease occurs mostly during the dry season, coinciding with monkey mortality.

- During the dry season the nymphal stages of the major vectors are abundant, whereas the adult stages are preponderant during the monsoon.
- The occurrence of human disease and preponderance of nymphal stages of *H. spinigera* and *H. turturis* coincided

Study of ticks biting man

During the epidemic season over 4600 forest frequenting villagers were examined for ticks

- On them; an average of 2.5 nymphs per person were collected; mostly *H.spinigera and H.turturis*
- There were also larval ticks collected from man,but this not have any epidemiological significance.
- No adult ticks were found biting man

Role of Bats in KFD Cycle

- KFD virus was isolated from the spleens of four insectivorous bats, *Rhinolophus rouxi* and one from a pool of *Ornithodoros* ticks collected from the same habitat, a disused well.
- From the 271 bats examined, no *lxodid* ticks were found.
- The role of bats in the natural cycle of KFD is yet to be understood. May be there is a closed cycle of bat-Ornithodors-bat maintained in nature. It is worth mentioning that the bats have evolved about 40 million years ago (Oligocene period).

Role of wild animals

 The panther, Panthera pardus, the porcupine, Hystrix indica and the wild dog, Cuon alpinus accounted for the largest number of Haemaphysalis adults collected which is very significant.

Ticks like *H. spinigera* and *H. turturis,* once parasitic on wild animals, might have left the original hosts and adapted themselves to alternate hosts like cattle which have moved in with man in the new areas. The construction of Sharavathy Hydro electric dam, resulted in the inundation of large forested areas, and the creation of new virgin land attracting man and his cattle.Contd

- Ticks like *H. spinigera* and *H. turturis,* once parasitic on wild animals, might have left the original hosts and adapted themselves to alternate hosts like cattle which have moved in with man in the new areas.
- The construction of Sharavathy Hydro electric dam, resulted in the inundation of large forested areas, and the creation of new virgin land attracting man and his cattle.
 A new biocoenotic relationship was the result

Adult Haemaphysalis ticks parasitizing 3 species of wild mammals

	No. + / No. Examined	H. spinigera			H. turtuis			H. kyasanurensis			
		L	N	Ad	L	N	Ad	L	N	Ad	
<i>Panthera pardus</i> (Panther)	5/5	66	10	46	151	14	705	-			
<i>Hystrix indica</i> (Porcupine)	15/15	2	77	0	162	603	488	9	117	511	
<i>Cuon alpinus</i> (Wild dog)	2/2	3	3	3	13	1	483	0	7	1	

Apart from monkeys, there are several species of small mammals (Rodents and shrews) in the forest, which are found heavily infested by ticks

Ticks on small mammals

Ticks engorging on a small mammal



Table – 4

Species of ticks collected from 3 small mammal species - 1961 to 1964

Species	No. examined	H.sp		H.t		H.k		D.aur		Ixodes		
		Larvae	Nymph	Larvae	Nymph	Larvae	Nymph	Larvae	Nymph	Larvae	Nymph	Adult
R.r.w	3551	104	63	98	3	884	1174	1967	672	8546	2916	31
R.b	860	114	59	16	0	559	1112	2139	362	2714	804	5
S.m	1571	[.] 4	1	6	0	14	12	453	29	6822	1051	0

R.r.w : Rattus rattus wroughtoni

- R.b : Rattus blanfordi
- S.m : Suncus murinus

- H.sp : H. spinigera
- H.t : H. turturis
- H. k : H. kinneari
- D.aur : Dermacentor auratus
- Ixodes (I. petauristae and I. ceylonensis)

How the villagers get exposed to the disease ?

- The villages are situated in the clearings in the middle of the forests, or at the edge (Interfaces).
- They have to walk through forests to go to another village or for any other chores within the forests like fetching firewood or gathering forest products.
- There are only narrow foot paths to walk through in the forests.
- They get infested by ticks in the forest and get infected with KFD

It can also infect a wide variety of birds and mammals such as jungle fowl, the red spur fowl, the

- mammals such as jungle fowl, the red spur fowl, the porcupine, bats and rarely even ruminants, apart from monkeys.
- There are possibilities that there are more than one independent natural cycle involving different vectors and hosts.

- The virus is entirely new to science and though it has close affinities with a few others, particularly the Omsk Hemorrhagic Fever (OHF) virus, KFD is perhaps the only disease in which ticks and monkeys are involved
- and one among only two or three arbovirus diseases in which monkeys are affected.

- The disease, in spite of numerous apparent opportunities, was restricted until recently to a comparatively small-forested area.
- The epidemiology is however most complex, even among arboviruses, more hosts and more vectors take part than with any other arbovirus disease

•For how long before 1955 was the disease in monkeys known ?

Some vague statements that monkeys had died in the area in 1918 exist but they were wholly unreliable. It would be difficult to believe that such a susceptible animal as the monkey would have remained unaffected if the virus was active in the natural cycle involving their ectoparasites.

Therefore, in all probability, the virus entered the Ectoparasitic world of the monkeys somewhere in 1954 or 1955.

Ticks themselves can be reservoirs as well as vectors.

Once infected as a larvae, the virus is passed <u>transtadially</u> from the larvae to the nymph and from the nymph to the adult, enabling the virus to persist in the habitat all the year round. Virus has been recorded in *H. kyasanurensis* adults 365 days after infection as larva. There are also reported instances of virus found in a tick for over 400 days after it was infected as a larva.

Where did the virus come from?

- Since KFD virus belong to the RSSE group, it was thought by some that birds migrating from Soviet Union must have visited KFD area and introduced the infection.
- A very extensive study of birds of Shimoga district where 8474 birds belonging to 184 species were collected and examined, and 1082 birds of 81 Species were found to be infested by ticks.
- The genus *Haemaphysalis* accounted for 99.5% of all the ticks collected and the remaining belonged to genera *Dermacenter, Amblyomma, Boophilus* and *Hyalomma. H. spinigera* was the commonest species, followed by *H. wellingtoni* and *H. turturis.* There was no evidence of birds bringing ticks not indigenous to India. (Ph.D.work of Dr PKR)

An enzootic cycle ?

• Was there a slow lurking enzootic Cycle involving small mammals and shrews in the forested areas (*Shrew-Ixodes-rodent-Haemaphysalis*) and their triggering a chain of epizootic in monkeys and epidemic in man ? Could be.

The small mammal and rodent cycle

• Small mammals were infested by several species of immature stages of *Haemaphysalis* and *Ixodes*.

•The shrew, Suncus murinus, apart from being capable of circulating high titres of virus for longer duration harbours very large numbers of *Ixodes* larvae and nymphs.

•Transovarial and transtadial transmission of KFD virus were demonstrated in *Ixodes*, adults of which are more prevalent in the dry season,

If the virus had entered this territory for the first time in 1955 from where did it come?

There is no other place in the world where an identical virus is known to occur. It may be occurring in a covert cycle but no one has yet recognized it. A study of ticks infesting birds in the KFD area did not show a single tick of distant lands, but this was not a final proof. If ticks falling off the bird had been the cause of starting the new cycle, why has it not occurred in other areas of India?

By pure laws of chance infected ticks must have fallen off at many places in and out of India. Why then was there no report of KFD-like disease anywhere else?

WAS KFD PRESENT

In this area even prior to 1955-1956, occurring in a natural cycle in which neither monkeys, the small

mammals and *Haemaphysalis* ticks were involved ? There is no record.

- The isolation of KFD virus from *Rhinolophus* sp., an insectivorous bat, in the Sagar area is significant. Virus was also isolated from softticks collected from the same habitat of the bat indicating a virus-soft tick-bat cycle.
- It could occur in nature completely isolated from the rodent- *Haemaphysalis*-monkey-man complex.

 KFD could have existed in an occult enzootic cycle in the forests without anybody knowing about it. At some stage the virus could have "transgressed" the exclusive cycle and entered the rodent-hard tick-complex.

- KFD is also possibly one such virus of the forests existing in an in apparent vector host cycle and had remained unnoticed.
- It is a virus, which can infect a variety of animals and birds and also several species of ticks.
- The period of viraemia in small mammals except the shrew, is of short duration, which means a quick population replacement is necessary. The reproduction cycle is short. Fresh individuals arriving, ensure the availability of many non immunes.

Spread of the virus

- For many years till 1967 or 1968, the virus remained more or less in the same area where it was found.
- The massive flowering of the bamboo which is said to occur only once in about 12 years also caused a large epidemic among people who visited the area near Sagar Town in 1967.

• The total area now infected is perhaps 4,000 Sq. Kms.

- The old theatre is still active with a few monkey deaths and human cases occurring every year. It is not as if the infection has burnt out in the old areas but the disease has spread to new areas.
- The manner of spread and the factors governing it are extremely important not only from the purely scientific but also from a practical point of view.

- Is KFD exclusively a disease of the forest?
- Does it need the climate and vegetation of the forest?
- Are the tick vectors specially adapted only to this environment?
- Are the now recognized natural hosts and their
 biology dependent upon the characteristics of the forest?

The main vector encodes of User anhyselis such as U

The main vector species of *Haemaphysalis* such as *H*. spinigera, H. turturis, etc. and the major natural hosts such as R. blanfordi, R. r. wroughtoni and even Suncus *murinus* are not denizens exclusively of the Sagar forests but are quite widespread. Therefore, why has there not been a more rapid spread of the disease? These are baffling questions. One has to recognize that the prevalence and survival of an organism like an arbovirus needs a combination of many factors all occurring in adequate quantities and of appropriate quality at a given time. Perhaps the Sagar area provides the best type of combination for it (Boshell's cup of coffee)

Late Dr. Boshell, my mentor in epidemiology, had stated the factors influencing the numbers of natural hosts and their vectors and had dealt with two types of exploitation of the forest which may have a bearing on the subject:

• Steady expansion of the paddy cultivation at the expense of the forest,

• The increased cutting of the forest trees for timber or firewood.

These led to miles and miles of "INTERFACES" between Forest and open land. His view was that it was this interface which provided greater opportunities for the field rodents and their vectors to multiply

Ecological changes ?

- Could the present EXPLOITATION of the forest be responsible for the introduction and persistence
- of the disease ?
- Does the conversion of a wet evergreen forest (e.g. Sharavathy forest) had any influence on KFD virus indirectly through the hosts and their ectoparasites ?

Long-term changes in the ecology can and do bring about great changes in the fauna and flora. The exploitation of the forest and the probable increase of interfaces has been going on for centuries.

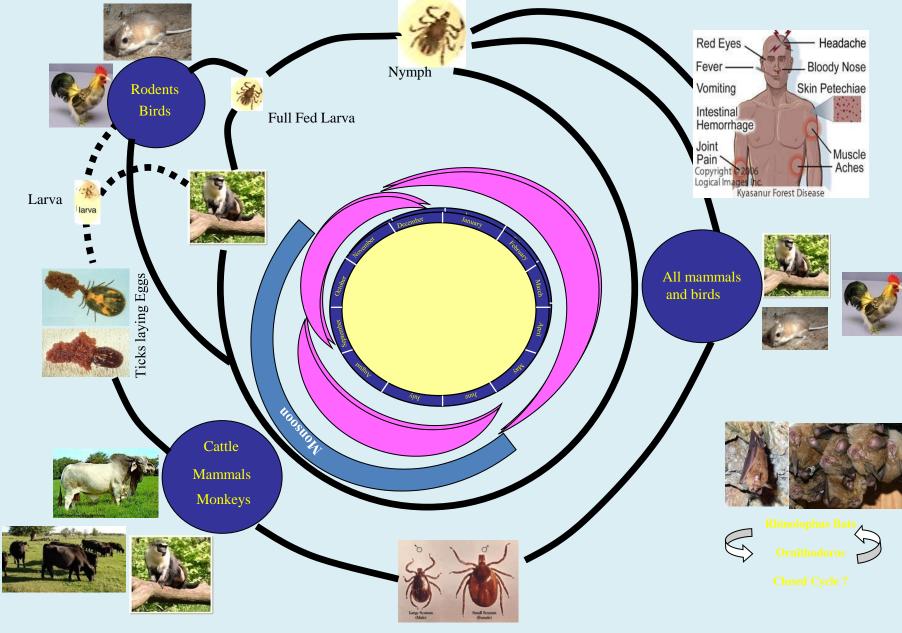
Also for centuries there has been a practice of the so-called "Kumri" cultivation adopted by the local people. It is the burn-cultivate-abandon method.

Many localities in the forest were cut down and burnt, cultivated for a few years and later abandoned. Such practices could also have led to creation of such interfaces.

The role of the habits and migration of monkeys is not clearly understood.

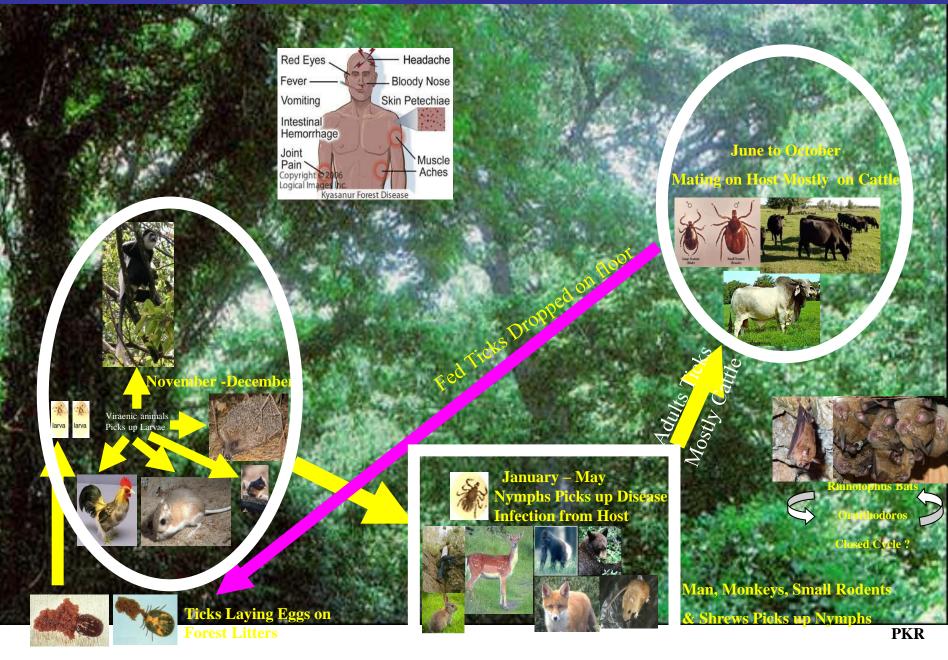
The question of home ranges, decimation of monkey populations due to KFD and the possibilities of some infected monkeys escaping out to nearby infected areas and introducing the virus to such localities have also to be considered.

Natural cycle of KFD virus



Adult Ticks

Natural Cycle of Kyasanur Forest Disease (KFD)



In Summary....

(1)

Since 1951 the human population of the Sagar-Sorab area had more than doubled, and the consequent alterations of the ecosystem have produced a set of conditions favouring overt expression of a hidden enzootic process.

(2)

Man's low rate of attack by

H. spinigera is offset ,and the probability of his infection increased by increased numbers of infected vectors

(3)

Experimental exposure of monkeys to the bite of tick larvae in the forest suggests that monkeys well may serve as efficient amplifiers in the KFD-virus cycle.

(4)

Adult ticks have been shown experimentally to be efficient transmitters of the virus

(5)

Adult ticks have been found attached to sick and dead monkeys. *Haemaphysalis* have been observed in nature to carry the virus in nymphal and adult instars for up to 14 months, spanning three seasons

(6)

Several species of small mammals in the Sagar forest have been found capable of multiplying KFD virus in their systems, and the virus has been detected in tick parasites of the genera *Haemaphysalis* and *Ixodes*.

(7)

The probabilities that the virus may have been imported into the Sagar area are considered and evaluated as not likely.

(8)

Immunity surveys carried out as early as 1952 and on later occasions indicate that **KFD virus (or some closely related agent)** has been active inconspicuously in various other regions of India, of contrasting ecology and widely separated from each other

• KFD is therefore an example of the interplay of many ecological and ethological features. But for the unfortunate occurrence of KFD, this biocoenose would have remained unknown. In spite of so much information being available, there are still many problems to be solved

- If, at present, KFD is not more generalized or frequent, the reason must be that
- one or more of the necessary conditions are either absent or below the required level of intensity; for the maintenance, latency, or explosion of most endemic and epidemic situations depends qualitatively on the coincidence of certain factors and quantitatively on the synchronization of their minimum thresholds PKR

There are still many gaps in our knowledge of KFD epidemiology and natural cycle.

For future studies of this nature, e.g. understanding the natural cycle of diseases some guidelines are given here,

Selection of study area should be carefully made. Ideal sylvan settings, interfaces, different ecotypes, etc.

Studies on biotic, climatic, and physical characteristics of the environment.

Demographic studies – all aspects.

Vector studies – all ecological parameters.

- Looking for small mammals, large mammals, birds, serological surveys, ectoparasite studies, etc. particularly population dynamics of small mammals, migration etc.
- Role of man, his life style, his domestic animals, etc to be studied.
- Investigation of enzootic foci in complex environments such as forests, river banks, swamps, ponds, deserts, fields etc.
- Outskirts of villages, interface between different ecotypes

	Reported occurrence of KFD virus in recent years in India
2012	100 confirmed cases in Karnataka; tick and monkey positivity
2012	KFDV confirmed in monkey specimen in Nilgiri, Tamil Nadu
2012– 13	Outbreak in the Bandipur National Tiger Reserve, Karnataka ; confirmed by human and monkey positivity
2013	Human case confirmed in Wayanad, Kerala
2014	Outbreak in seven health centers in Thirthahalli , Shimoga, Karnataka
2014	Human case confirmed in Wayanad, Kerala
2014	Outbreak in a tribal population, Malappuram, Kerala
2014	Anti-KFD IgG antibody positivity in a tribal population of the Palakkad and Wayand districts, Kerala
2015	Confirmed in monkey specimen in Nilambur, Malappuram, Kerala
2015	Tick positivity for KFDV in Wayanad, Kerala
2015	Outbreak in Wayanad, Kerala [18 confirmed cases; Pulpally (7), Mullankolly (8), Chethayalayam (1), and Poothadi (2)]
2015	Outbreak in Shimoga, Karnataka [35 confirmed cases]
2015	Outbreak in Pali village, Sattari Taluka, northeast Goa [18 confirmed cases]

A word about Diagnostic procedures

- Over the last decade or more, complete ,"Molecularisation" of almost all laboratory procedures have taken place.
- Now you have ready made commercially available
- test kits to diagnose everything under the sun.
- Every body supports this, because it is so EASY.

BUT HOW RELIABLE ARE THEY ?

I would like to end by quoting a famous saying "

"There is no substitute for hard field work. Understanding disease, understanding its nuances, learning about the interactions of the reservoir, vector, and host, and witnessing the interplay of many variables of each ecological niche; all this means the permanent and alert presence of the investigator at the end of the line ".

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The motivation was only our curiosity & challenges in the field. Those were <u>the golden years of field research</u> and your work was encouraged and appreciated by the powers that be.

Thank You