enzyme activity. The intensity of the colour developed by the substrate is proportional to the quantity of the enzyme on the bead and therefore is indicative of the amount of rotavirus antigen in specimen. The results of a diluent control, unknown specimen and the positive control assayed are compared either subjectively (visual method-comparison to a colour chart).

Rural Survey (Indakaw)

Study Area and Population:

The study area, study population and study period were the same as in the rural survey for bacterial aetiologic agents carried out by the Bacteriology Research Division.

Virologic Studies:

Rota cell kit, manufactured by Meguro Institute Co. Ltd., 29-7, Masu micho, Ikeda City, Osaka, Japan was utilized for the detection of rotavirus in the stool specimens. The test utilizes "reversed passive haemagglutination system" and involves a screening procedure and confirmatory procedure to identify and confirm the presence of rotavirus in the stool specimens. In the screening procedure fixed sheep red cells coated with antibody to bovine rotavirus (NCDV Virus) was utilized to detect the presence of rotavirus in the stool specimens. The presence of rotavirus in the stool specimens detected in the screening procedure was then confirmed by a blocking test utilizing antirotavirus serum. The test was done in a microtitre system. Due to limited laboratory facilities, only randomized subsample of diarrhoea stools and control stool specimens were tested for the presence of rotavirus.

Results

Table 47, 48 and 49 shows population distribution of under five years children, no. of diarrhoea and control stool specimens collected, and no. of subsampled diarrhoea and control stool specimens for virologic studies to be carried out during the various study periods.

Rotavirus was not detected in 166 diarrhoea stool specimens and 75 control stool specimens tested during the wet season study period in the rural survey. However, rotavirus was detected in 17 out of 116 diarrhoea stool specimens tested during the dry season study period (Table 50) in the rural survey.

In the urban survey rotavirus was detected 11 out of 78 stool specimens tested during the wet season study period (Table 51).

The highest prevalence was detected in the 0 to 11 months old age group in both the rural and urban surveys (Fig. 33 & Fig. 34).

Discussion

The aetiologic role of rotavirus in diarrhoea cases was first demonstrated in Burma by Electron Microscopic examination of diarrhoeic stools in 1978 (3). Rotavirus has also been demonstrated in the stools of gastro-enteritis cases in hospitalized cases of under 3 years old children (residing in Rangoon) throughout the year (4).

In the present study area rotavirus was not detected in diarrhoeic stools during the wet season study period in the rural survey. This could be due to two reasons. Firstly, it is simply that the rotavirus is not prevalent in the study area during the wet months. This rather unlikely as rotavirus was detected in diarrhoeic stools during the wet season in the urban survey (North Okkalapa). The two areas have essentially similar climatic conditions. Thus, the plausible explanation is that we may be able to detect rotavirus in the diarrhoeic stools if we have studied the whole wet season period (which lasts about four to five months) rather than only a month of the wet season. A similar unusual pattern of endemicity of rotavirus has also been reported from an area. In that area it was demonstrated that rotavirus was the cause of an outbreak of gastroenteritis among children which occured between August and September 1980, although it had not been detected in the population in the preceeding 13 months. Epidemics of human rotavirus associated with gastroenteritis appear to occur every second year in that population (5). The precise climatic factors involved in the seasonal distribution of human rotavirus have not been clearly defined, as is the means by

which the virus survives between the epidemics. However, in smaller population groups, outbreaks may be less frequent, for example every 2-3 years, presumably occuring only when a critical number of susceptible children are present (6).

Rotavirus is the most common cause of viral gastroenteritis in children. Most cases occur between the ages of 1 and 6 years, with a peak between 1 and 3 years. In some reported series the highest incidence has been in children aged between six and eleven months, the age when maternal antibodies have just been lost and children are susceptible to any passing virus. In more primitive communities rotavirus infections may occur slightly earlier in life. In a comparative study of Aboriginal and white children admitted to hospital in Western Australia rotaviruses were detected mainly between 6 months and a year in non-aboriginal children and in children less than six months of age in the Aboriginals (7).

Our findings are similar to the above reports as the model age group of rotavirus diarrhoea seems to be under 3 years. Moreover, rotavirus infections appear to occur earlier in life in this community. This study also provides evidence for the first time in Burma the prevalence of rotavirus in rural community.

Summary

A study was undertaken to determine the relative importance of rotavirus as an enteric pathogen in the causation of acute diarrhoea in children under 5 years residing in an urban and a rural community. The study was carried out during the dry season and wet season. Rotavirus was detected in 14.6% of diarrhoea cases during the dry season but no rotavirus was detected during the wet season in the rural community although rotavirus was detected in 14.1% of diarrhoea cases during the wet season in the urban community. The study provides evidence for the first time in Burma the prevalence of rotavirus in a rural community.

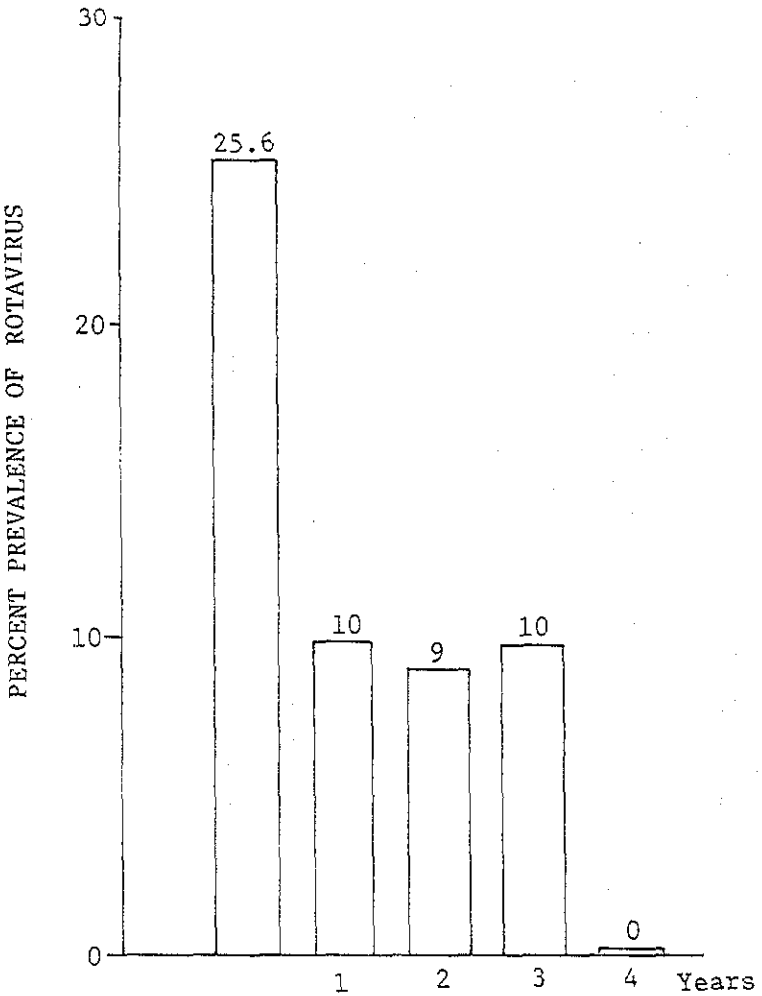

Figure 33 Age distribution of rotauirus prevalence in a rural community

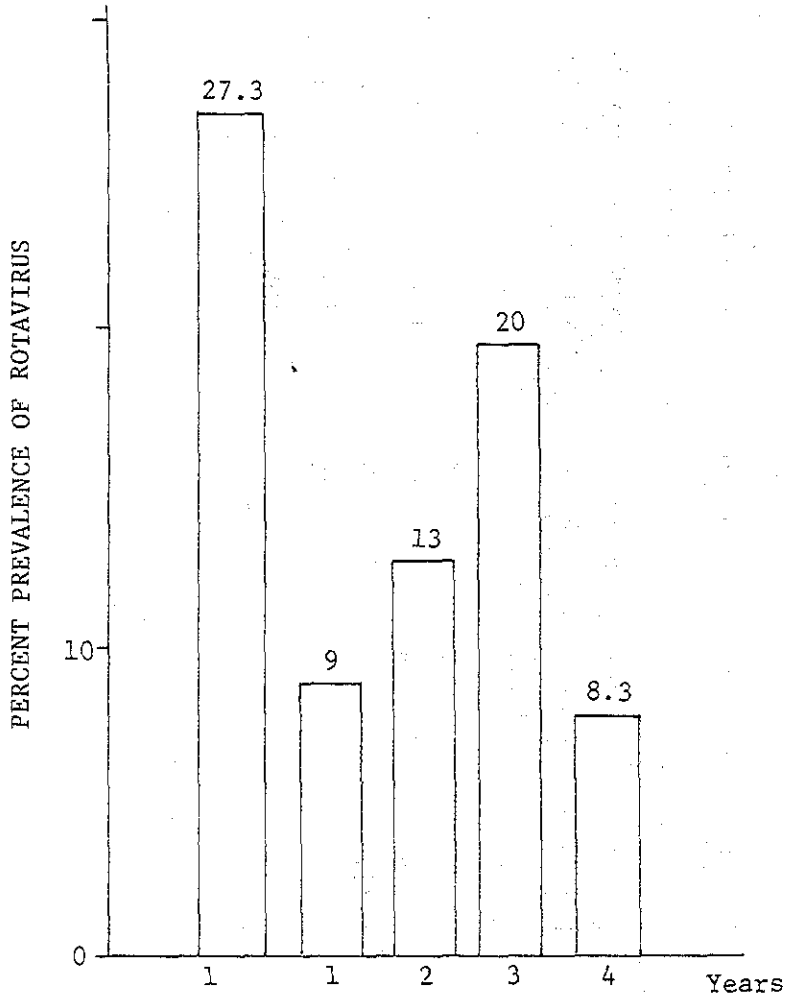

Figure 34 Age distribution of rotavirus prevalence in at urban community

Table 47 Data on study population of rural survey (wet season)

Age (months)	Under five total population	Surveyed Population		*Subsample Population	
		Diarrhoes	Control	Diarrhoea	Control
0-11	178	126	65	61	21
12-23	134	81	59	41	19
24-35	146	62	46	30	18
36-47	137	46	39	19	11
48-59	120	27	24	15	6
0-59	715		233	166	75

* to undertake virologic studies

Table 48 Data on study population of rural survey (dry season)

Age (months)	Under five total population	Surveyed Population		*Subsample Population	
		Diarrhoea	Control	Diarrhoea	Control
0-11	156	58	47	39	14
12-23	170	59	55	40	17
24-35	130	31	20	21	7
36-47	152	14	12	10	4
48-59	124	8	8	6	2
0-59	732	170	142	116	44

* to undertake virologic studies

Table 49 Data on study population of urban survey (wet season)

Age (months)	Under five total population	Surveyed Population		*Subsample Population	
		Diarrhoea	Control	Diarrhoea	Control
0-11	318	56	21	11	5
12-23	327	64	33	22	8
24-35	320	51	18	23	12
36-47	312	28	24	10	7
48-59	264	17	11	12	2
0-59	**1545	216	107	78	34

* to undertake virologic studies

** includes 4 children of unknown age

Table 50 Prevalence of rotavirus in rural survey (dry season)

Age (months)	Tested Population		Rotavirus Positive		Percent Positive	
	Diarrhoea	Control	Diarrhoea	Control	Diarrhoea	Control
0-11	39	14	10	1	25.6	7.1
12-23	40	17	4	0	10.0	0
24-35	21	7	2	0	9.5	0
36-47	6	4	1	0	0	0
48-59	6	2	0	0	0	0
0-59	116	44	17	1	14.6	2.2

Table 51 Prevalence of rotavirus in urban survey (wet season)

Age (months)	Tested Population		Rotavirus Positive		Percent Control	
	Diarrhoea	Control	Diarrhoes	Control	Diarrhoea	Control
0-11	11	5	3	0	27.3	0
12-23	22	8	2	0	9.0	0
24-35	23	12	3	0	13.0	0
36-47	10	7	2	0	20.0	0
48-59	12	2	1	0	8.3	0
0-59	78	34	11	0	14.1	0

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3-5 A Preliminary Study of the Biological Variation of Dengue 2 Virus Recovered from Different Disease Severity Manifestations in Dengue Haemorrhagic Fever.

Dengue infections has long been considered to be an annoying but benign disease occuring in various parts where the vector Culicines are present. (1,2 & 3). However since the last 3 decades a change in the clinical manifestations from the benign to severe form resulting with leading cause of hospitalization and death among children has been reported for in a number of countries in Southeast Asia (4). The life threatening forms of dengue are usually associated with haemorrhage hypovolemic shock or both and generally are described by the term Dengue Haemorrhagic Fever (DHF). Questions have been raised as to whether or not it is new, but it is clear that it is now more prevalent than any time in the known past (5). Although various descriptive hypothesis have been carried with regards to the pathogenesis of DHF, an universal acceptance of any particular hypothesis have not yet been achieved. Epidemiological studies to prove the sequential infection hypothesis with respect to the pathogenesis of DHF (6), and genetic variation of dengue virus have been carried out with the hope of furnishing insights to the pathogenesis of DHF. Further studies in relation to the variation in the biological characters as attributes of markers for virulence have been documented for Venezuela equine encephalitis virus, a member of group A alphavirus (7,8,9 & 10). These studies stimulated and initiated an attempt to study the biological patterns and variation among the dengue 2 serotype which has been the predominent common serotype recovered from DHF patients since 1976 in Rangoon (11). Further if differences in variation in the biological characters are seem, attempts to correlate with disease severity may reveal markers to specify virulent strains of dengue causing severe DHF.

Materials and methods:

Source of dengue viruses

The source of dengue viruses were from clinical material of patients admitted to Rangoon Childrens Hospital during 1976-1980

with a clinical diagnosis of DHF of grades I-IV. Grading of DHF was according to the recommendation of WHO Technical Advisory Committee on Dengue Haemorrhagic Fever (1975). In order to avoid confusion in classifying the disease severity grade I, & II have been classified as non shock (NS), representing milder infections in DHF, and grade III & IV as shock (S) the more Severer forms of DHF resulting with the Shock Syndrome (DSS). Clinical diagnosis of DHF was only done so after a positive Hess test generally done at admission. All impending shock cases of DHF are monitored by haemconcentration (PCV) test, so that all grades 3 & 4 are well categorized shock cases of varying degrees of severity and duration. Among some of the very severely ill cases of shock death was the ultimate end and in some autopsy liver and heart blood were processed for virus isolation.

Prototype virus, either as suckling mouse brain (smbr) or tissue culture fluid (TCF). Prototype strains of dengue were used to standardize test conditions for the various biological test systems. Dengue 1 (Hawaii), Dengue 2 (New Guinea) Dengue (H-87) and Dengue 4 (H-241), strains were used.

Isolation system

Dengue viruses were recovered by inoculation of Toxorhynchites Splenders by the intrathoracic route, and stored at -80°C deep freeze or in liquid nitrogen.

Identification and typing

Isolates will be identified as dengue by the direct fluorescent antibody test (DFAT) on mosquito head squash preparations, and further serotyped by direct FAT using type specific dengue monoclonal antibodies.

Biological characterizations were done by using plaque assay, effect of temperature on the growth of viruses, and mouse virulence.

Plaque assay

Monolayer culture of C6336 in 6 x 4 wells infected with tenfold dilutions of infected TCF preparation of wild dengue viruses. After

an adsorption period of 90 mins, an overlay medium of 1% methyl cellulose in maintainance medium was added and incubated at 34°C in a Co_2 incubator for 7 days.

Plaques were stained by the immuno-enzyme staining method of Kansai Medical University, Department of Microbiology, Osaka, with a slight modification in the use of high titered convalescent serum of DHF patients as the primary antibody and antihuman IgG (goat) conjugated with peroxidase conjugate from Miles and Yeda, Isreal. Infectious PFU and plaque size were determined and designated as small if the size is less than that of the prototype, intermediate if of the same size and large if larger than the prototype size.

Effect of temperature on the replication and growth of dengue virus in Vero cell cultures.

Monolayer culture of Vero prepared in Lab Tek chamber slides were infected with 10 fold dilution of the wild dengue viruses and adsorbed at permissive temperature of 37°C, and an arbitary non-permissive or restrictive temperature of 39°C for 90 imins and incubated for 7 days at these temperatures.

Replication was assayed by detection of viral antigen by the direct fluorescent antibody (DFAT) technique, and results were recorded as $IFID_{50}$(13) of the viruses by the method Reed and Meunch (14).

Effect of increasing temperature on the replication of wild dengue virus in Vero cell culture.

Replication of the dengue virus in Vero cell culture was studied at increasing temperature of 34°, 37° and 39°C on 3,5,7 days after incubation. Strains replicating at nonpermissive temperature will be designated as t^- temperature resistant, and if strains grew at 37°C only it was designated at t^+ temperature sensitive.

Virulence assay in mice

Preliminary experiments were done to assess the virulence of two randomly selected strains of dengue recovered from 2 different

clinical disease manifestation. Shock (S) and nonshock (NS) and a prototype strain. Each virus was titrated in newborn to 1 day old and weanling mice by the intracerebral (IC) intracerebral and intraperitoneal IC/IP, and intraperitoneal IP route. As the result of the experiment revealed (1) marked differences in lethality between the 2 age groups and (2) vague mixed response and resistant to IP infection at the standard does of 0.02 ml of the 10 fold virus dilutions. Consequently newborn to 1 day old mice were used for the comparative studies.

Serial 10 fold dilution in Hanks 2% △ FeCS were incubated simultaneously into groups of newborn to 1 day old mice by the IC, IC/IP and IP routes. Mice were observed daily for 221 days and illness resulting with paralysis or CNS involvement and deaths were recorded and LD_{50} were calculated by the method of Reed and Meunch (14).

To conclude the certainity of death due to dengue and not of adventitious causes, brain smears of mice dying at the highest dilution was checked by DFAT, and from time to time was randomly selected for virus isolation in C6 36 tissue culture, and presence of viral antigen detect by DFAT.

Outbred Swiss mice ICR strain were obtained as special pathogen free, from the Animal Supply Centre of DMR. The mice were shown to be free from haemagglutination inhibition (HI) antibodies to dengue 2 and JE (Jaguar) virus.

Infectivity assay

10 fold dilution of virus was assayed in monolayers of C6 36 cell culture prepared in Lab Tek chamber slides. Adsorption was carried at 34°C for 90 mins. After addition of MEM + Glutamine + NEA and 2% FeCS pH 7.3 it was incubated at 34°C Co_2 incubator for 7 days and viral antigen was detected by DFAT and infectivity expressed as $IFID_{50}$ was calculated by the method of Reed and Meunch (14).

Results

Plaque assy

Results of the plaque assay of the dengue 2 virus recovered from DHF shock and patients have shown, that 5 out of 9 exhibited large plaques, 3 out of 9 intermediate plaque, and 1 out of 9 presented small plaques.

6 out of 9 strains had high PFU/ml and 3 had low PFU/ml Table 52a.

None of the dengue 2 recovered from DHF nonshock patients showed large plaques. 2 out of 9 strains grew as intermediate plaques and majority presented as small plaques. Moreover none of these strains had high PFU/ml, but all of them (9) had low PFU/ml Table 52b.

The plaque assay comparison between strains of the same serotype of dengue 2 from Shock and nonshock patients of DHF showed correlation and association with disease severity, whereby large plaques was only seen from 5 out of 9 shock patients Table 53.

Further comparison of plaque size and the PFU/ml in relation to the type of cell used (C6 36 or Vero) for the test did not present any marked difference.

In the 4 wild strains of dengue 2 recovered from DHF patients with shock heterogenous or mixed population of plaques were seen, however characterization of the size i.e. large, intermediate or small was only done so when 70% or more of a particular plaque size appeared as a homogenous population. Fig. 35 a and 35 b.

An interesting finding was that of the 9 strains of dengue 2 recovered from Shock patients only 2 strains grew plaques which appeared like doughnuts. On the other hand none of the 9 strains recovered from DHF non shock patients presented doughnut plaques. Since this biological appearance was a constant finding even when taken up to 5 TCF passage or whether or not the passage level of cell line used was same or different.

These findings have indicated that large plaque size correlates with disease severity as shown in table 53. Moreover the doughnut appearance of some strains appear to present biological character which denotes variation among strains of dengue 2 recovered from different disease manifestation in DHF.

Effect of temperature on the replication and production of viral antigen.

The prototype dengue 2 strain was found to replicate at lower temperatures of 34°C but was incapable of growth at 39°C and thus 39°C was designated as the non-permissive temperature. Of the 8 strains recovered from DHF Shock patients only 4 was capable of growth at nonpermissive temperature, and ratio of $IFID_{50}$ log 10/ml at 37°C and 39°C was 1, indicating that the strain recovered from shock were temperature resistant t^-, on the other hand 4 of 8 were temperature sensitive t^+ Table 54a None of the strains recovered from patients with nonshock DHF were capable of growth at non-permissive temperature and could be regarded as temperature sensitive ts^+ strains Table 54b A comparison of growth at non-permissive temperature between strains of dengue 2 of two different manifestation clearly shows only 4 of the strains recovered from shock was capable of growth at non-permissive temperature Table 54c.

Adsorption efficiency was measured at the permissive and non-permissive temperature for the dengue 2 virus. Virus adsorption was carried out at 34°C, 37°C permissive and 39°C non-permissive temperature for 90 mins. It was found that adsorption was not markedly affected as shown in Table 55. The results indicate that probably RNA synthesis and production of infectious progeny are inhibited and hampered with at non-permissive temperature for most strain of dengue virus. Replication of 1 prototype, 2 strains each recovered from shock and nonshock DHF patients was studied after absorption and incubating at varying temperature of 34°C, 37°C and 39°C. Growth was detected by the presence of viral antigen in the cytoplasm generally overriding a pole or both poles of the nucleus by DFAT on harvested material of day 3, 5 and 7. The results show that viral replication appears to be suppressed at restrictive or non-permissive temperatures for the prototype and 2 strains recovered from Nonshock patients of DHF, whereas, growth was not much affected in the 2 strains recovered from shock patients.

There is a strong positive correlation between strains dengue 2 recovered from 2 different disease severity and its ability to grow and replicate at permissive and non-permissive temperature Table 56.

Virulence assay in mice

Table 57a and 57b shows the LD 50 log 10/ml of the dengue 2 strains recovered from shock and nonshock patients of DHF when inoculated by the IC, and IP routes. Three patterns were identified (1) strain of high virulence when LD_{50} log 10/ml ratios if IC/IP was lesser than 1. (2) strain of low virulence caused low mortality and IC/IP ratio was greater than 1. (3) Strain of no virulence as infection was not apparent over the 21 days observed for illness or when death occured due to adventitious cause and the IC/IP ratio could not be calculated.

Of the 8 strains of dengue 2 recovered from shock patients 4 fell into the group of high mouse virulence, and 2 were of low mouse virulence and 2 could not express any signs of infection in the mice. Of the 5 strains of dengue 2 recovered from nonshock patients none of them presented high mouse virulence and 1 was of low virulence, and the remaining 4 did not express any signs of infection in mice.

Further when LD50 log 10/ml ratio of prototype to the wild strains was determined, it also was found that the degree of disease response in mice could also be classified as either high or low virulence, and in some did not have any response to infection. The results showed that among the dengue 2 strains recovered from shock, 4 presented characteristics of high virulence and 4 of low virulence Table 58a. On the other hand among the dengue 2 strains recovered from non shock DHF patients, none presented characteristics of high virulence. 2 strains show response of low virulence and 3 strains did not respond to infection Table 58b.

Further comparison and correlation of dengue virus recovered from shock and nonshock DHF patients with mouse virulence when expressed by ratio of the prototype to test strain, it was also found that of the 8 strains recovered from shock patients, 4 expressed high virulence, 2 low virulence and 2 no response to infection. On the other hand neither of the 5 strains recovered from shock patients expressed high virulence features, but 1 presented low virulence and 4 did not show any response to infection Table 59.

Table 60 summarizes the findings of each of the biological test processed to aid in correlation with disease severity. The summary depicts clearly that in 4 out of 7 strains recovered from DHF shock patients presented large plaques, were temperature resistant and showed high virulence in mice. However the remaining 3 strains did not show the consistent pattern as mentioned above, but presented attributes generally associated with attenuation, and like wise for all the 4 strains of dengue 2 recovered from non-shock DHF patients presented biological attributes general associated with attenuation or strains capable of causing milder infections.

Discussion

The biological markers have been employed extensively in differentiating strains of the same virus strains. Mixed population of large and small plaques have been frequently found in wild dengue virus type 2 isolates from human and mosquitoe in both Asia and the Caribbean region. Previous studies with dengue 2 demonstrated that large and small plaque clones could be readily derived from silates and small plaque clone had a markedly reduced virulence of suckling mice. Further naturally occuring and induced temperature sensitive mutants of other type of viruses have been associated with lower causal virulence (17). Preliminary study on biological variation of dengue 2 ciruses recovered from 7 patients with severe illness (shock) and 5 from patients with less severe nonshock infection in DHF have shown that plaque size, replication at non-permissive temperature of 39°C and high mouse virulence correlated with disease severity in 4 only, as seen in the summarized table of the biological marker systems, Table 61 and 62. A definite conclusion for applicability of positive findings of virulence was not seen in any of the 4 strains of dengue 2 recovered from nonshock DHF patients.

In relation to the appearance of doughnut plaque only from 2 strains recovered from shock patients and none from nonshock patients may lead us to consider in the light of the occurence of CPE in pathogenic viruses, especially as dengue viruses by rule do not produce CPE in most arthropod or mammalian cell cultures. The

possible explanations may be that infected cells when stained by the immuno-enzymatic method when examined under the microscope showed that the center part of the doughnut presented a large amount of dengue antigen which had and quickly replicated resulting with degeneration of cells and pyknosis occured after further peripheral spread of the virus thus leaving a clear rim outside the heavily stained plaque area. These findings strongly indicate the possibility of differentiation of strain variation in dengue 2 by biological markers such as plaque size, temperature markers and mouse virulence.

More over as the biological attributes of large plaque growth at nonpermissive temperature of 39°C and high mouse virulence correlated with disease severity in DHF may lead on to hypothesize that the biological variation is of genetic in nature as already shown by some workers for strains of dengue 2 from different geographic regions (6) and thus provide insight to the pathogenesis of the severe from of DHF/DSS.

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Table 52a Plaque assay of dengue 2 virus recovered from shock patients of DHF by the Immunoenzymatic method.

Sr. No.	Serum No.	C6 36 Cells		Vero cells	
		PFU/mℓ	Size in mm	PFU/ml	Size in mm
1	10048	2.3×10^7	0.8 -	3.3×10^6	- 1.2
2	10049	2.9×10^7	(1) 1.0 - 1.2 (2) 0.2 - 0.4	7.1×10^7	(1) 0.8 - 1.2[D] (2) 0.4 - 0.5
3	14935	7.4×10^7	(1) 1.6 - 1.8 (2) 0.6 - 0.8	1.3×10^6	(1) 1.8 - 2.0[D] (2) 0.4 - 0.6
4	11021 Enceph.	4×10^7	(1) 0.8 - 1 (2) 0.2 - 0.4	2×10^6	(1) 1.2 - 1.4 (2) 0.4 - 0.6
5	13116	1.2×10^6	0.4 - 0.6	2.1×10^4	0.6 - 0.8
6	10210	4×10^7	0.4 - 0.5	2×10^6	0.6 - 0.8
7	10665	9.3×10^5	0.6 - 0.8	1.3×10^5	0.6 - 0.8
8	13888	2×10^5	0.5 - 0.6	2×10^4	0.5 - 0.6
9	17969 autopsy liver	2×10^7	(1) 1 - 1.2 (2) 0.6 - 0.8 (3) 0.2 - 0.4	2×10^6	(1) 1.0 - 1.2 (2) 0.6 - 0.8 (3) 0.2 - 0.4

Enceph. - isolate from a patient with encephalitis.

D - doughnut appearance.

Distribution of plaque and association with disease severity

Large plaque > .8 mm _____ 5

Intermediate plaque .6 - .8 _____ 3

Small plaque < .6 mm _____ 1

n = 9

Distribution of PFU infectious unit and association with disease severity

High PFU titer > 4.7×10^6 _____ 6

Low PFU titer < 4.7×10^6 _____ 3

Table 52b Plaque assay of dengue 2 virus recovered from nonshock patients of DHF by the Immunoenzymatic method.

Sr. No.	Serum No.	C6 36		Vero Cells	
		PFU/ml	Size in mm	PFU/ml	Size in mm.
1	13255	2.5×10^6	.6 - .8	1.3×10^5	.6 - .8
2	13319	1×10^6	.6 - .8	2×10^4	.6 - .8
3	10478	2.3×10^5	.3 - .4	2×10^5	.6 - .8
4	10208	2.3×10^5	.3 - .4	2×10	.5 - .6
5	18084	1×10^5	.2 - .3	1×10^3	.6 - .8
6	17874	7.5×10^2	.4 - .6	2×10^2	.4 - .6
7	18660	5×10^2	.4 - .6	5×10^2	.4 - .6
8	20733	2.8×10^2	.1 - .3	1×10^2	.4 - .5
9	18066	5×10^3	.4 - .6	2×10^2	.6 - .8

Distribution of plaque and association with disease severity.

Large plaque	> .8 mm	0
Intermediate plaque	.6-.8 mm	2
Small plaque	< .6 mm	7

n = 9

Distribution of PFU infectious unit and association with disease severity

High titer	> 4.6×10^6	0
Low titer	< 4.7×10^6	9

n = 9

Figure 35a The distribution of plaque size of dengue 2 recovered from shock patients of D.H.F.

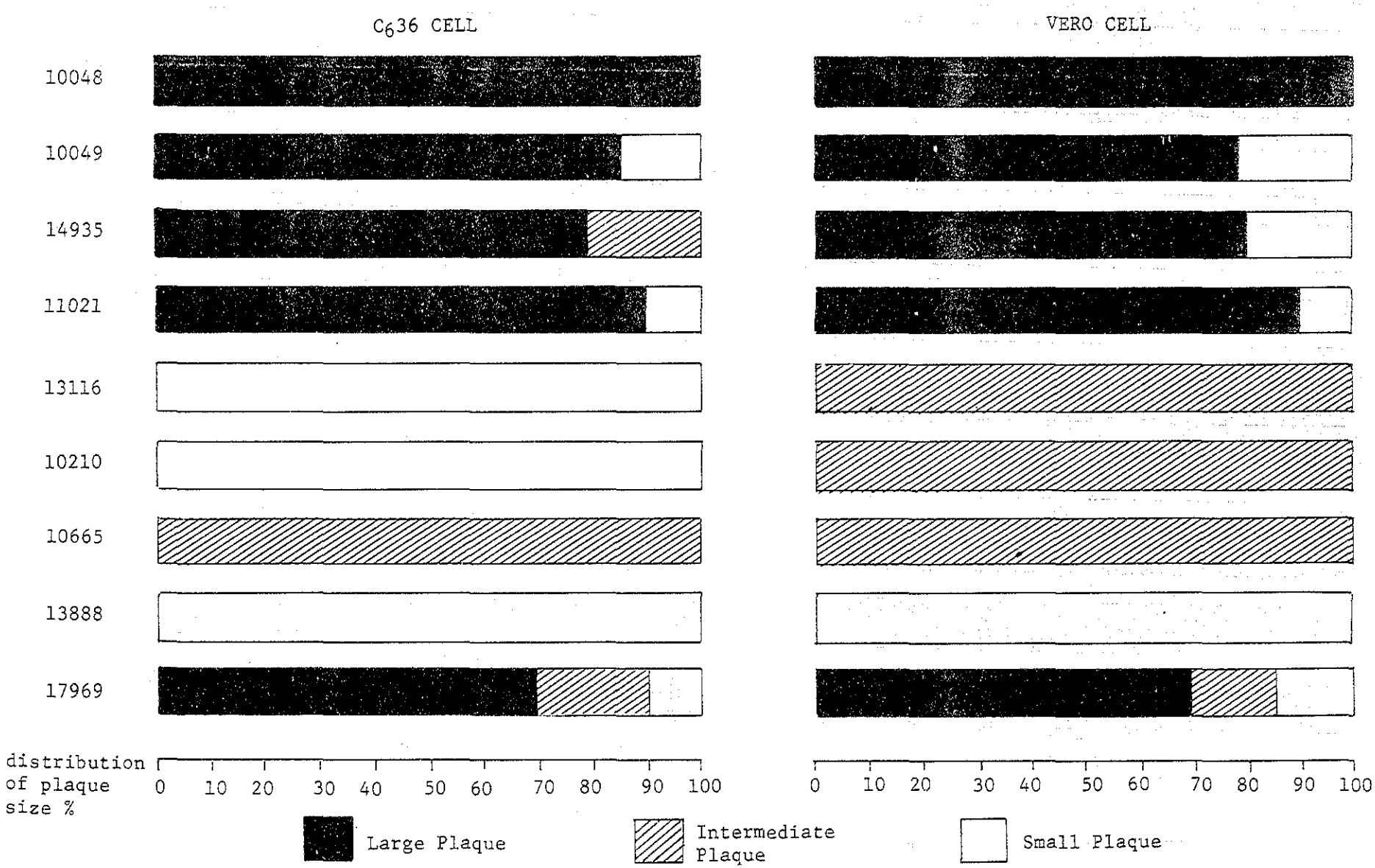


Figure 35b The distribution of plaque size of dengue 2 recovered from non-shock patients of D.H.F.

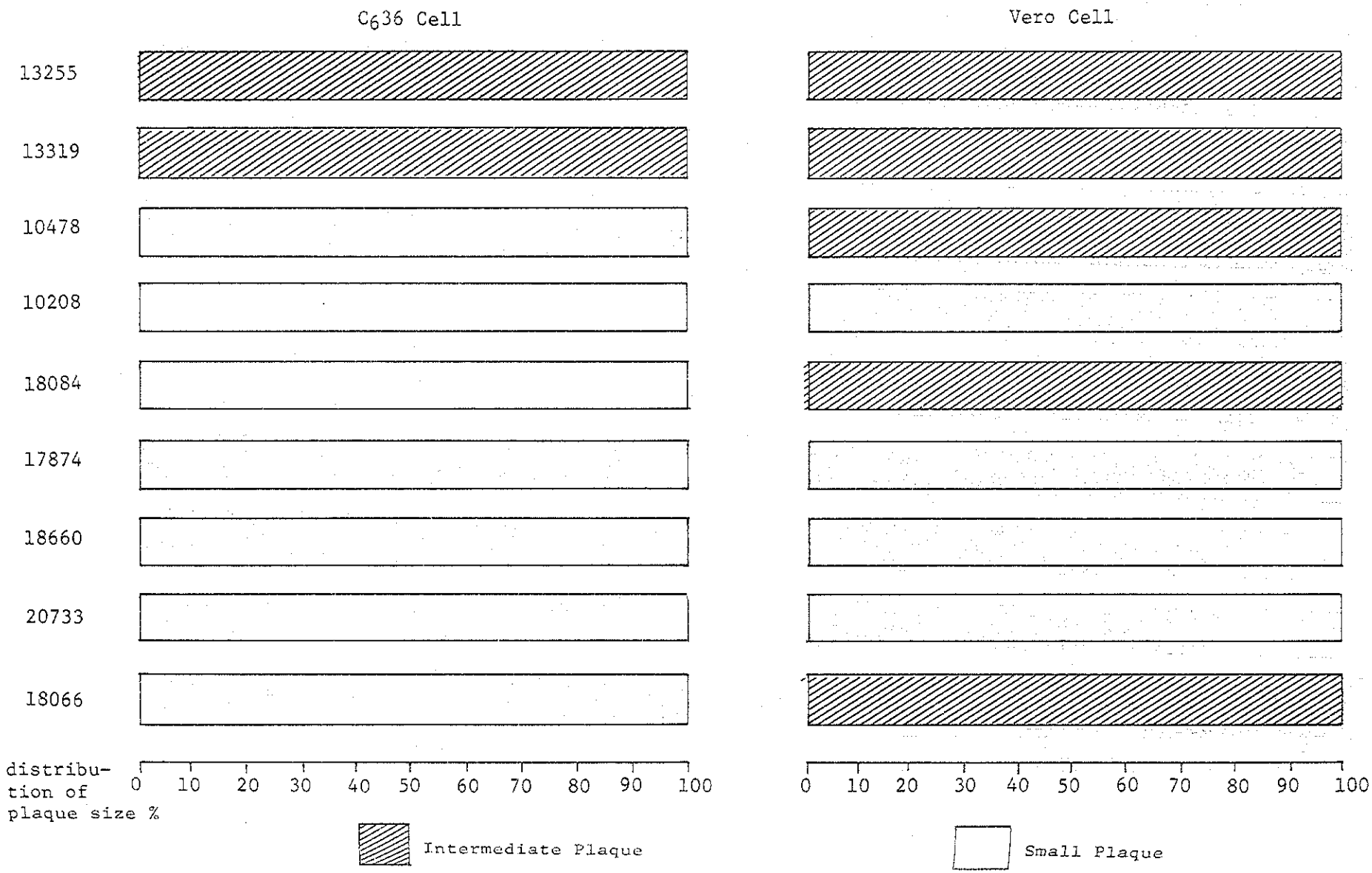


Table 53 Comparison of plaque assay between Shock/nonshock patients of DHF and correlate plaque size with disease severity

Disease Severity	Plaque size						Plaque forming units/ml.			
	C6 36			Vero			C6 36		Vero	
	L	I	S	L	I	S	High	Low	High	Low
S n = 9	5	1	3	5	3	1	6	3	6	6
NS n = 9	0	2	7	0	-	9	-	9	0	9

S = Shock, NS - Nonshock, L = large plaque, I = intermediate plaque, S = small plaque

Table 54a The effect of temperature on the relication and production of viral antigen in dengue 2 virus recovered from shock patients of DHF.

Sr. No.	Serum No.	Original Title	Titer at 37°C	Titer at 39°C	Ratio of 37°C/39°C	t Marker
1	10048	$10^{7.5}$	$10^{6.5}$	$< 10^{1}$	> 5.5	NC
2	10049	$10^{7.5}$	$10^{7.7}$	$10^{6.7}$	0	t^-
3	14935	$10^{6.5}$	$10^{6.5}$	$10^{6.5}$	0	t^-
4	11021en	$10^{6.5}$	$10^{6.5}$	$10^{5.5}$	0	t^-
5	13116	$10^{5.5}$	$10^{5.5}$	$< 10^{1}$	> 4.5	NC
6	10210	$10^{6.7}$	$10^{5.5}$	$< 10^{1}$	> 4.5	NC
7	10665	$10^{5.5}$	$10^{5.5}$	$< 10^{1}$	> 4.5	NC
8	17969 autopsy liver	$10^{7.5}$	$10^{6.7}$	$10^{6.7}$	0	t^-

IFID 50 log 10/ml

Ratio of IFID 50 log 10/ml / Ratio of IFID 50 log 10/ml 37°C / 39°C

Table 54b The effect of temperature on the replication and production of viral antigen in dengue 2 virus recovered from nonshock patients of IHF

Sr. No.	Serum No.	Original Titer	Titer at 37°C	Titer at 39°C	Ratio of 37°C/39°C	t makrer
1	13255	$10^{5.2}$	$10^{5.2}$	$< 10^{1}$	> 4.2	t^{+}
2	13319	$10^{5.2}$	$10^{4.7}$	$< 10^{1}$	> 3.7	t^{+}
3	10478	$10^{3.2}$	$10^{3.2}$	$< 10^{1}$	> 2.2	t^{+}
4	10208	$10^{3.2}$	$10^{3.2}$	$< 10^{1}$	> 2.2	t^{+}
5	18084	$10^{3.2}$	$10^{4.2}$	$< 10^{1}$	> 3.2	t^{+}
6	17874	$10^{2.2}$	$10^{2.2}$	$< 10^{1}$	> 1.2	t^{+}
7	18066	$10^{4.2}$	$10^{4.2}$	$< 10^{1}$	> 3.2	t^{+}

IFID 50 log 10/ml

Table 54c Comparison of growth at nonpermissive temperature between strains of dengue 2 recovered from shock and nonshock patients

Disease Severity	Growth at permissive temp. 37°C	Growth at nonpermissive temperature 39°C
Shock n = 8	8	4
Nonshock n = 7	7	0

Table 55 Effect of temperature on adsorption

Virus	% of inoculum adsorbed at		Ability to replicate	
	34°C	39°C	34°C	39°C
Prototype n = 5	50	40	Yes	No
Shock n = 5	70	60	Yes	Yes
n = 5	65	45	Yes	No
Nonshock n = 5	40	42	Yes	No

Table 56 Comparison of replication of dengue virus recovered from Shock and Nonshock cases of DHF when cultivated at 37°C and 39°C

Disease severity or prototype strain.		Growth 37°C Permissive temp.		Growth 39°C Non permissive temp.
Prototype	n = 1		+	-
Shock	n = 8	4	+	+
	N	4	+	-
Nonshock	n = 7	6	+	-
		1	+	-

Table 57a Mouse virulence assay of dengue 2 virus recovered from Shock patients of DHF

Sr. No.	Serum No.	LD50 log 10/ml IC	LD50 log 10/ml IP	Ratio value IC/IP in Log	Degree of virulence
1	10048	$10^{1.5}$	10^{1}	0.5	low
2	10049	$10^{2.8}$	$10^{2.9}$	-0.1	high
3	14935	$10^{2.4}$	10^{3}	-0.6	high
4	11021En.	10^{4}	$10^{4.4}$	-0.4	high
5	13116	$10^{5.7}$	$10^{4.4}$	1.3	low
6	10210	10^{1}	$< 10^{1}$	> 0	NR
7	13888	10^{1}	$< 10^{1}$	> 0	NR
8	17969 autopsy liver	$10^{3.4}$	$10^{4.8}$	-1.5	high

When ratio of LD 50 Log 10/ml of IC/IP is < 1 = high virulence - 4

> 1 = low " - 2

No response to infection - 1

n = 8

Table 57b Mouse virulence assay of dengue 2 virus recovered from nonshock patients of DHF.

Sr. No.	Serum No.	LD50 log 10/ml IC	LD50 log 10/ml IP	Ratio value IC/IP in Log	Degree of virulence
1	10478	$< 10^1$	$< 10^1$	NC	NR
2	10208	$< 10^1$	$< 10^1$	NC	NR
3	18084	$10^{2.3}$	$< 10^1$	> 1.3	NR
4	17874	10	10^1	0.7	Low
5	20733	$< 10^1$	$< 10^1$	NC	NR

When ratio of LD50 log 10/ml of IC/IP ratio is

< 1 = high virulence - 0

> 1 = low virulence - 1

No response to infection - 4

n = 5

Table 58a Mouse virulence assay by IC/IP route infection of dengue 2 virus recovered from DHF Shock patients.

Sr. No.	Serum No.	IC/IP LD 50 log 10/ml	Ratio LD50 log 10/ml Prototype/Shock strain.	Degree of virulence
1	10048	$10^{2.3}$	2.3	low virulence
2	10049	10^6	.9	high virulence
3	14935	$10^{3.8}$	.79	high virulence
4	11021	$10^{4.4}$	.9	high virulence
5	13116En.	10^5	1.8	low virulence
6	10210	$10^{2.5}$	2.1	low virulence
7	13888	$10^{2.9}$	1.9	low virulence
8	17909 autopsy liver.	$10^{5.8}$	.9	high virulence

Wh n LD50 log 10/ml of Prototype/Shock Strain = n = 8

< 1 = high virulence - 4 strain

> 1 = low virulence - 4 strain

Table 58b Mouse virulence assay by IC/IP route infection of dengue 2 viruses recovered from DHF Non-shock patients.

Sr. No.	Serum No.	IC/IP LD_{50} log 10/ml	Ratio of LD50 log 10/ml Prototype/Non shock strain	Degree of virulence
1	10478	No response	Not calculatable	No response.
2	10208	No response	Not calculatable	No response.
3	18084	10^2	2.7	Low virulence.
4	17874	$10^{2.4}$	2.3	Lov virulence.
5	20733	No response	Not calculatable	No response.

When LD50 1g 10/ml of Prototype/Non shock strain = n = 5

< 1 = high virulence = 0

> 1 = low virulence = 2

No response to infection = 3

Table 59 Comparison and correlation of dengue virus recovered from shock/nonshock DHF patients with mouse virulence when ratio of prototype/test strain is expressed.

Disease severity in DHF	High mouse virulence	Low mouse virulence	No response to infection
Shock n=8	4	2	2
Nonshock n=5	0	1	4

Table 60 Summary of the biological markers of dengue strains recovered from DHF patients.

Sr. No.	Serum No.	Grade of illness	Plague marker	t Marker	Mouse virulence marker	Correlation with disease severity in DHF
1	10048	S	L	t^+	Lmv	low virulence
2	10049	S	L + S	t^-	Hmv	high virulence
3	14935	S	L + S	t^-	Hmv	high virulence
4	11021^{EN}	S	L + S	t^-	Hmv	high virulence
5	13116	S	S	t^+	Lmv	low virulence
6	10210	S	S	t^+	NR	No reponse to infection
7	17969 autopsy liver	S	L	t^-	Hmv	high virulence
1	10478	NS	S	t^+	NR	low virulence
2	10208	NS	S	t^+	NR	low virulence
3	18084	NS	S	t^+	NR	low virulence
4	17874	NS	S	t^+	Lmv	low virulence

S = Shock n = 7

NS = Nonshock n = 4

L = Large plaque

S = Small plaque

t^- = Capable to growth at 39°C

t^+ = Not capable of growth at 39°C

Hmv = High mouse virulence

Lmv = Low mouse virulence

NR = No response to infection

Table 61 Summary of the biological markers of dengue virus strains recovered from DHF shock patients

Sr.	Serum	Grade	Response	Plaque marker			t Marker	MV	IFID 50
				L	I	S			
1.	10048	S	2°	+	-	-	t^+	LMV	h
2.	10049	S	1	+85%	-	+15%	t^-	hMV	h
3.	14935	S	2°	+80%	-	+20%	t^-	hMV	h
4.	11021	S	2°	+90%	-	+10%	t^-	hMV	h
5.	13116	S		-	-	+	t^+	hMV	h
6.	10210	S	2°	-	-	+	t^+	NR	h
7.	10665	S		-	+	-	t^+	NT	h
8.	13888	S		-	-	+	NT	NR	l
9.	17969 PM liver	S		+	-	-	t^-	hMV	h

L = Large plaque
I = Intermediate plaque
S = Small plaque
t^- = Growth at non permissive temperature (39°C)
t^+ = No growth at non permissive temperature (39°C)
MV = Mouse virulence
hMV = High mouse virulence
LMV = Low mouse virulence
NR = No response to infection
h = High immunofluorescence infecting dose 50
l = Low " " "
NT = Not tested

Table 62 Summary of the biological markers of dengue virus strains recovered from DHF nonshock patients

Sr.	Serum	Grade	Response	Plaque marker			t Marker	MV Marker	IFID 50
				L	I	S			
1.	13255	NS	2°	-	+	-	t+	NT	L
2.	13319	NS	2°	-	+	-	t+	NT	h
3.	10478	NS	1°	-	-	+	t+	NR	L
4.	10208	NS	2°	-	-	+	t+	NR	L
5.	18084			-	-	+	t+	NR	h
6.	17874			-	-	+	t+	LMV	h
7.	18660			-	-	+	t+	NT	h
8.	20733			-	-	+	NT	NR	L
9.	18066			-	-	+	NT	NT	NT

l = Large plaque
I = Intermediate plaque
S = Small plaque
t- = Growth at non permissive temperature (39°C)
t+ = No growth " " "
MV = Mouse virulence
hMV = High nouse virulence
LMV = Low mouse virulence
NR = No respense to infection
h = High immunofluorescence infecting dose 50
l = Low " " "
NT = Not tested

3-6 The Studies of Dengue Virus Type 2 in Toxorhynchites Cell Line(TRA171)

-------- Replication and morphological observation --------

In the field of arbovirus research, cultured mosquito cells were widely used for the propagation of dengue and other arboviruses, because of its superior results compared to suckling mouse and cultured mammalian cells inoculation. We had already reported the growth and morphological characters of Japanese encephalitis, dengue and yellow fever viruses using *Culex pipiens* var *molestus* cell line.

In the present report, using non-hematophagous *Toxorhynchites amboinensis* mosquito cell line (TRA 171) established by Kuno which at present widely used in arbovirus research, we have studied the growth and morphological characters of dengue virus wild strains isolated from the sera of shock and nonshock dengue hemorrhagic fever (DHF) patients from Rangoon Children's Hospital, Rangoon, Burma. The wild strains were identified as dengue virus type 2 (D-2). The results of these wild strains were compared to those of D-2 prototype (Tr 1751) which have been adapted and passaged many times in suckling mouse brain. As there are very few reports on *in vitro* and morphological studies of wild strains from shock and nonshock DHF patient, it is expected that present study will help to understand the pathogenesis of DHF.

Materials and Methods

viruses: Dengue wild type strains (BR 006 and BR 116) were isolated from DHF patients (shock and nonshock) in *Tx. splendens* mosquitoes using mosquito inoculation technique and identified as D-2 at the Department of Medical Research, Rangoon, Burma. Later these wild strains were passaged in *Tx. splendens* mosquitoes and further in *Aedes albopictus* cloned C6/36 cell line both for 2-3 times. D-2 prototype (Tr 1751) was prepared in suckling mouse brain.

Cultured cells: Cultured *Tx. amboinensis* cell line (TRA 171) was obtained from Department of Microbiology, Faculty of Medicine, Kobe University. Cells were subcultured at 30°C with MM/VP medium supplimented with 10% fetal bovine serum. Vero and C6/36 cells were used for plaque assay and for other studies.

Virus inoculation and titration: D-2 prototype virus was prepared as 10% mouse brain emulsion and the supernatant was diluted into 1:10 dilution. Five days after incubation, C6/36 cells, inoculated with D-2 wild type viruses (BR 006 and BR 116) were frozen and thawed for 3 times and diluted into 1:10 dilution. Each strains were then inoculated to 2-3 days old monolayered TRA 171 cell cultures. After 90 min adsorption, maintenance medium was added and incubated at 30°C. An aliquot of cultured medium was harvested every day and assayed for the virus titer by the plaque assay method using 24 multiwell plate (ϕ=16 mm). Serial ten fold diluted materials were inoculated onto Vero cells monolayers, cultured in the plates. After adsorption, overlay medium containing 1.2% methylcellulose was added and incubated at 37°C for 6-7 days. After incubation, the overlay medium was removed and stained with Giemsa for counting the plaque numbers and their morphological characters. At the same time the other wells were stained by the enzyme focus forming assay (EFFA) method using peroxidase labelled antibody.

Electron microscopy: For morphological studies, infected TRA 171 cells were harvested daily and ultrathin sections were observed for both conventional electron microscope (EM) and immuno-electron microscope (IEM). Hitachi model H-500 electron microscope was used in the study.

Light microscopy: For light microscopic observation, TRA 171 cells were prepared on coverslip in Leighton's tubes and infected. Coverslips were then stained with Giemsa and the other for indirect fluorescent antibody (IFA) staining.

Results and Discussion

Virus replication in TRA 171 cells: Virus replication of the wild strains were detected at 2 days after infection. The virus titer rose until 5 days and showed 10^7PPU/ml for BR 006 and 10^{5-6} PPU/ml for BR 116. These titers had been kept until 10 days after infection. As for the titer of Tr 1751, the virus titer rose until 7 days after infection, with 10^8PFU/ml. On the other hand, virus replication of the wild strains in the Vero cells (control) showed same as those of Tr 1751 strain. All the virus titers were 10^{6-7}PFU /ml at 4-5 days after infection (Fig. 36).

Plaque morphology: Wild and prototype strains of viruses replicated in the TRA 171 cells showed different plaque morphology on the Vero cells (Fig. 37, 38). Both BR 006 and BR 116 showed various plaque sizes. Their sizes were 0.6 to 1.5 mm in diameter and some plaques showed doughnut like shape. The reason is healthy cells were remained at the center of the plaque just like an island in a lake. According to the sizes of plaques two groups were divided (Table 63). One, 1.2-1.5 mm in diameter (large plaque) and the other, 0.6-0.8 mm (small plaque). The number of large plaque to those of small plaque was higher in BR 006 strain and same in BR 116 strain. On the other hand, Tr 1751 replicated in the TRA 171 cells showed uniform plaques of 1.2-1.5 mm in diameter, whereas no plaques were observed in TRA 171 control cells.

As we want to understand the characters of different plaque sizes in wild strains, virus clones (L_1 and S_1) were isolated from large and small plaques of BR 116 strain, respectively. These virus clone isolates were replicated in the Vero cells and their infectious titers were assayed at various incubation temperature for 4 days (Fig. 39). The titer of L_1 was $4X10^5$ and $1.4X10^5$PFU/ml at 39°C respectively. The titer of S_1 was $1.6X10^5$ and $3X10^4$PFU/ml at 37°C and 39°C. No virus replication was seen in the Vero cells at 30°C. Both clones were shown to be D-2 by FA method using D-2 monoclonal antibody produced by lymphocyte hybridoma cell line (93H5).

Light microscope observation: Cells (TRA 171) infected with wild and prototype viruses showed same results by light microscope observation. Cells were partially aggregated and partial CPE was observed at 48 hr after infection. Remaining cells were healthy and cell division and replication was also observed.

By fluorescence microscope observation, aggregated fluorescent dots appeared first in the perinuclear cytoplasm in the early stage of infected cells. Later these positive areas spread throughout the whole cytoplasm.

Electron microscope observation: TRA 171 cells, infected with D-2 prototype and wild strains (BR 006 and BR 116) were harvested and prepared for ultrastractural observation. Dengue virus particles of 40 nm in diameter and enveloped were observed (Fig. 40). These virus particled were found within the cytoplasmic vacuoles.

The small tubular vacuoles with virus particles were also found in the cytoplasm. This structure was the typical characteristics of the arbovirus infected cells. The present findings were also similar to those of previous studies of D-2 in other cell lines.

IEM haves also shown that dengue virus particles in all virus strains (prototype and wild strains) react with the antisera raised against D-2 prototype.

In the present study we had also unexpectedly found 4 types of virus like particles in nucleus, cytoplasm and large vacuoles. In the nucleus, electron dense particles about 10 nm in diameter were observed by lattice like arrangement (Fig. 41). They arranged massively in one part and linearly in another part and sometime they are in very regular crystalline arrangements. In the cytoplasm another crystalline like structures were found. These particles were electron dense particles with 15-20 nm in diameter. Enveloped virus like particles, about 25 nm in diameter, were found in the cytoplasmic vacuoles (Fig. 42). Finally we had also found larger particles of 100 nm in diameter in the nucleus and cytoplasm. At present the nature and the character of these virus like particles present in the TRA 171 was unknown. Further morphological studies of these particles will be reported later.

Conclusion

In the present study we had found that D-2 prototype strain replicate well in the TRA 171 cell line, and also in D-2 wild type strains. At present there is no report on the replication of the wild type strains in TRA 171 cells. As wild strains replicate well in the TRA 171, there is no doubt that TRA 171 cell line is useful for the isolation and identification of arboviruses.

The results of our previous report in *Toxorhynchites* splendence, *in vivo*, by D-2 prototype virus is comparable to those of the present results, *in vitro*.

But on the other hand, as this TRA 171 cell line contains 4 types of unknown virus like particles, ultrastructurally, it is also assumed that there is inferiority in this cell line due to the contamination of these particles. The further cloning of the cell free from these contaminated particles is required for further usage of TRA 171 cell line.

3.7 Training Given

Post-graduate Diploma in Bacteriology students are provided with theoretical and practical training in relation to basic and recent virological techniques. Some of the students did their dissertation at the division under the supervision of the division's staff. Students of the Rangoon University are allowed to do the thesis for their Masters Degrees of Science (Zoology) at the division under the supervision of the division's staff. The following dissertations and thesis were completed.

(1) Effect of temperature on infection of dengue 2 viruses recovered from different clinical manifestation in Dengue/DHF.

(2) Relationship between haemagglutinating units and haemagglutinating inhibition titres of arboviruses.

(3) Effect of temperature on growth and replication of dengue viruses.

3.8 Training Received

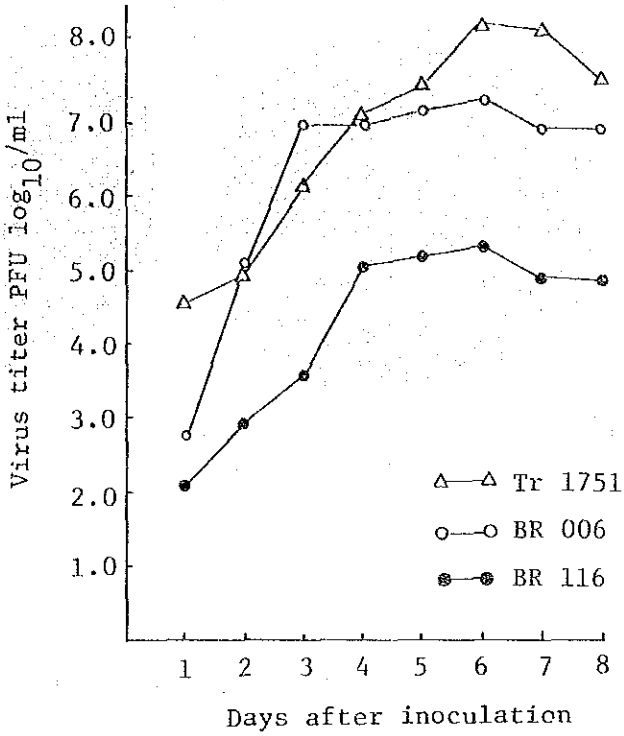
One scientist (Senior Research Officer) was trained for the research in Entero-Viruses and one scientist (Research Officer) is now undertraining for the Ultrastructural Studies of arboviruses in Japan. Moreover, Japanese Experts visited the Virology Research Division for varying periods and collaborated with scientists and gave training for salient techniques.

3.9 Academic Activities Relevant to The Project

Virology, Pathology and Biochemistry Research Divisions conducted a workshop on "Intercountry Workshop on Electron microscopy and Immune Electron-microscopy" in November 1983. The workshop is to provide the participants EM and IEM techniques for the laboratory diagnosis of viruses causing diarrhoea. See Annex attached to this Report.

Fig. 36 Growth curve of D-2 virus

(Toxorhynchites amboinensis cell line TRA 171)



A. Ohyama, T. Ito, E. Tanimura, N. Yamamoto (KMU)
Mi Mi Khin, Thet Win (DMR)

Fig. 37 BR 006 strain plaques after 7 days infection on Vero cell monolayer with 1.2% MC overlay medium, Giemsa staining, plate of 25 mm in diameter

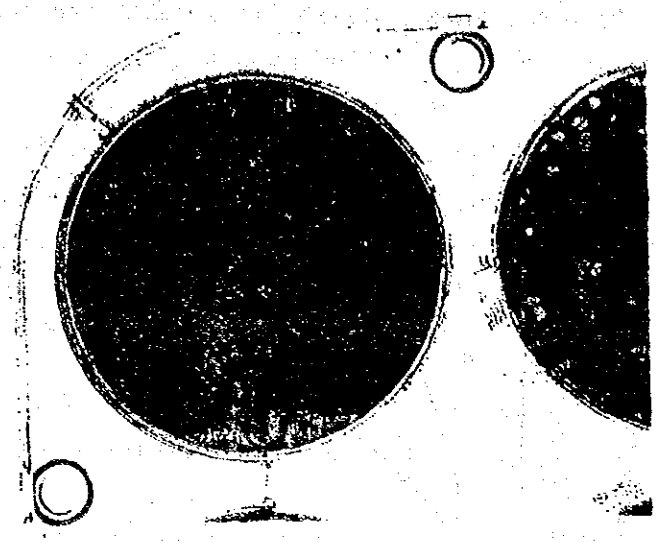


Fig. 38 BR 116 strain plaques after 7 days infection on Vero cell monolayer with 1.8% MC overlay medium, Giemsa staining, plate of 52 mm in diameter

Table 63 The distribution of plaque size of D-2 on vero cells

Plaque size(mm)	Distribution of plaque size (%)		
	Tr 1751	BR 006	BR 116
1.4-1.8	90	19	10
0.8-1.2	10	50	29
0.4-0.6	0	31	61

Incubation at 37°C for 7 days with 1.2% methylcellulose

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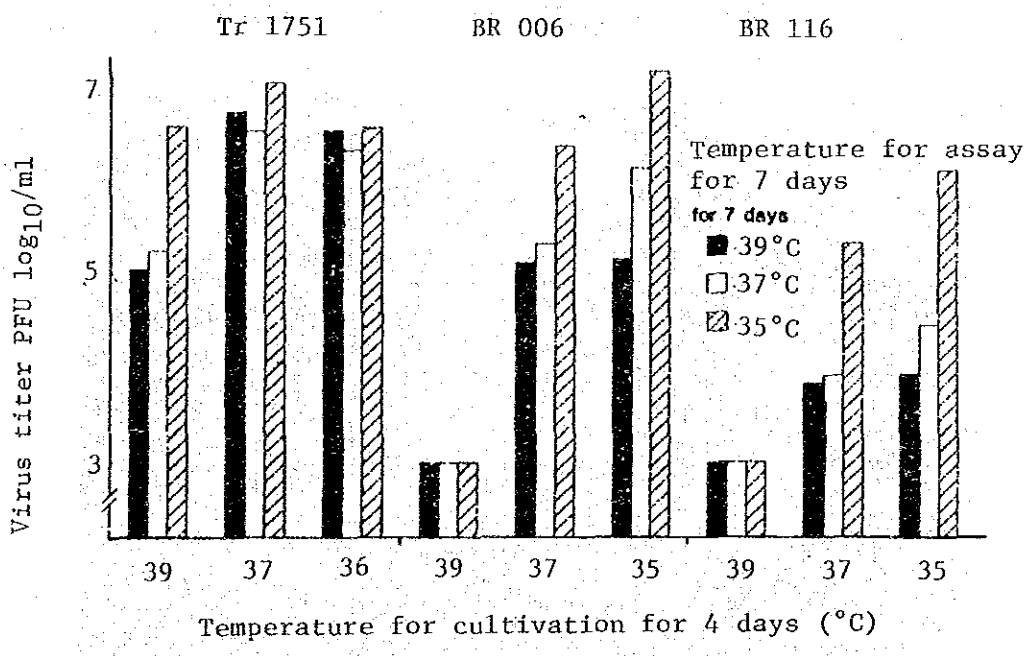
Mi Mi Khin, Thet Win (DMR)

Fig. 39

Temperature sensitivity of D-2 for the plaque formation



A. Ohyama, T. Ito, E. Tanimura, N. Yamamoto (KMU)
Mi Mi Khin, Thot Win (DMR)

Fig. 40 TRA 171 cell, infected with BR 116.
DV; Virus particles, N; Nucleus,
rER; Rough endoplasmic reticulum
x 40,000.

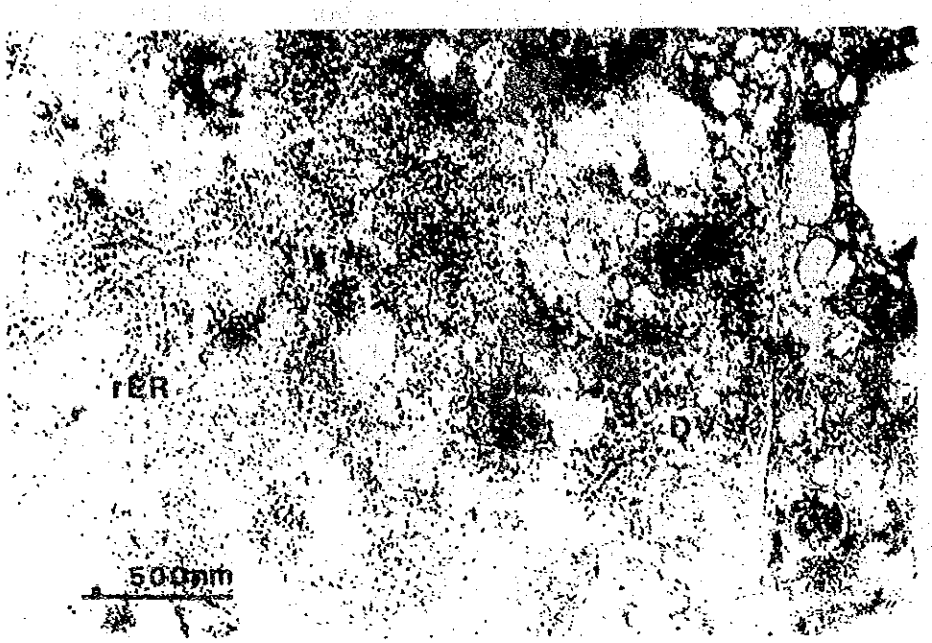
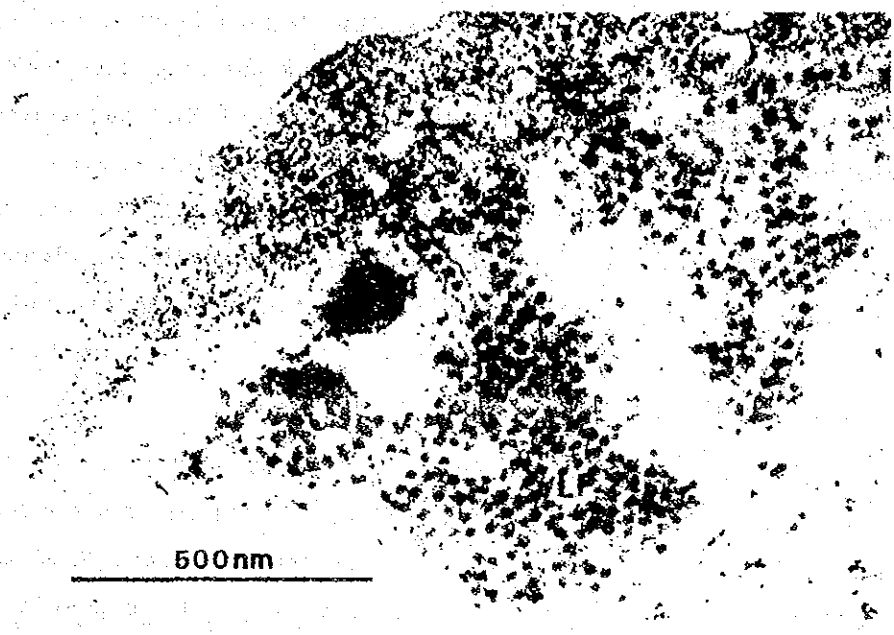


Fig. 41 Uninfected TRA 171 cell.
VLPs; Virus like particles. x 40,000

Fig. 42 Uninfected TRA 171 cell. x80,000



4. Research and Development in Immunology

4.1 Aims of Research and Development

The aim was to develop experties in advanced immunological techniques which would help the DMR to undertake more productive research in the understanding, diagnosis and treatment of the major health problems in Burma, especially in the area of bacterial enteric and arboviral diseases.

Hybridoma technology was identified as one such advanced technology which would be worthwhile of development under the project.

4.2 Development and use of Enzyme Linked Immunoabsorbent Assays (ELISA)

ELISA was developed and used in DMR (and Burma) for the first time in the Immunology Division. A workshop on ELISA was conducted by the immunology Research Division in collaboration with other Divisions of DMR, for scientists from DMR as well as from other institutions in Rangoon. ELISA is now being used extensively by many Divisions of DMR.

4.3 Development of 'Hybridoma Technology'

With the help of Japanese Expert, we attempted fusion of NSI cells with BALB/c spleen cells primed with SRBC. The resultant hybrids were screened for antibody production by simple agglutination test. Positive wells were cloned by limiting dilution technique. Positive clones were expanded and maintained in culture medium and some in frozen state. Monoclonal antibody produced was tested for specificity against a battery of red cells of rabbit, rat, guinea pig and goat. The immunoglobulin class of the antibody belongs to IgG_3. These hybrids been maintained in culture over 6 months and still secreting antibody. Myeloma cell line (NSI) has been maintained in our laboratory since introduction by Prof. Morikawa.

4.4 Research Work

See section on other DMR Research activities which interact all the DMR/JICA Project.

4.5 Training Received

The Senior Research Officer had attended a Workshop on Hybridoma Technology at the Chiba University in Japan in 1981, and the Head of Division attended a WHO Workshop on Hybridoma in Singapore in 1981.

Japanese Expert visited the Division for 1 month in 1983, and helped procure supplies and equipment and initiated hybridoma work.

One senior technician will be trained for 1 year in Japan in general immunological technology and hybridoma techniques.

4.6 Training given

Conducted a workshop on Enzyme-linked immunoabsorbent assay, for scientists of DMR and other institutions.

4.7 Other activities

Gave many lectures and seminars - see annex

4.8 Papers published and read

See - Annexes

5. Research and Development in Pathology

5.1 Aims of Research and Development Programme

The general aim of development of pathology is:-

Firstly to develop the capability to study defects in hemostasis, clotting and alterations in the microcirculation in important diseases prevalent in Burma in which such defects are considered to be of significance (eg. dengue haemorrhagic fever, malaria and snake bite)

Secondly, to develop electron microscopic facilities and utilize then for ultrastructural investigation in the study of major disease problems, especially dengue haemorrhagic fever, severe malaria, snake envenomation and hepatitis.

5.2 Development of Electron Microscopy

A new electron microscope laboratory was established in the pathology research division and a new transmission electron microscope was installed in 1982. This was in addition to the old electron microscope which already existed in another electron microscope laboratory, under the pathology research division.

The head of division received training in electron microscopy in Japan, and short small group training courses were also held at department of medical research by visiting Japanese experts, so as to initiate the use of electron microscope by scientists in other divisions of department of medical research.

5.3 Development of Facilities for Clotting Studies

Basic clotting tests, as well as special tests for assay of nest of the clotting factors were set up. Necessary equipment and supplies were provided by JICA as well as by DMR. The head of division received training in research related to clotting defects in Japan.

Clotting Tests established so far are as follows

Activated partial thromboplastin time
Thrombin time
Fibrinogen estimation
Factor V, VII, VIII, and X assays
Antithrombin III

a_2 antiplasmin

plasminogen

Fibrin degradation products

5.4 Services Provided to Other Research Divisions

(1) Diarrhoea research project: Electron microscopic identification of rota virus particles in stool samples.

(2) Electron microscopic examination of other material sent by research divisions.

5.5 Training given

(1) Technicians from clinical research units on snake bite and malaria were trained for some of the clotting tests.

(2) Supervised one M.Sc thesis titled "Effect of eclipta alba on acute alcoholic liver disease."

5.6 Training received

(1) The head of division received training in pathology research with special emphasis on clotting and electron microscopy, for one year in Japan (1982-83)

(2) One research Officer is being trained in Japan in experimental pathology (1983-85)

(3) Japanese experts visited the division for varying periods and collaborated with scientists and gave training in pathological techniques to technicians

5.7 Research Findings

See section on other DMR research activities interacting with DMR/JICA Project.

5.8 Other Activities

(1) Jointly conducted together with virology research division the WHO/DMR inter-country workshop on electron microscopy and immune electronmicroscopy.

(2) Held many lectures and seminars.

6. Development of Laboratory Animal Technology and Facilities

6.1 An essential support facility for effective research in Bacterial enteric Diseases and arboviral Diseases is the development of laboratory animal Technology and facilities and this was vigorously implemented.

New stock of laboratory animals were received from JICA. The Japanese expert who arrived near the end of 1980 and the Burmese counterpart very quickly organized the services so that high quality animals were already available by the time Bacterial Enteric Diseases Research Program started.

The laboratory animal services are now fully operational. Satisfactory work is being done both in the breeding and experimental sections. Sufficient animals are now being supplied to all Research Division for their need which has greatly increased in variety and in volume, example are the Bacteriology and Virology Research Divisions are demanding and receiving thousands of animal per month for their surveys, the Pathology and Parasitology Research Division are inducing rodent malaria and animal model of cerebral malaria for their research and the Physiology Research Division is making parabiotic rats for growth studies.

A laboratory has been established and equipped in the laboratory animal Building for necessary laboratory work in disease control and genetic control. Disease control is being carried out with cooperation of the Bacteriology and other relavent research Division of DMR. A most rewarding information available for quality control was a recent report from NIH Tokyo that the animals are free from major diseases and in SPF (Specific Pathogen Free) condition free from Corynebacterum kutscheri, Bordetella bronchiseptica, Ectromelia virus for mice, Rabbit pox virus for rabbits, Sendai virus, Murine hepatitis virus, mouse adenovirus, Reovirus type 3, Tyzzen's disease organism Bacillus piliformis & mycoplasma pulmonis.

The breeding record and supply records that is not only to DMR but also to other medical institute as well are shown in the accompanying tables.

6.2 Training received

The senior technician received one year training in Japan in laboratory animal technology. 3 Japanese experts visited the division for varying period to set up and train the Burmese technicians.

6.3 Training given

Technician from various divisions were given training by the laboratory animal staff.

6.4 Research

A new Recombinant inbred strains was developed since 1981 are now leading to their F-10 generation. These strains are obtained from the cross between the two highly inbred strain of BALB/C and C_{57} BL/67.

6.5 Publications

A paper has been prepared in relation to intestinal adenomatosis associated with campylobacter like bacteria in guine pig. (see annex).

Table 64

Total Number of Animal Produced & Supplied During 1981 to 1983

(November 28th)

	Species	1981 Production / supplied	1982 Production / supplied	1983 Production / supplied	Total Production / supplied
1	MOUSE	39124 / 31088	59314 / 53411	52182 / 53307	150620 / 137836
2	RAT	992 / 570	3201 / 1395	618 / 839	4811 / 2804
3	RABBIT	87 / 11	488 / 206	502 / 170	1077 / 387
4	GUINEA PIG	257 / 37	444 / 280	342 / 147	1043 / 464
5	HAMSTER	232 / 103	352 / 140	158 / 144	742 / 351

Table 65

Total Number of Animals Supplied to Various Divisions of D.M.R. and Other Institutions During 1981 to 1983

	DMR	Mouse	Rat	Rabbit	Guinea pig	Hamster
1	Bacteriology	89,298	5	12	319	-
2	Virology	21,476	-	11	-	-
3	Pathology	5,922	80	45	16	245
4	Immunology	2,789	25	240	9	-
5	Parasitology	2,956	62	5	-	-
6	Physiology	4	322	-	-	-
7	Biochemistry	3,232	413	7	8	50
8	Entomology	519	8	31	-	-
9	Pharmacology	319	10	10	5	-
10	Clinical Research	934	518	-	1	-
11	Nuclear Medicine	572	-	-	-	-
12	Exp. Medicine	-	-	8	10	-
13	Nutrition	-	93	-	-	-
	Total	128,021	1,536	377	368	295

Table 66

	Others	Mouse	Rat	Rabbit	Guinea Pig	Hamster
1	National Health Laboratory	2,764	40	-	78	-
2	Burma Pharmaceutical Industry	2,847	-	2	-	-
3	Medical College Rangoon	31	117	-	18	-
4	Medical College Mandalay	-	320	-	-	-
5	Arts & Science University Rangoon	1,267	-	4	-	-
6	Veterinary Science Institute	361	-	-	-	-
7	Children Hospital	673	-	-	-	-
8	Rangoon General Hospital	-	10	-	-	-
9	Orthopaedic Hospital	-	70	-	-	-
10	Ear, Nose, Throat Hospital	-	6	-	-	-
11	Radio-Isotope Rangoon	-	-	2	-	-
12	Chemical Examination Department	-	-	2	-	-
13	Rodent Control Unit	363	-	-	-	-
14	D.C.P.T. Insein	425	-	-	-	-
15	Rangoon Zoological Garden	1,084	705	0	-	56
	Total	9,815	1,268	10	96	56

7. Development of Instrumentation Technology and Facilities

Development of the capability to maintain and repair the equipment and instruments for the Project was recognized as an important support service for the DMR/JICA Research Project.

The project assisted the development of instrumentation technology by training in Japan. Government has also strongly reinforced the scientific and technical personnel in the Instrumentation Division so that better and more regular maintenance & repair services are now possible. In order to maintain the efficiency and full work-horse capacity of the various electronic medical equipment after completion of the DMR/JICA project, importance is stressed on the availability of tools & test equipments, spare part, follow-up service and training facilities for engineers and technical staff.

8. Development of Library Facilities

Provision of adequate Library facilities is essential to research in any subject.

This Project could not give high priority to Library development. However, the Government budget has continued to provide fairly satisfactory supply of journals and has also expanded the staff considerably.

The Library provides services to all health related personnel all over the country and the dissemination of current biomedical literature is done through our Current Awareness Service which is carried out in collaboration with the Burma Medical Association. The Service was started in 1982 with the publication of contents lists of selected journals in the Burma Medical Bulletin. Through this service photocopies of articles can be requested on payment of a certain fee.

Bibliographies are compiled and regularly updated by this Library on major research projects carried out by this Department such as Snake-bite, Diarrhoea, Malaria and Dengue Haemorrhagic Fever.

Linkages have been made with the four major medical libraries namely the libraries of the Institute of Medicine (1), Institute of Medicine (2) and the Institute of Medicine, Mandalay such that a net work of medical libraries has been found with the compilation of a Union Catalogue of Books and Journals of these libraries in 1968, and the Interlibrary Loan System has been in operation since then. The advantages of the Interlibrary Loans System are enhanced with the introduction

of Coordinated Acquisition Scheme among these libraries which makes it possible to avoid unnecessary duplication and ommission of acquisition of journals and to extend the range of acquisition. Thus maximum use of available library resources is achieved.

Internationally this Library participates as the National Focal Point in the Health Literature, Library and Information Services (HELLIS) Network of Medical libraries of the Southeast Asian Region sponsored by WHO. Linkage has also been made with the Network Library of Medicine in the U.S.A from which MEDLINE services is acquired.

The provision of library services to the users is done directly as well as through the Interlibrary Loan System in the country and internationally through the HELLIS Network.

9. Field Work

9.1 North Okkalapa - a northern suburb of Rangoon and Intakaw groups of villages 40 miles from Rangoon were chosen as the site for the field work in connection with Bacterial Enteric Diseases. The North Okkalapa General Hospital which treats the people residing in North Okkalapa was also the site for some of the hospital based work on diarrhoea.

9.2 The Children Hospital, Rangoon, was the source of all the patient material for research and DHF. Various townships in Rangoon were the site for field studies in connection with viral and enterological studies on Dengue and Japanese encephalitis.

9.3 The People Council at various levels in the townships ward and villages cooperated very willingly and closely during the many field activities.

9.4 Staff from many Divisions including the Epidemiology, Medical Statistics, Parasitology and Clinical Research Divisions collaborated at various time in the DMR/JICA field work. The Administrative staff gave good support for the field activities especially for transport and vehicles.

9.5 The Mobile Research Laboratory which arrived in 1983 proved very useful for field work.

10. Training

Training is one of the components of the DMR/JICA project and consisted of the following:

(1) Training of Burmese scientists and technicians in Japan.

(2) Training of Burmese scientists and technicians in DMR by Japanese Experts and Burmese counterparts.

Training in Japan

Altogether 5 scientists-2 in Pathology, 2 in Virology, 1 in Instrumentation and 3 senior technician-1 in Virology, 1 in Entomology & 1 in Bacteriology were being trained at various institutions in Japan, 2 Scientists and a senior technician are scheduled to leave for Japan early next year. A scientist will be trained in Bacteriology & another in Virology. The senior technician will receive training in Immunological techniques.

In addition to the trainees mentioned above one engineer and a laboratory animal technician were trained by JICA under the regular country program. (but not under DMR/JICA project) in anticipation of the implementation of the present DMR/JICA project. The trainees have greatly accelerated development of facilities in DMR and made possible the rapid progress in research.

Training in Burma

In addition to the exchange of knowledge and skills which takes place between Japanese and Burmese scientists and the on-the-job training which accompanies the collaborative conduct of research work, Japanese scientists gave small-group training, in various subjects so as to impart knowledge to a wider audience. Small-group courses were conducted as follows:-

(a) Immuno-pathology by Professor Y. Hamashima

(b) Gas chromatography by Professor Y. Kanemasa

(c) Electron-microscopy by Professor K. Nakane

11. Japanese Experts

This component of the Project provides Japanese Experts to come to D.M.R. to help and advise with the execution of research and development of technology.

Japanese Experts in Bacteriology, Virology, Patho-immunology, animal technology, Electron-microscopy and hybridoma technology have come to D.M.R. under this Projects according to the needs of the Projects.

The Japanese Experts generally contributed in a valuable way towards progress of research and development of the necessary technology.

12. Supplies and Equipment

This vital component of the Project was vigorously implemented and involved extensive and repeated consultation between Japanese and Burmese scientist. List of requirements for 1982-1983, 1983-84 were drawn up according to the needs of project and was sent to JICA on September 1982 and June 1983 respectively and JICA undertook the ordering of supplies on the advice of an advisory groups of Japanese scientist in Japan. First consignment of supplies for 1982-1983 arrived in July 1983 and have since been arriving in batch consignments, consisting of the following categories:-

(a) Equipment and instruments.

(b) Chemical reagents, laboratory supplies and glasswares.

(c) Spare-parts for instruments and equipments.

Major items of equipment are:-

1. Electron microscope
2. Vacuum evaporator
3. Ultra microtome
4. Cryostat
5. Pellet machine
6. Oven 2K.V.A for Animal Supply Center
7. Spectrophotometers
8. Refrigerators
9. CO_2 incubator

10. Autoclave
11. Boiling sterilizer
12. Osmometer
13. Fluorescense Microscope
14. Microscopes
15. Dehumidifiers
16. Densitometer
17. Electronic Balance
18. Spare parts for airconditioning and refrigerator system
19. Spare parts for major equipments and instruments.

Major Equipments expected to arrive

1. Tissue -Tek II microtone
2. Food mixer
3. Ultra deep freezer
4. Ultrasonic washer
5. Electrophoresis system
6. Dry ice making machine attachment
7. Air gas generator
8. Refrigerated centrifuge
9. Roller drum incubator
10. Water bath incubator
11. Microcoagulometer
12. Vacuum pump
13. Exhaust fan
14. Oscilloscope as Synchroscope
15. Signal generator
16. Digital Multimeter
17. Frequency counter
18. Digital circuit tester
19. Ultra deep freezers gas charging equipment
20. Spare parts for various major equipment

Supplies and equipment for the final year 1983-84 mentioned above have been requested to JICA and will be arriving within the following year.

13. Other DMR Research Activities which Interact with DMR/JICA Project

The Department of Medical Research undertakes a wide range of research activities in its 15 Research Divisions & 3 Clinical Research Units attached to hospitals and through collaborative arrangement into their institution in the country. Although not concerned with major arboviral diseases or bacterial enteric diseases and therefore not directly under the DMR/JICA Project some of these research activities interact with those under the project since there can be no clear cut boundaries in scientific research.

Also some research activities, although not concerned with arboviral or bacterial enteric diseases are carried out in division strengthened by this project and have benefited by the development in technology.

Some of these other DMR research activities which interact with the research and developmental activities of the DMR/JICA Project are summarized below, in order that a proper perspective and assessment of the project may be made.

13.1 Pathology

(a) Measurement of F.D.P. and clothing defects in human cerebral malaria.

(b) Histopathological and ultrastructural studies of the brain in cerebral malaria. Morphological study up to ultrastructural level at the brain of a patient who died of cerebral malaria was carried out with particular attention on the interaction of infected red cells and endothelial cells lining the blood vessels. Multiple electron dense knob protrusion were found on the vessel. They were found to be points of adhesion to the surface of endothelial cell. We want to further strengthen the postulation that these knobs with their endothelial attachment are responsible for the deep vascular schizogony, obstruction of small blood vessels and ultimate cerebral complication in man to *Plasmodium falciparum* infection.

(c) Effect of exchange transfusion on histopathologic lesions in the liver of *plasmodium berghei* infected mice.

BALB/c mice were infected with *Plasmodium berghei* intraperitoneally and exchange transfusion was done on different days after inoculation. They were sacrificed one and two days after blood exchange. For each and every mouse undergoing exchange transfusion, two matched control mice were selected with similar degree of parasitaemia on the same day after inoculation. One served as negative control and sacrificed immediately without treatment. The other was sacrificed after two days of chloroquine therapy to be regarded as positive control. Thus, each experimental mouse has its own carefully matched positive and negative controls. Liver sections were made for light microscopy from all of them. The degree of mononuclear cell infiltration and sinusoidal pigmentation were assessed qualitatively and compared between test animal and their respective controls. Mononuclear cell infiltration was also quantified and compared similarly. The results showed that blood exchange reduced both mononuclear cell infiltration and pigmentation in the test animal.

(d) Role of disseminated intravascular coagulation in the pathogenesis of cerebral malaria. In collaboration with clinician from Defense Services General Hospital.

Coagulation parameters were studied in 40 cases admitted to the Defence Services General Hospital, with severe falciparum malaria in order to investigate the role of disseminated intravascular coagulation in the pathogenesis of cerebral malaria.

The control group consisted of twenty non cerebral malaria cases, while the test group consisted of twenty cases with cerebral malaria. The results showed disseminated intravascular coagulation to be present in about 5% of the cases with cerebral malaria. It is concluded that disseminated intravascular coagulation may not play a major role in the pathogenesis of cerebral malaria.

(e) Study of Hemostatic defects in experimentally envenomented rabbits is in progress.

(f) Study of Hemostatic defects in snake bite victims is in progress in collaboration with Clinical Research Unit and Research Division.

13.2 Immunology

(a) Development and testing of russells viper venom toxoid

Preliminary work in experimental animals was completed in the previous year during 1980-81. Human volunteers were tested with a toxoid of refined viper venom developed in DMR. Satisfactorily high level of antibody response was obtained lasting up to 6 months. There were no severe side effects this is the first time a viper venom toxoid has been developed and tested on human subject.

(b) Development of ELISA - for viper venom and observation of the kinetics of viper envenomation in animals.

(c) Russell's viper venom levels in serum of snake bite victims in Burma

Serum levels of venom antigen were measured using Enzyme-linked Immunosorbent Assay (ELISA) in 38 Russell's viper bite victims before and after administration of 40 ml of monovalent liquid antivenom. Initial serum levels ranged from 4.5 ng to 290 ng/ml in one case a level of 75 ng/ml was detected 27 hours after the bite. Serum venom levels after liquid monospecific antivenom therapy indicated that venom clearance was similar in each case to the natural clearance of venom in the absence of antivenom therapy. In one case a venom level of 11.5 ng/ml was detected 66 hours after liquid antivenom therapy whereas in two fatal cases, serum venom levels of 95 ng/ml and 185 ng/ml were detected after the same interval. Failure of complete neutralisation of venom is probably the result of loss of potency of antivenom during improper storage. The amount of venom excreted in the urine was not related to initial serum level.

(d) The efficacy of a bolus dose of Russell's viper antivenom in human viper bite victimes

Serum venom levels before and after administration of an intravenous bolus dose of 40 ml potent lyophilised monospecific antivenom in nine Russell's viper bite victims were measured by enzyme-linked immunosorbent assay (ELISA). In 3 out of 9 cases we studied, venom levels were still detectable in blood for more

than 2 hrs after therapy. Therefore, a single 40 ml bolus dose of antivenom is not sufficient to completely neutralise circulating venom in some cases of Russell's viper bite victims.

(e) Collaboration with the DMR Clinical Research Unit for snake-bite research in clinical study and clinical trial of viper bite patient at Tharawaddy Hospital.

(f) Collaboration with the DMR Clinical Research Unit for snake-bite research and with the Research Divisions on the pharmacokinetics of viper venom and antisera.

(g) Collaboration with clinicians of township hospitals on the efficacy of First aid measures against viper bite.

13.3 Bacteriology

(a) Monitoring of drug-resistant M. Leprae using mouse-foot pad method. This study has shown that drug resistance to Dapsone has developed in Burma. Further studies on factors influencing drug-resistance are being pursued.

(b) WHO multicentre hospital-based control study of the aetiology of diarrhoea in different geographic regions of the world. This study has provided information which further strengthens previous findings of the importance of enterotoxigenesis of *Escherichia coli* as a major bacterial enteric pathogen as found in the DMR/JICA Project on enteric bacterial diseases.

(c) Research on Bacteriological control of water has been undertaken as a preperatory task to aid in evaluation of health impact of the dry zone tube-well water supply programmes funded by UNICEF.

13.4 Virology

(a) Research in transovarial transmission of dengue virus in nature was begun earlier & was continued and completed in 1981. This work has an important and direct bearing an Arboviral research activities being undertaken under DMR/JICA Project.

(b) WHO multicentre multidisciplinary epidemiological study on DHF.

(c) WHO hospital based control study of aetiology of diarrhoea in different geographic regions.

13.5 Nutrition and Clinical Research

(a) Study of breast feeding and weaning practices in urban working mothers. This study has a relationship with bacterial enteric diseases research since breast feeding and weaning practice will influence the prevalence and distribution of acute diarrhoea in children.

(b) The efficacy and acceptability of oral rehydration therapy administered by village mothers. This study has shown that oral rehydration therapy given by village mother is effective in preventing dehydration but the long term nutritional improvement claimed by others is not confirmed. It has direct bearing on research on bacterial enteric diseases.

(c) Clinical trial of Becozamycin, tetracycline and placebo in acute diarrhoea.

(d) Change in aminoacid absorptive transport mechanisms in response to toxins of *V. cholerae* and enterotoxigenic *E. coli*. This basic research project, carried out at cellular and subcellular levels will provide information regarding the activity of aminoacid absorptive transport processes during diarrhoea; this information will provide the basis for clinical and field research projects involving feeding during diarrhoea.

(e) Effect of feeding during acute diarrhoea on clinical outcome. This clinical trial uses objective criteria to evaluate whether continued feeding during acute diarrhoea adversely affects its clinical course and outcome.

(f) Clinical trial of berberine in adult patients with diarrhoea. An indigenous drug berberine, which is an alkaloid from Coptis teeta wall (ခနိုးေတာက်ျမစ်), is studied in a randomized, double blind placebo controlled trial to evaluate its anti-secretory and vibriocidal effects.

(g) Composition and contamination of oral rehydration solutions (ORS) prepared in the home by village mothers. This is an operation research project to study the feasibility of village mothers preparing ORS using readily available household measures

(condensed-milk tins) and the extent of bacterial contamination that occurs when unboiled and boiled well water are used for preparation of ORS.

(h) Endotoxemia and septicaemia in neonatal gastroenteritis. In this study, neonates with acute gastroenteritis with and without clinical evidence of septicaemia are studied for bacterial pathogens in stools and blood, and for presence of endotoxin in blood. Result from this study will provide insight to measures to be recommended for preventing high mortality associated with gastroenteritis in neonates.

(i) Clinical trial of incomplete formula oral rehydration solutions in acute diarrhoea. Incomplete formula preparations containg salt and sugar, and salt and jaggery are evaluated in comparison to complete formula WHO (oralyte) solutions in a controlled clinical trial.

13.6 Epidemiology

(a) Development of epidemiological models for Ascaris infection and enteric diseases. Development of this epidemiological model for ascaris infection has enabled a similar model to be developed for bacterial enteric disease (acute diarrhoea).

13.7 Medical Entomology

(a) Genetic and enzyme stability of adult and isoenzyme pattern of aquatic stage of *An. balabacensis* spp. complex.

(b) Cytogenetic and hybridization studies among three strains of *An. balabacensis* spp. complex.

(c) Studies on ecology and biology of *An. balabacensis* spp. complex and *An. mininus*.

(d) Colonization cytogenetics and cross-fertilization studies of *Anopheles balabacensis* with different behavioral characters from two areas of Burma.

(e) Colonization of *An. mininus* & maintenance in the laboratory. Extensive studies in relation to the Anopholine Vector of malaria in Burma have been one of the major research programmes

of Medical Entomology Progress have been made in relation to characterization of species by modern techniques such as cytogenetics and isoenzyme studies of the various species complex of *An. balabacensis*. This would lead towards a better understanding of the ecology of the vector & direct effective control measures.

(f) Biology, ecology and control of *Aedes aegypti* a vector for DHF. In this context field studies have been conducted to provide infection on the population dynamic & vector competence of the vector of Dengue Haemorologic Fever. Further bio control by the use of dragon-fly nymphes *Bardinopyga gaminita rahme* have been used with success and is now in the phase where research has been conducted to assess the acceptance & participation of the community in establishing & maintenance of storing the dragon-fly nymphes in the house hold water containers.

(g) Rearing of *Toxosihychites splenders*. Apart from the research activities. Service is another component of work in the Medical Entomology Research Division Mosquitoes are reared to provide a constant demand of the Virology Research Division which utilize the mosquitoes for isolation of dengue viruses and in the study of biological variation of dengue viruses recovered from different diseases manifestation of Dengue Haemorrhagic Fever.

14. Academic Activities Relevent to the Project

The DMR conducts and participates in a variety of academic activities which are:-

(a) Attendance at meetings, seminars and workshops, both national, and international.

(b) Scientific talks and discussions in DMR

(c) Supervision of research of post-graduate students from Institutes of Medicines and Arts and Sciences Universities for the degree of M.Med.Sc, Diploma in Baceriology and M.Sc.

At present there are 2 master students in D.M.R., working on topics relevant to the project or in Divisions strengthened by the project. Some of the various academic activities are listed in the Annex. Since 1982 a total of 22 had completed post graduate research for the M.Med.Sc, Diploma in Bacteriology and M.Sc degrees.

15. Staff Development and Other DMR Inputs into the Project

15.1 Staff Deployment

The following staff are employed in Research Division, directly concerned with the Research Project.

Virology Research Division	- 5 scientists
	9 technicians
Bacteriology Research Division	- 4 scientists
	12 technicians
Pathology Research Division	- 3 scientists
Immunology Research Division	- 1 scientist
	6 technicians
Entomology Research Division	- 3 scientists
	11 technicians
Animal Supply Center	12 technicians
Instrumentation Division	- 3 engineers
	16 technicians

In addition, staff from Clinical Research Epidemiology and Medical Statistics Division are deployed on a large scale during field studies on Bacterial Enteric Diseases and collaboration is obtained from hospital and other health staff in the field area and from the community leaders.

15.2 Staff Development

Altogether 138 new staff posts were created in DMR since 1980 comprising 34 scientists, 43 technicians, 61 administrative staffs including lower echelon workers, 14 scientists, 26 technicians are for Research Division concerned with DMR/JICA Project or for technical support services.

15.3 Others

Motor vehicles of DMR are utilized extensively during the field studies for the project.

The Division concerned continue to draw from DMR central stores for part of their requirements.

16. Overall Assessment by DMR of the Project

The Aim of the Project is to conduct research on major arboviral diseases, bacterial enteric diseases and to apply these results for the control of these diseases in Burma.

The expressed aim of the Project may be considered to have been achieved. Research on acute diarrhoea which is the foremost bacterial enteric diseases in Burma and on dengue haemorrhagic fever which is the major arboviral diseases in Burma have been satisfactorily carried out. The results obtained are of general scientific significance as well as applicable to the control of these diseases in Burma. The quality of research is deemed satisfactory and in some instances may be regarded as of a high standard comparable to some internationally recognized institutions in other countries.

Hitherto unknown facts and understanding of the epidemiology and aetiology of childhood diarrhoea in Burma has been obtained. This will improve the control and treatment of this major health problem in Burma. Biological characteristics of the dengue virus and the dengue strains causing disease of varying severity has been studied and defined. It has added to the understanding of the pathogenesis of dengue haemorrhagic fever and will help prevent and tact this major health problem.

In addition to research of an applied nature a balanced development of basic research in bacteriology, virology, pathology and immunology has also taken place.

Sound expertise, technology and infrastructure for the study of arboviruses and bacterial enteric pathogens has been established on the basis of which fresh research efforts in these areas may be further launched. This improvement in self reliance in these two areas has been an implicit aim of the Project.

Apart from achieving the aim of the Project an equally important accomplishment has been the spin off of benefits to other research activities being carried out by DMR outside the DMR/JICA Project as a result of the build-up expertise and technology in the Divisions directly involved in the Project.

A vitally important but intangible achievement is the cross-fertilization of ideas and stimulation which has further taken place in DMR as a result of the implementation of the DMR/JICA Project.

ANNEX 1

MASTER PLAN OF THE PROJECT

1. Objective

The Project aims to conduct Research on major arbo-viral diseases, bacterial enteric diseases and the application of its results for the control of these diseases.

2. Implementation

The Department of Medical Research has overall responsibilities for the Project with the guidance of the Coordinating Committee. The Biomedical Research Centre which is an integrated functional component of the said Department is the executing organ for the achievement of the above mentioned objective.

3. Activities under the Project

Activities under the Project will be carried out at the Biomedical Research Centre premises including the pilot area:

Activities will include the following:

(a) Research on major arbo-viral diseases and the application of its achievement for their control.

(b) Research on major bacterial enteric diseases and the application of its achievement for their control.

(c) To further develop technology of laboratory and other services.

ANNEX 2

JAPANESE EXPERTS 1980-1984

Name/disignation	Subject	Date	Period
1980-81			
1. Professor Y. Hamashima Professor, Kyoto University	Pathology	30-10-80 to 29-12-80	2 months
2. Dr. H. Hayashi Associate Professor Okayama University	Bacteriology	1-11-80 to 29- 4-81	6 months
3. Dr. M. Nakagawa Head, Experimental Animal Laboratory National Institute of Health	Laboratory Animal Services	21-10-80 to 21-12-80	2 months
4. Professor A. Ohyama Professor, Kansai Medical School	Virology	16-12-80 to 13- 2-81	2 months
5. Professor Y. Kanemasa Professor, Okayama University	Bacteriology	19- 6-81 to 8- 8-81	1 3/4 months
1981-1982			
1. Professor Y. Hamashima Professor Kyoto University	Pathology	15-12-81 to 19- 1-82	1 month
2. Professor A. Ohyama Professor, Kansai Medical School	Virology	17-12-81 to 24- 1-82	1 1/4 months

3.	Professor P. Nakane Professor Tokai University	Pathology Electron Microscopy	16- 1-82 to 29- 1-82 16- 2-82 to 15- 3-82	1 1/2 months
4.	Dr. T. Asano Researcher National Institute of Health	Laboratory Animal Services	19- 1-82 to 16- 3-82	1 1/2 months
5.	Dr. H. Hayashi Associate Professor Okayama University	Bacteriology	19- 1-82 to 10- 3-82	1 3/4 months
6.	Dr. T. Ito Associate Professor Kansai Medical School	Virology	24- 1-82 to 23- 4-82	3 months
7.	Prof. Y. Hamashima Professor, Kyoto University	Pathology & Evaluation	19- 2-82 to 24- 2-82	1 week
8.	Prof. Y. Hamashima Professor, Kyoto University	Pathology	15- 6-82 to 21- 6-82	1 week
9.	Dr. A. Okabe Associate Professor Kagawa Medical School	Bacteriology	15- 6-82 to 14- 9-82	3 months
10.	Professor A. Ohyama Professor, Kansai Medical School	Virology	27- 4-82 to 3- 5-82	1 week
11.	Mr. E. Tanimura Senior Scientist Kansai Medical School	Virology	28- 8-82 to 27-10-82	2 months

12.	Dr. M. Nakagawa Head of Experimental Animal Laboratory National Institute of Health	Laboratory Animal Services	25-11-82 to 24-12-82	1 months

1982-1983

1.	Professor A. Ohyama Professor Kansai Medical School	Virology	18-12-82 to 14- 1-83	1 month
2.	Mr. N. Yamamoto Senior Scientist Kansai Medical School	Virology	13-11-82 to 10- 1-83	2 months
3.	Professor Y. Kanemasa Professor, Okayama University	Bacteriology	6- 1-83 to	3 weeks
4.	Dr. T. Muto Senior Researcher National Institute of Health	Laboratory Animal Services	17- 1-83 to 26- 2-83	1 1/2 months
5.	Prof. Y. Hamashima Professor Kyoto University	Pathology Immunology	5- 3-83 to 25- 3-83	3 weeks
6.	Prof. S. Morikawa Professor, Shimane Medical School	Immunology	5- 3-83 to 25- 3-83	3 weeks
7.	Professor T. Kanda Professor St. Marianna University	Medical Entomology	7- 4-83 to 14- 4-83	1 week
8.	Dr. T. Asano Senior Researcher National Institute of Health	Laboratory Animal Services	3-11-83 to 28-12-83	2 months

9.	Prof. H. Hayashi Professor, Kagawa Medical School	Bacteriology	21-11-83 to 9- 1-84	1 1/2 months
10.	Prof. A. Igarashi Professor, Nagasaki University	Virology	18-12-83 to 15- 1-84	1 month
11.	Prof. Y. Hamashima Professor Kyoto University	Pathology	18-12-83 to 15- 1-84	1 month
12.	Prof. P. Nakane Professor, Tokai University	Pathology Electron Microscopy	18-12-83 to 15- 1-84	1 month

ANNEX 3

Director General of the Department of Medical Research visited Japan in October 1982 for 14 days at the invitation of the Government of Japan, to visit various Medical Universities & Research Institutions.

Burmese Trainees Japan

Name	Educational Qualification	Position in DMR	Subject of Training	Period
(1980-1981)				
1. Daw Myat Myat Thu	M.Sc. (Zoology)	Technician Grade I Entomology	Entomology Technology	12- 3-80 to 11- 3-81
(1981-1982)				
1. U Aung Myint	B.Sc. (Chemistry)	Technician Grade I Virology	Virology Technology	23- 2-81 to 22- 2-82
2. U Myint Soe	B.E.	Research Officer Instrumentation	Instrument Maintenance and Repair	23- 2-81 to 22- 2-82

(1982-1983)

1.	Dr. Daw Than Than	M.B.,B.S. Ph.D.	Head Pathology	Pathology Research	25- 2-82 to 24- 2-83
2.	Dr.Kyi Kyi Khin	M.B.,B.S. D. Bact.	Senior Research Officer	Virology Research	25- 2-82 to 24- 2-83
3.	Daw Khin Sann Aung	B.Sc. (Bot)	Technician Grade I Bacteriology	Bacteriology Technology	1- 7-82 to 30- 6-83
4.	Dr. Mg Mg Oo	M.B., B.S. D.P.	Research Officer Pathology	Pathology Research	28- 2-83 to 2 years

(1983-1984)

1.	U Thet Win	M.Sc. (Zoology)	Research Officer Virology	Virology Research	9- 1-83 to 8- 1-84

(1983-1984) (Schedules to train in 1983)

1.	Daw Myint Myint Than	M.Sc (Zoology)	Technician Grade I Immunology	Immunology Technology
2.	Dr.Phyu Phyu Win	M.B., B.S. M.Sc. (Micro.)	Research Officer Bacteriology	Bacteriology Research
3.	Dr. May La Lin	M.B., B.S.	Research Officer Virology	Virology Research

Burmese Trained in Japan by JICA Before Project Implementation but in Anticipation of Project

(1977-1978)

1. U Soe Myint	M.Sc.	Senior Research Officer, Instrumentation Division	Instrument Maintenance and Repair.	1 year
2. U Khin Mg Zaw	B.Sc. (Zoo.)	Technician Grade I Animal Laboratory Services	Laboratory Animal Technology	1 year

ANNEX 4

LIST OF ACADEMIC ACTIVITIES RELEVANT TO THE PROJECT (1980 - 83)

I. WORKSHOP (INTER COUNTRY)

1. WHO Intercountry workshop on mosquito inoculation and tissue culture techniques for arbovirus isolation was held at the Virology Research Division of DMR: 15 March to 3 April, 1982.

2. WHO Regional workshop on Electron Microscopy and Immune Electron Microscopy was held at the Virology Research Division and Pathology Research Division of DMR: 28 November to 2 December, 1983.

II. WORKSHOP (INTERNATIONAL)

1. Dr. U Tun Pe, Head, (Immunology Research Division), and U Thet Win (Research Officer, Virology Research Division) attended the regional workshop on "Symposium on the properties of the monoclonal antibodies (produced by hybridoma technology) and their application to the study of diseases" held at the National University of Singapore: 19-23 October 1981.

2. Dr. Khin Ohn Lwin, (Senior Research Officer, Immunology Research Division) attended the training course on "Derivation of hybridoma-producing monoclonal antibodies" at Chiba University Japan which was sponsored by the Asian Molecular Biology Organization: 26 October to 4 November 1981.

3. Dr. Khin Maung Oo, (Head, Clinical Research Division) attended the workshop on "Clinical Trial in Acute Diarrhoea" held at ICDDR, B(International Centre for Diarrhoea Disease Research Bangladesh) in Dhaka sponsored by WHO/CDD programme: 14-22 November 1983.

III. WORKSHOP (National)

1. A workshop on ELISA methodology was held by the Immunology Research Division at DMR: 12-13 January, 1981.

IV. SEMINAR

1. Research Seminar on acute diarrhoea in childhood was held at the Department of Medical Research: 27 February, 1982.

V. SCIENTIFIC MEETINGS

a. International Participation

1. Ohyama A., Thet Win & Tanimura E. (1981) Isolation and identification of Dengue virus (Type 11) by Toxorhynchites mosquitoes. The 23rd Annual Meeting of Japanese Society of Tropical Medicine, 1981 Tokushima, Japan.

2. Ohyama A., Thet Win & Tanimura E. (1981) Development of Dengue virus in Toxorhynchites mosquitoes after intercerebral inoculation technique. The 29th Meeting of Japanese Virologists, 1981 Tokyo, Japan.

3. Ito T., Tanimura E., Tamamoto N., Nalamichichi A., Aung Myint & Ohyama A. (1981) Quantitative detection of Dengue and Rubella Viruses using cultured cells. The 29th Meeting of Japanese Virologists, 1981 Tokyo, Japan.

4. Khin Maung U (1980) Laboratory and Field studies in Diarrhoea of Adults and Children Tropical Climates Presented at Clinical Gastroenterology Meeting on 12-12-80 at the MRC Clinical Research Centre, Harrow, Middlesex, England.

5. Khin Maung U (1981) Intestinal Transport Studies of Calcium and Phosphate: a model for ionic transport study in diarrhoea. Presented at Clinical Cell Biology Seminar on 3-4-81 at the MRC Clinical Research Centre, Harrow, Middle-sex, England.

6. Khin Maung U (1981) Operational Research Methodology for domiciliary oral rehydration of acute diarrhoea among village children in Burma, and its effect on their growth and nutrition, Presented at Harvard University School of Public Health on 7-5-81 Boston, Massachusetts, USA.

7. Khin Maung U (1981) Effect of oral rehydration on growth and nutrition in rural Burma. Presented at International Food and Nutrition Programme Seminar, on 7-5-81 United Nations University, Massachusetts Institute of Technology, Boston, Massachusetts, U.S.A.

8. Khin Maung U (1981) Operation Research Method to study feasibility, acceptability and effectiveness of oral rehydration for acute diarrhoea among village children by their mothers at home, and its effect on their growth and nutrition. Presented at Inter-University School of Medicine, Baltimore, Maryland, U.S.A.

9. Mi Mi Khin, (1980) Development of rapid laboratory diagnostic test for Arboviral infections. Participation as Member Scientific Group Meeting on development of rapid laboratory techniques for viral infections. 29th Sept.: - 4 Oct: 1980 at WHOHQ Geneva, Switzerland.

10. Mi Mi Khin, (1982) Viral infections in Burma. To present at the International Seminar on Viral Diseases in Southeast Asia and Western Pacific, 8-12 Feb. 1982 at Canberra, Australia.

11. The studies of dengue virus type 2 in *Toxorhynchites* cell line (TRA-711)- Replication and morphological observation - The 25th Annual Meeting of Japanese Society of Tropical Medicine, 1983 Osaka, Japan.

12. Dr. Khin Maung Tin (Deputy Director, Research) attended the Regional Advisory Committee on Medical Research Meeting on Dengue Haemorrhagic Fever held in New Dehli, India: 30 March 1981.

13. Dr. Daw Tin Aye(Head, Bacteriology Research Division) attended the Second Asian Conference on Diarrhoeal Diseases held in Calcutta, India: 22-25 February 1982.

14. Dr. U Thane Toe (Deputy Director, Research) attended the International Conference of Oral rehydration therapy, he d at Washington DC: 7-10 June 1983, sponsored by UNICEF.

b. Scientific meetings and talks at DMR

1.	Dr. Mi Mi Khin	(27-12-80)	Detection of Dengue antigen in LLCMK/2 cell lines
2.	Dr. H. Hayashi	(21-2-81)	Structure and function of erythrocyte membrane
3.	Dr. Myo Thein	(7-3-81)	Stamina
4.	Dr. H. Hayashi	(21-3-81)	Current review on the basis of immune reaction
5.	Dr. Aung Khin	(4-4-81)	The strategy of life
6.	Ma Phyu Phyu	(18-4-81)	Protective action of G-6-PD deficiency against malaria
7.	Daw Thawka Kyin	(16-5-81)	Induction of immunity in mice by *Ascaris suum* eggs
8.	Dr. Ne Win and Dr. Khant	(6-6-81)	Recent trend in drug abuse
9.	U Tun Khin	(13-6-81)	Quality control in medical laboratory technology
10.	Dr. B. Murphy	(20-6-81)	Laboratory diagnosis of viral hepatitis
11.	Dr. Maung Maung Oo	(4-7-81)	Basic principles of calculation in digital computers

12.	Prof. R.L. Smith	(17-7-81)	Pharmacokinetics and drug metabolism
13.	Dr. Y. Kanemasa	(25-7-81)	Phospholipid composition of *Staphylococcus aureus*
14.	Dr. R.D. Piyasena	(15-8-81)	Recent testicular function control-recent-research development
15.	Dr. Willougby Tun Lin	(5-9-81)	People's Health programme in Burma
16.	Dr. David A. Sack	(30-9-81)	Recent advances in Research on aetiology and therapy of acute diarrhoea
17.	U Thet Win	(3-10-81)	Detection of Dengue virus by immunofluorescence after intracerebral inoculation of *Toxorhynchites splenders* mosquitoe from mononuclear cells of Dengue haemorrhagic fever patients
18.	U Khin Maung Zaw	(17-10-81)	Breeding of nude mouse at DMR animal house
19.	Dr. Y.H. Bang	(24-10-81)	Applied research for National Vector Borne Disease Control Programme
20.	Daw Htay Htay Aye	(7-11-81)	Practical use of calculators in Biomedical Research
21.	Dr. Daw Tin Aye		Experience in International Centre for Diarrhoeal Diseases Research
22.	Dr. Alton I. Sutmik	(30-11-81)	Hepatitis B and liver cancer
23.	Dr. Y. Sawai	(8-12-81)	Recent advances in pathogenesis and medical treatment of snake-bite

24.	Dr. Thein Hlaing	(19-12-81)	Conceptual epidemiological model of Ascariasis
25.	Dr. U Tun Pe Dr. Daw Khin Ohn Lwin U Thet Win	(20-1-82)	Hybridoma
26.	Professor P. Nakane	(23-1-82)	Peroxidase labelled antibody method
27.	Professor B. Cvjetanovic	(12-2-82)	Epidemiological models of acute bacterial and viral diseases
28.	Dr. Arlene Mclean Ph. D	(25-2-82)	Clinical Trials: Design and conduct
29.	Professor P. Nakane	(6-3-82)	Mechanism of IgA Transport in Gastrointestinal Tract
30.	U Tin Win	(21-1-83)	Anticholinesterase activities of some anthelmintic agents and some medicinal plants
31.	Dr. Thein Than	(4-2-83)	Snake Venoms
32.	Mg Mg Thwin	(25-2-83)	The use of radiolabelled Russell's viper venom in distribution studies and prediction of venom doses in mice.
33.	Prof. Y. Hamashima	(18-3-83)	New findings in Kawasaki's disease
34.	Prof. Tozo Kanda	(12-4-83)	Genetics of culicine family
35.	Miss Helena Shayn	(27-4-83)	1. Electrophoresis
	Dr. Thomas Warving		2. Isoelectric Focussing
	"	(28-4-83)	1. Affinity Chromatography
			2. Chromato-focussing
	"	(29-4-83)	1. Fast Protein Liquid Chromatography
36.	Dr. V.M. Duncombe	(13-5-83)	Research techniques in Intestinal absorption

37.	Dr. D.S. Warrell	(27-5-83)	Management of Snake bite
38.	Daw Htay Htay Aye	(27-5-83)	Composite Scheme of Socioeconomic Index to be used in Infant Feeding and Weaning Practices Survey
39.	Dr. D.A. Warrell	(2-6-83)	Recent advances in the mangement of severe Falciparum Malaria
40.	U Chit Mg	(3-6-83)	Medicinal and dietetic application of Bee-Products in some major user countries
41.	Dr. W. Tun Lin	(17-6-83)	Development and testing of mass screening techniques for detection of malaria sporozoites in female anopheline mosquitoes
42.	Dr. Saw Mya Yee	(19-7-83)	Performance characteristic of sandwiched enzyme linked immunosorbent assay (ELISA) for serum alpha fetoprotein
43.	U Tun Pe	(18-8-83)	Production of monoclonal antibodies against sheep red blood cells
44.	U Soe Myint	(2-9-83)	Spectophotometric measurements
45.	Dr. T.D. Bolin & Dr. V.M. Duncombe	(15-9-83)	Disaccharidase deficiency
46.	Dr. G.B. White	(16-9-83)	Malaria vectors in Burma: Some problems to solve
47.	Dr. Kyi Kyi Khin	(7-10-83)	Health information for health planning in Japan
48.	Dr. Thein Hlaing	(18-11-83)	Malaria models
49.	Dr. J.M. Davies	(24-11-83)	Hybridoma researches in Washington University

50.	L. Molineauz	(9-12-83)	Malaria models
51.	Prof. A. Igarashi	(6-1-84)	Epidemiological Studies on encephalitis in Chiang Mai area, Thailand (1982)
52.	Prof. P.K. Nakane	(11-1-84)	Use of enzyme labeled antibodies

Postgraduate Students Conducting Research in Topics Relevant to DMR/JICA Project or in Divisions Strengthened by This Project
(1980-1984)

1. Aye Aye Myint : (1980-81) M. Sc. (Zoology)
Rangoon University

Title of Thesis : Establishment, Standardization and Application of ELISA in Russell's viper evenomation
(Immunology Research Division, DMR)

2. Dr. Phyu Phyu Win : (1980 to date) M. Sc. (Micorbiology)
Institute of Medicine, Mandalay

Title of Thesis : A study of Cell-mediated Immunity in Leprosy patients
(Immunology Research Division, DMR)

3. Dr. Soe Thein : (1981- to date) M. Med, Sc. (Microbiology).
Institute of Medicine, Mandalay

Title of Thesis : Viability of JE virus in glyceronized mosquitoes and tissues.
(National Health Laboratory & Virology Research Division, DMR)

4. Khin Mar Aye : (1981-to date) M. Sc. (Zoology)
Rangoon University

Title of Thesis : Virulence and temperature sensitivity of Dengue viruses
(Virology Research Division, DMR)

5. Dr. Mya Mya Ohn : (1981-to date) M. Med, Sc. (Paediatrics)
Institute of Medicine (1), Rangoon

Title of Thesis : Neonatal diarrhoea - major etiological agents & mortality patterns
(Clinical & Bacteriology Research Division, DMR)

6. Thazin Lay : (1983--1935) M. Sc. (Zoology)
Rangoon University

Title of Thesis : Replication and persistence of dengue virus in Aedes albopictus (C636) cell culture
(Virology Research Division, DMR)

7. Dr. Tin Aung : (1982-1983) M. Med. Sc.
Institute of Medicine I, Rangoon

Title of Thesis : Effect of E. coli enterotoxin on aminoacid absorption by animal intestinal tissues

ANNEX 5.

LIST OF PAPERS READ OR PUBLISHED (1980-1984)

5.1 Papers read or published on topic relevant to DMR/JICA Project

(a) Bacterial enteric diseases and diarrhoea

1. Khin Maung U (1980). Laboratory and Field studies in Diarrhoea of adults and children in Tropical Climate. Presented at Clinical Gastroenterology Meeting on 12-12-80 at the MRC Clinical Research Centre, Harrow, Middlesex, England.

2. Khin Maung U (1981). Intestinal Transport Studies of Calcium and Phosphate a model for ionic transport study in diarrhoea. Presented at Clinical Cell Biology Seminar on 3-4-81 at the MRC Clinical Research Centre, Harrow, Middlesex, England.

3. Khin Maung U (1981). Operational Research Methodology for domiciliary oral rehydration of acute diarrhoea among village children in Burma, and its effect on their growth and nutrition. Presented at Harvard University School of Public Health on 7-5-81 Boston, Massachusetts, U.S.A.

4. Khin Maung U (1981). Effect of oral rehydration on growth and nutrition in rural Burma. Presented at International Food and Nutrition Programme Seminar on 7-5-81 United Nations University, Massachusetts Institute of Technology, Boston, Massachusetts, U.S.A.

5. Khin Maung U (1981). Operation Research Method to study feasibility, acceptability and effectiveness of oral rehydration for acute diarrhoea among village children by their mothers at home and its effect on their growth and nutrition. Presented at Inter-university School of Medicine, Baltimore, Maryland, U.S.A.

6. Tin Aye, K. Wachmuth, J.C. Feeley, R.J. Gibson and S.R. Johnson, (1981). Plasmid profiles of Legionella Species. Current Microbiol., 6: 389-394

7. Tin Aye, Mar Mar Nyein. (1982). Etiological agents responsible for acute diarrhoea in children. Proceedings of the Research Seminar on Acute diarrhoea in childhood, Department of Medical Research, Rangoon, Burma.

8. Tin Aye, Mar Mar Nyein (1982). Recent knowledge on Bacterial enteric Pathogens of acute diarrhoea diseases. Proceedings of the Research Seminar on acute diarrhoea in childhood. pp 73-78, Department of Medical Research, Rangoon, Burma.

9. Khin Maung U (1982). Community management of acute diarrhoea. Proceedings of Research Seminar on Acute Diarrhoea. 113-142. Department of Medical Research, Rangoon, Burma.

10. Khin Maung U (1982). Recent advances in the Pathophysiology and management of acute diarrhoea. Proceedings of Research Seminar on Acute Diarrhoea. 143-158. Department of Medical Research, Rangoon, Burma.

11. Kyi Kyi Khin (1982). Viral agents causing diarrhoea in childhood. Proceedings of the Research Seminar on Acute Diarrhoea in childhood. 79-93. Department of Medical Research, Rangoon, Burma.

12. Aung Myo Han (1982). Epidemiology of acute diarrhoea in childhood. Proceedings of the Research Seminar on Acute Diarrhoea, 1-20, Department of Medical Research, Rangoon, Burma.

13. Aung Myo Han, Thein Mg Myint, Thein Hlaing and W. Tun Lin. (1982). Incidence of acute diarrhoea in children under five years in North Okkalapa township. Proceedings of the Research Paper Reading Session, Medical Sciences Division, Abstr., 4, Rangoon, Burma.

14. Tin Aye (1983). A comparative study of heat-labile toxin of ETEC by CHO, GM_1 ELISA and Biken assay systems. Presented at 2nd Asian Conference on diarrhoea diseases on 21-24 Feb. 1983, Calcutta, India.

15. Tin Aye, Mar Mar Nyein, Y. Kanemasa and H. Hayashi (1983) Etiological agents responsible for acute diarrhoea in children in an urban community in Burma. Microbiol. Immunol. 27(6), 551-556.

16. Thane Toe, Khin Maung U, Tin Aye, Mar Mar Nyein and Ye Htut. (1984). Oral rehydration therapy in the home by village mothers in Burma. Trans. of the Roy. Soc. of Trop. Med. & Hyg., (in Press).

(b) Dengue Haemorrhagic fever and arboviral infections

1. Anthony Sebastian, Myat Myat Thu and Myint Myint Sein (1980) The use of dragonfly nymphs in the control of Aedes aegypti. Southeast Asian J. of Trop. Med. & Hyg. Publ. Hlth, 1:(11), 104-107.

2. Oyama, A., Thet Win & Tanimura E. (1981). Isolation and identification of Dengue virus (type 2) by Toxohynchites mosquitoes. The 23rd Annual Meeting of Japanese Society of Tropical Medicine, 1981 Tokushima, Japan.

3. Ito T., Tanimura E., Yamamoto N., Nakamichi A., Aung Myint and Ohyama A., (1981). Quantitative detection of Dengue and Rubella viruses using cultured cells. The 29th Meeting of Japanese Virologist 1981, Tokyo, Japan.

4. Thet Win and Ohyama, A., (1981). Detection of Dengue 2 protype virus in Toxorhnchites mosquitoes using intracerebral inoculation techniques. Dengue Newsletter. Southeast Asian and Western Pacific Regions, W.H.O., 7: 51-52.

5. Mi Mi Khin, (contributor). Rapid Laboratory techniques for the diagnosis of viral infections. Report of a WHO Scientific Group, Technical Report Series 661, WHO Geneva 1981.

6. Ito, T., Tanimura, E., Yamamoto, N., Aung Myint, and Ohyama, A. (1981). Macroscopic observation of slow appearing cytotoxic viruses in cultured cells by EFFA (Enzyme Focus Forming Assay). Proceedings of 29th Annual Meeting of the Society of Japanese virologist (in Japanese). 20-23 October 1981, Tokyo, Japan.

7. Thet Win (1982). Detection of Dengue virus by immunofluorescence after intracerebral inoculation of mosquitoes. Lancet, 2: 53-54.

8. Mi Mi Khin (1982). Viral infection in Burma. Mackenzie, J.S. ed, Viral diseases in Southeast Asia and the Western Pacific. Sydney, Academic Press, 210-216.

9. Kyi Kyi Khin (1982). Neutralizing capacity of the sera of dengue haemorrhagic fever patients to Japanese encephalitis virus. Journal of the Kansai Medical University, Supplement of Vol. 34.

10. Mi Mi Khin, Khin Aye Than and Tin U (1982). Recovery of Dengue Viruses from autopsy material. Proceedings of the Research Paper Reading Session, Medical Sciences Division.

11. Mi Mi Khin and Khin Aye Than (1983). Transoviral transmission of dengue 2 virus by Aedes aegypti in nature. Am. J. Trop. Med. Hyg., 32: 590-591.

12. Fujiwara, H., Kao, T-C., Shimizu, J., Fujiwara, T., Maung Maung Oo and Hamashima Y.: Microorganism in the heart in Kawasaki disease.
Lancet, Sep 10, 621, 1983.

13. Ikehara, S., R. A. Good, Nakamura, T., Sekita, K., Inoue, S., Maung Maung Oo, Muso, E., Ogawa, K. and Hamashima, Y.: Rationals for bone marrow transplantation in the treatment of autoimmune diseases.
Nature, Submitted. 1984.

14. Ikehara, S., Inoue, S., Nakamura, T., R.A. Good, Maung Maung

Oo and Hamashima, Y.: Demonstration of age-related increase in T cell functions of nu/nu mice by sensitive assay methods. J. Immunol. 1984 (submitted)

15. Maung Maung Oo, Ikehara, S., Sekita, K., Nakamura, T., Inoue, S., Susuki, M., Igarashi, I. and Hamashima, Y.: Immune mechanisms in murine malaria: 1. periods of survival correlate with immune complex levels.
Clin. Exp. Immunol. 1984 (submitted)

16. Maung Maung Oo and Than Than.: In vitro effect of Russett's viper verom on platelet aggregation.
Archires of Tonicology. 1984 (submitted)

5.2 Papers read or published on other topics

(a) Snake-bite research

1. M. Aung Khin, Khin Ohn Lwin & Thant Zin, (1980). Immunogenicity of the toxoid of Russell's viper venom. The Snake, 12: 45-53.

2. Aye Kyaw, Thant Zin & Aung Khin (1982). Partial purification and some properties of Caseinolytic activity in Russell's viper venom. The Sanke, 14: 18-22.

3. Khin Ohn Lwin and Aye Aye Myint (1982). The use of enzyme linked immunosorbent assay (ELISA) in detection of Russell's viper venom in body fluid. The Snake, 14: 77-82.

4. Khin Ohn Lwin, Aye Aye Myint, Tun Pe, Theingie Nwe and Min Naing. (1984). Russell's viper venom levels in serum of snake-bite victims of Burma. Transaction (in press).

5. Tun Pe, Aye Aye Myint, Min Naing and Thein Win. The efficacy of bolous dose of Russell's viper antivenom in human viper bite victims. Presented at 2nd Conference of Medical Specialities. 13-17 October, 1983, Rangoon.

6. Mg Mg Thwin, Thein Than and Hla Pe. The relationship of administered dose of venom levels in blood following

experimental envenomation of Russell's viper (vipera russelli) venom in mice. Toxicon. (in press)

(b) Nutrition

1. Tin Tin Oo, and Khin Maung Naing (1980). Breast feeding and weaning practices in Burma. The Proceedings of a Research Seminar on Breast feeding and weaning practics in Burma. Department of Medical Research, Rangoon, Burma.

2. Khin Maung Naing, Tin Tin Oo, Kywe Thein and Nwe Nwe Hlaing (1980). A study on lactation performance of Burmese mothers. Am. J. Clin. Nutr. 33: 2655-2668.

3. Tin Tin Oo and Khin Maung Naing (1982). A comparison of milk output of Burmese mothers by three different methods. Reference Food and Nutrition Bulletin, 4: (4), 66-68.

4. Thane Toe and Thein Than (1982). Serum Ferritin studies in Burmese Pregnant Women. Am. J. of Clin. Nutr., 35: 95-99.

5. Iles, C.A., Sharman, I.M., Thane Toe, Wardsworth, G.R.(1983) Material diet and nutrition during early pregnancy and after delivery in North London. In "Prevention of Spina Bifida and other Neural tube Defects", Ed: John Dobbing, Academic Press, London, 1983.

(c) Viral Hepatitis and Liver Diseases

1. Khin Mg Tin and Tun Khin. Alcohol and the liver. The Proceedings of the 28th Burma Medical Conference, 1981, Rangoon, Burma.

2. Khin Maung Tin, Tin Htut, Hla Myint & Tun Khin. Prevalance of Hepatitis A & B in Burma. The Proceedings of the 1st Conference of Combined Medical Specialities, 1981, 1: 26-31, Rangoon, Burma.

3. Khin Maung Tin, Myo Thwe and Myo Han (1982). Chapter 9, Liver and Biliary tract. Text book of Internal Medicine, Section of the Burma Medical Assoc. 9.1-9.48, Rangoon,

Burma.

4. Aye Kyaw, Thidar Aung, Tin Htut, Hla Myint and Khin Maung Tin (1983). Lysosomal enzyme activities in normal and in patients with chronic liver diseases. Clinical Chimica Acta (Japan). 131: 317-323.

5. Hla Myint, Khin Maung Tin, Nwe Nwe Oo, Tun Khin, and Myint Myint Soe. Non-A, Non-B Hepatitis in Rangoon, The 2nd Conference of Combined Medical Specialities, 1983, Rangoon, Burma.

(d) Malaria

1. Long, E., Lwin M., Targett, G, Dornhoff, M. (1981). Factors affecting the acquisition of resistance against *Schistosoma mansoni* in the mouse, VIII Failure of concurrent infections with *Plasmodium chabaudi* to effect resistance to reinfection with *S. mansoni*. Am. Trop. Med. Parasitol. 75(1): 79-86.

2. Lwin M., Last, C., Targett, G.A., Dornhoff, M.J. (1982). Infection of mice concurrently with *Schistosoma mansoni* and rodent malarias: contrasting effects of patent S. mansoni infections on *Plasmodium chabaudi, P. yoclii* and *P. berghei* Ann. Trop. Med. Parasitol., 76: (3) 265-273.

3. Hla Pe, Phu Phu, Tin Win, Win Myint Than and Franco Tin (1982). Protective action of glucose-6 phosphate dehydrogenase deficiency against malaria. The Proceedings of the Research Paper Reading Session, Medical Sciences Division, 1982, Abstr., p 5, Rangoon, Burma.

4. Myint Oo, Myint Lwin, Min Zaw & Ye Htut (1983). Biochemical characterization of chloroquine-resistant isolates of *Plasmodium falciparum*. Trans. Roy. Soc. Trop. Med. Hyg. (in press).

(e) Pharmacology and Indigenous drugs

1. S.J. Tha, Aye Than, Thawka Kyin and Chit Mg (1981). Drug oxidative status of Burmese with reference to Antipyrine.

Proceedings: 1st Conference of Combined Medical Specialities, 1981, 82-86, Rangoon, Burma.

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1. プロジェクト協力期間中JICAより送付した機材実績

1－1　機材供与実績

年度	輸送	主たる品目	価格 EX GO DOWN	CIF (円)
55	56. 3. 16 AIR CARGO	薬品	587,860	637,395
	56. 3. 29 WAKAMATSU MARU	〃	1,315,530	1,394,309
	56. 5. 6 CRISTOBAL MARU	パーツ類	14,556,060	15,065,656
	56. 8. 11 WAKAMATSU MARU	〃	15,743,940	16,546,152
	56. 10. 29 WAKAMATSU MARU	〃	5,200,000	5,810,778
56	57. 2. 9 BOSTON MARU	電顕	25,000,000	25,515,217
	57. 7. 1 NOSHIRO MARU	機器	23,315,570	24,050,209
	57. 7. 2 FUSO MARU	〃	10,204,430	10,423,046
	57. 8. 28 AIR CARGO	薬品	2,134,860	2,509,161
	57. 9. 14 CLOVER	機器	1,695,140	1,762,094
	58. 3. 22 SIT TWAY	〃	2,569,290	3,440,000
57	58. 7. 1. CORINTO MARU	機器	35,150,000	36,566,098
	58. 8. 2 NOSHIRO MARU	車輌	3,783,500	4,187,514
	59. 3. 25 AIR CARGO	薬品	1,103,280	1,228,341
58	59. 3. 27 NAGANO MARU	〃	1,990,720	2,207,804
	59. 4. 29 LONG BEACH	機器	33,337,070	35,014,513
	59. 5. 28 NAGANO MARU	〃	2,694,300	2,841,326
	59. 5. 27 AIR CARGO	薬品	4,027,780	4,298,959

Description of Goods		Quantity
O. Nitrophenyl Galactophyranoside	25g	1 pc.
Alkaline Phosphatase Type VII	5mg	2 pcs.
Gelatin, Type I	100g	1 pc.
Adenosine-5-Phosphoric Acid Sod. Salt Type III	1g	5 pcs.
Lithium Lactate	50g	2 pcs.
Tetrohydrofolic Acid Grade III	250mg	2 pcs.
Dihydrofolic Acid	100mg	2 pcs.
L-L-Lecithin	100mg	1 pc.
Peroyidase	5000U	5 pcs.
Streptokinase	50000U	5 pcs.
Adenosine-5-Phosphoric Acid	5g	1 pc.
Ethylhydrocupreine HCL	1g	1 pc.
M.T.T.	1g	1 pc.
B-Nicotinamide Adenine Dinucleotide Grade III	1g	1 pc.
B-Nicotinamine Adenine Dinucleotide Phosphote Sod. Salt	1g	1 pc.
P-Nitrophenyl Phosphate Disodium	1g	3 pcs.
Alkaline Phosphatas Type I	500mg	4 pcs.
Pyridoxal-5-Phosphate	1g	2 pcs.
Dipalmitoyl-L-£-Phosphatidylchocine	1g	1 pc.
Agarose	100g	2 pcs.
HEPES	100g	2 pcs.
Trizma Base	500g	2 pcs.
Albumin, Bovine	1g	2 pcs.
Rhodamine Isothiocyanate	100mg	1 pc.
Complete Adjuvant	10ml x 5A	1 pc.
Heparin Sodium Grade I	100000U	2 pcs.
Pokoweed Mitogen	10ml	2 pcs.

TOTAL:		55 pcs.	¥587,860
	F.O.B. Expensive		¥13,435
	F.O.B. Japan		¥601,295
	Air Freight		¥34,026
	Insurance		¥2,074
	C.I.F.		¥637,395

Description of Goods		Quantity
Acetic Acid	G.R.	500g x 10
Acetone	G.R.	500g x 1
Acetylacetone	G.R.	500g x 1
Ethyl Ether	G.R.	500ml x 1
Ethyl Alcohol	G.R.	500g x 8
Ethyl Acetate	G.R.	500g x 1
Methyl Acetate	G.R.	500g x 1
Propylene Oxide	G.R.	500g x 3
Xylene	G.R.	500g x 5
Iso-Amyl Acetate	G.R.	500g x 1
Tween 20		500g x 3
Tween 40		500g x 1
Tween 60		500g x 1
Tween 80		500g x 1
Ethylene Glycol	G.R.	500g x 1
Collodion 2%		100g x 2
Glutaldehyde 25%		25ml x 10
Balsam Canada	E.P.	100g x 1
Giemsas Sol.		500ml x 1
Sodium Borohydride		25g x 6
Acetyl Bromide	G.R.	25g x 1
Potassium Hydroxide	G.R.	500g x 2
Sodium Hydroxide	G.R.	500g x 3
Trichloroacetic Acid	G.R.	500g x 2
Sodium Hypochlorite	E.P.	500g x 3
Ammonia Water 28%	G.R.	500g x 5
Cupper Sulfate Anhyd	G.R.	500g x 1
Sodium Formate	G.R.	25g x 1
Ammonium Persulfate	G.R.	500g x 1
Sodium Nitrite	G.R.	500g x 3
Hydrogen Peroxide	E.P.	500g x 3
Lead Peroxide	G.R.	100g x 1
Ammonium Sulfate	G.R.	500g x 4
Sodium Chloride	G.R.	500g x 1

Description of Goods		Quantity
Boric Acid	G.R.	500g x 1
Sodium Periodate	G.R.	25g x 2
Sodium Iodate	G.R.	500g x 1
Chloroform	G.R.	500g x 10
Potassium Dichromate	G.R.	500g x 1
Potassium Dichromate	G.R.	500g x 10
Calcium Chloride	G.R.	500g x 1
E.D.T.A.	G.R.	500g x 1
Potassium Chloride	G.R.	500g x 2
Sodium Dedecylsulfate	E.P.	250g x 1
Sodium Borate	G.R.	500g x 1
Sodium Phosphate Monobasic	G.R.	500g x 5
Ammonium Acetate	G.R.	500g x 2
Sodium Axide		25g x 2
Sodium Gyanide	G.R.	500g x 1
Phenol Reagent		100ml x 5
Sodium Barbital	G.R.	500g x 1
Barbital	G.R.	500g x 1
Sodium Barbital	G.R.	500g x 1
3.3-Diaminibendizine		25g x 1
3.3-Dimenthoxybendizine	E.P.	25g x 2
Lead Hydroxide	E.P.	500g x 1
Lead Citrate	E.P.	500g x 1
Osmic Acid		1g x 3
2.4-Dinitrofluorobenzene	G.R.	25g x 3
Sulfuric Acid	G.R.	500g x 12
Chromium Standard Sol.	A.A.	250ml x 2
Lead Standard Sol. 1,000 PPM.	A.A.	250ml x 2
Cadmium Standard Sol. 1,000 PPM.	A.A.	250ml x 2
Calcium Standard Sol.	A.A.	250ml x 2
Iron Standard Sol. 1,000 PPM.	A.A.	250ml x 2

Description of Goods		Quantity
Magnesium Standard Sol. 1,000PPM.	A.A.	250ml x 2
Manganese Standard Sol. 1,000PPM.	A.A.	250ml x 2
Potassium Standard Sol. 1,000PPM.	A.A.	250ml x 2
Sodium Standard Sol. 1,000PPM.	A.A.	250ml x 2
Tin Standard Sol. 1,000PPM.	A.A.	250ml x 2
Aluminum Standard Sol. 1,000PPM.	A.A.	250ml x 2
Copper Standard Sol. 1,000PPM.	A.A.	250ml x 2
Mercury Standard Sol. 1,000PPM.	A.A.	250ml x 2
Hydrochloric Acid	G.R.	500g x 5
Hydrochloric Acid	A.A.	500g x 1
Tetramethyl-p-Phenylenediamine Hcl	G.R.	5g x 2
Potassium Gluconate		25g x 4
Sodium Malonate	G.R.	25g x 2
L-Arginine Hydrochloride	G.R.	25g x 2
Casaminoic Acid	Dif	114g x 1
Sodium Ricenolate		25g x 1
Sodium Tartrate	G.R.	25g x 1
Sodium Benzoate	G.R.	25g x 1
Sodium Pyrubinate	E.P.	25g x 1
Sodium Succinate	G.R.	25g x 1
Pyridoxal Hydrochloride	G.R.	1g x 1
DL-Phenyl-L-Aranine	G.R.	25g x 5
Coomassie Brilliant Blue R250		25g x 2
YP-Nitrophenyl Phosphate		(5mg x100p) x1
Water Blue	G.R.	25g x 1
Alizarin Yellow GG	G.R.	25g x 1
Thymol Blue	G.R.	25g x 1
Methyl Red	G.R.	25g x 1
Chlorophenol Red	G.R.	1g x 3
Bromo Gresol Purple	G.R.	25g x 1
Bromo Thymol Blue	G.R.	25g x 1
Neutral Red	G.R.	24g x 1
Phenol Red	G.R.	25g x 1

Description of Goods		Quantity
Cresol Red	G.R.	25g x 1
Phenol Phthalein	G.R.	25g x 1
Litmus	G.R.	25g x 1
Fuchsin Acid	G.R.	25g x 1
Bromo Phenol Blue	G.R.	1g x 5
Bromo Cresol Green	G.R.	1g x 3
Resazurin	G.R.	1g x 3
Dextrun M.W.200,000/300,000		25g x 6
Dithiothreitol	G.R.	100mg x 4
Trimethoprime		5g x 1
Choline Chloride	G.R.	25g x 10
Vitamine A		1g x 1
" D2	G.R.	1g x 1
" E	G.R.	5g x 2
" K1	G.R.	1g x 1
Cholesterol	G.R.	25g x 1
Anforin Carrier PH.416		25ml x 2
" PH 5/8		25ml x 2
" PH 7/9		25ml x 2
" PH 8/9.5		25ml x 2
Methyl Green	M.	25g x 1
Sephadex G-25		100g x 5
" G-50 Fine		100g x 5
" G-75		100g x 2
" G-75 Super Fine		100g x 2
" G-100		100g x 1
" G-50 Super Fine		100g x 5
Ficol Pack		(100ml x 6) x 2
L-Lysine Hcl	G.R.	25g x 6
5-Aminosalicyric Acid	G.R.	25g x 6
Hormbal		100g x 1
Toluidine Blue O	G.R.	25g x 1
F.I.T.C.	E.O.C.	1g x 1
Dithiothreitol	G.R.	1 L x 1
Lecithin from Egg		25g x 1

Description of Goods		Quantity
Sodium Acelate	G.R.	500g x 3
Sodium Tungstate	G.R.	500g x 1
Saccharose	G.R.	500g x 14
Sodium Arginate		500g x 1
Tris-Hydroxymethylaminomethane	G.R.	500g x 1
Glucose	G.R.	500g x 40
Calcium Chloride	G.R.	500g x 1
Calcium Carbonate	G.R.	500g x 5
Sodium Phosphate Dibasic	G.R.	500g x 5
Potassium Phosphate Monobasic	G.R.	500g x 5
Potassium Phosphate Dibasic	G.R.	500g x 5
Sodium Carbonate	G.R.	500g x 2
Albumin EGG Plake	E.P.	500g x 2
Polyvinylpyrrolidinone K-30		500g x 10
" K-90		500g x 5
Magnesium Sulfate	G.R.	500g x 1
Phosphotungstic Acid	G.R.	500g x 1
Sodium Citrate	G.R.	500g x 1
Sodium Carbonate, Anhyd	G.R.	500g x 1
Sodium Bicarbonate	G.R.	500g x 1
Diethanolamine	G.R.	500g x 2
Diethanolamine	G.R.	500g x 2
Monoethanolamine	G.R.	500g x 2
Dimethyl Sulfoxide		500g x 1
Silica Gel Blue, Mid.		14kg x 9

Description of Goods		Quantity
Nitric Acid 1.38	G.R.	500g x 5
TOTAL: Twelve (12) W. Cases		25g x 82, 25ml x 18, 100g x 25, 500ml x 2, 1g x 23, 5mg x100, 250ml x 26, 5g x 5, 114g x 1, 100mg x 4, 14kg x 9, 100ml x 17, 250g x 1
	F.O.B. YOKOHAMA	¥1,315,530
	Ocean Freight	69,886
	Insurance Premium	8,893
	C.I.F. RANGOON	¥1,394,309

Description of Goods	Quantity
SPARE PARTS FOR MEDICAL EQUIPMENT	
Spare parts for High Pressure Autoclave Model S-90N 1. Silicon Gasket ...8 pcs.	1 set
Spare parts for High Pressure Autoclave Model SD-30N 1. Heating element, 2 KW ...2 pcs. 2. Pressure control switch ...2 pcs. 3. Silicon gasket ...8 pcs.	1 set
Spare parts for Automatic Tissue Processor, Model RH-12E 1. Motor, SH-1021 2. Thermoregulator for paraffine bath ...1 pc.	1 set
Spare parts for Refrigerated Centrifuge Model RS-20-2 1. Poly tube, 300ml. ...24 pcs. 2. - ditto -, but 50 ml ...60 pcs. 3. - ditto -, but 300 ml ...20 pcs. 4. - ditto -, but 10 ml ...80 pcs. 5. Glass tube, 15 ml ...48 pcs. 6. Carbon brush ... 4 pcs. 7. Rotor, No. 6N ... 2 sets 8. Rotor, No. 8N ... 2 sets 9. Rubber cushion ...72 pcs. 10. Poly tube, 10 ml ...24 pcs. 11. - ditto -, but 50 ml ...24 pcs.	1 set
Spare parts for Disc Type Electrophresis Apparatus, Model SJ-1061 1. Electrophoresis column ...62 pcs. 2. Disting column ...62 pcs. 3. Bar ... 1 pc. 4. Packing for electrophresis ...42 pcs. 5. Packing for disting ...42 pcs. 6. Timer ... 1 pc.	1 set
Spare parts for Freesing Dryer Model FD-5 1. Ampule Adaptor, No.75934-01... 1 pack pack of 10 pcs. 2. Vacuum oil ... 2 gallons 3. Gasket top cover ... 2 pcs. 4. Rubber tube, No. 76324 ... 2 pcs.	11 set

Description of Goods	Quantity
5. Rubber tube, No. 76244 ... 2 pcs. 6. Thermometer, No. 19509 ... 1 pc. 7. Compressure, No. 14685 ... 1 pc.	
Spare parts for Microscope Model BHA-534-SW	1 set
1. Objective Lens, 100x ... 1 pc.	
Spare parts for Microtome Model LR75-D	1 set
1. Knife handle ... 3 pcs. 2. Leather strap ... 6 pcs. 3. Microtome knife, 17 cm ... 6 pcs. 4. Microtome oil, 500 cc ... 6 pcs.	
Spare parts for Ultraviolet Absorbent Monitor, Model SJ-1541	1 set
1. Recording paper ...20 rolls	
Spare parts for Electrophoresis Apparatus Model AE-2B	1 set
1. Water bath ... 4 pcs. 2. Line viewer ... 1 pc. 3. Gel cutter set ... 2 sets	
Spare parts for Low Speed Refrigerated Centrifuge, Model CD-100R	1 set
1. Compressure ... 3 pcs. 2. Evapolator ... 3 pcs. 3. Thermoregulator ... 3 pcs. 4. Capillary Tube ... 5 vials 5. Carbon brush ...20 pcs. 6. Timer ... 2 pcs.	

Description of Goods	Quantity
MEDICAL INSTRUMENTS	
Isolation Tape, red	4 pcs.
- ditto -, but blue	4 pcs.
- ditto -, but green	4 pcs.
Fluorescent Lamp, FL-4W	6 pcs.
Blade, pack of 10 pcs.	10 packs
Dissecting Instrument Set Cat. No. 5-17201	1 set
Dressing Forceps, 11 cm long	4 pcs.
Binding agent, 50 grm	12 pcs.
Objective Lens, 4x	1 pc.
- ditto -, but 10x	1 pc.
- ditto -, but 20x	1 pc.
- ditto -, but 100x	1 pc.
Filtering Apparatus Model UHP-150 complete with 100 pcs. of filter papers	1 set
Micropipette, 20 - 200 ul. Cat. No. P-200	1 pc.
Tip for above box of 1000 pcs.	1 box
Film Developer, 0.6 liters	20 tins
- ditto -, but 4 liters	40 tins
- ditto -, but 8 liters	20 tins
Fixer, 2 liters	40 tins
Black and White Paper	
a) WP-FW-2, 13x18cm pack of 250 pcs.	10 boxes
b) WP-FW-4, " " "	10 boxes
c) WP-FW-2, 20.3x25cm " of 100 pcs.	10 boxes
d) WP-FW-4, " " "	10 boxes
Driwel, 2 liters	3 pcs.

Description of Goods	Quantity
Slide Film, 36Ex.	3 pcs.
Slide Paper Mount, for 25 pcs.	40 pcs.
Film, 35 mm	2 pcs.
Film Clip, box of 12 pcs.	10 boxes
Belt for Auto Print Dryer	2 pcs.
Pilot Lamp, 100 V.	12 pcs.
Flow-Cell, Cat. No. 12	1 pc.
- ditto -, but Cat. No. 52	1 pc.
Filter Paper, box of 10 pcs., 25 mm Cat. No. XM-50	1 box
- ditto -, but 43 mm, Cat. No. XM-50	1 box
- ditto -, but 25 mm, Cat. No. XM-100	1 box
- ditto -, but 43 mm	1 box
- ditto -, but 25 mm, Cat. No. XM-300	1 box
- ditto -, but 43 mm	1 box
Developing Tank, s.s.	2 pcs.
Developing Tray, plastic Size: 8" x 12"	6 pcs.
Spare Bulb for Enlarging Machine	4 pcs.
Dark Room Timer, 60 min.	2 pcs.
Dark Room Ventilator with Stepdown Transformer	1 pc.
Metal Enlarging Easel	1 pc.
Developing Tank, plastic	2 pcs.
Film, 5.9 x 18.3 cm, box of 100 pcs. for No. FG-JEM-7A	3 boxes
Microtiter Plate, box of 100 pcs. Cat. No. 76-212-05	1 box
Pipette, 5" long box of 250 pcs.	5 boxes

Description of Goods	Quantity
Pipette, 9" long box of 250 pcs.	5 boxes
Test Tube Basket, square, 10 cm	12 pcs.
- ditto -, but 15 cm	12 pcs.
- ditto -, but 20 cm	6 pcs.
Stainless Steel Bucket 36 x 36 cm	5 pcs.
Stainless Steel Beaker, 500 cc	3 pcs.
- ditto -, but 1,000 cc	3 pcs.
- ditto -, but 2,000 cc	3 pcs.
Homogenizer complete with standard accessories, for use on 100V, 50Hz, A.C. Model AM-9	1 set
Graduated Cylinder, 500 ml	6 pcs.
- ditto -, but 1,000 ml	6 pcs.
Test Tube, 18 x 165 mm	1,000pcs.
Petri Dish, 95 mm diam.	30 doz.
Funnel, 55 mm diam.	50 pcs.
- ditto -, but 75 mm diam.	50 pcs.
- ditto -, but 160 mm diam.	10 pcs.
Spatula, 15 cm long	10 pcs.
Stop Watch	5 pcs.
Stainless Steel Tray Size: 8" x 5" x 2"	6 pcs.
Erlenmeyer Flask, 30 ml	24 pcs.
- ditto -, but 50 ml	24 pcs.
- ditto -, but 250 ml	48 pcs.
- ditto -, but 100 ml	48 pcs.
Graduated Cylinder, 100 ml	24 pcs.
- ditto -, but 100 ml with stopper	12 pcs.

Description of Goods	Quantity
Graduated Cylinder, polyethylene 10 liters	6 pcs.
Disposable Test Tube, 12 x 75 mm box of 1,000 pcs.	3 boxes
Volumetric Flask, with stopper, 50 ml	30 pcs.
- ditto -, but 100 ml	30 pcs.
Graduated Cylinder, 50 ml	6 pcs.
- ditto -, but polyethylene, 1,000 ml	12 pcs.
Micropipette, 20 ul	2 pcs.
- ditto -, but 50 ul	2 pcs.
- ditto -, but 100 ul	2 pcs.
- ditto -, but 200 ul	2 pcs.
- ditto -, but 1 ml	2 pcs.
Tips for micropipette, box of 1,000 pcs.	1 box
Safety Pipetter, rubber, 60 ml	1 pc.
White Gold Crucible, 20 ml 32 ø x 34 (H) mm	2 pcs.
Test Tube, plastic Size: 12 x 75 mm, box of 1,000 pcs.	1 box
- ditto -, but 10 x 75 mm	1 box
Test Tube, glass, 12 x 75 mm	1,000 pcs.
- ditto -, but 10 x 75 mm	1,000 pcs.
Capillary Tube, heparinized	1,000 pcs.
Capillary Tube, plain	1,000 pcs.
Micropipette, 2 - 10 ul, No. 3001	1 pc.
Tip for above	1 box
- ditto - , but 10 - 50 ul Cat. No. 3002	1 pc.
Tips for above	1 box
Polyethylene Bottle, with nozzle 20 liters	12 pcs.

Description of Goods	Quantity
Polyethylene Bottle, with nozzle 10 liters	12 pcs.
- ditto -, but 5 liters	12 pcs.
Ampule, 1.5 ml	10 gross
Homogenizer, glass, 7 ml	6 pcs.
- ditto -, but 15 ml	6 pcs.
Petri Dish, polyethylene, 60 x 15 mm	60 pcs.
Culture Bottle, polyethylene, 125 x 16 mm	500 pcs.
Culture Bottle, glass, 50 ml	60 pcs.
- ditto -, but 300 ml	30 pcs.
Reagent Bottle, clear, 1 liter	2 pcs.
- ditto -, but 2 liters	2 pcs.
Volumetric Pipette, 10 ml	60 pcs.
- ditto -, but 5 ml	60 pcs.
- ditto -, but 2 ml	60 pcs.
Vial, with screw cap, 45 x 15 mm	1,000 pcs.
- ditto -, but 55 x 19 mm	1,000 pcs.
Micropipette, 2 - 10 ul with 960 pcs. of tips Cat. No. 3001	30 pcs.
- ditto -, but 10 - 50 ul with 500 pcs. of tips Cat. No. FP-11	30 pcs.
- ditto -, but 50 - 250 ul Cat. No. FP-12	30 pcs.
- ditto -, but 250 - 1,000 ul Cat No. FP-13	30 pcs.
- ditto -, but 1,000 - 2,000 ul with 200 pcs. of tips, Cat. No. FP-14	30 pcs.
Charcoal Activated Powder, 500 grm.	200 pcs.

Description of Goods	Quantity
Desiccator, large, Cat. No. LD-1	3 pcs.
Disposable Syringe, 1 cc with needle, box of 100 pcs.	60 boxes
- ditto -, but 5 cc	10 boxes
- ditto -, but 10 cc	10 boxes
- ditto -, but 50 cc	100 boxes
Stainless Steel Column, 4 Ø	3 pcs.
- ditto -, but 3 Ø	3 pcs.
Liquid Nitrogen Tank, 35 liters Cat. No. Dalic-35	2 pcs.
- ditto -, but 10 liters Cat. No. Cric-10	2 pcs.

TOTAL: 9 cases 14 sets, 8,932 pcs., 159 boxes, 10 packs, 120 tins, 30 doz. & 10 gross

Shipping Charge	¥100,043
Ocean Freight	¥321,040
Insurance	¥88,513
C.I.F.	¥15,065,656
	¥14,556,060

Description of Goods		Quantity
Spare parts for Air Conditioning Device consisting of;		1 set
1. Indoor Unit	... 3 sets	
2. Outdoor Unit	... 3 sets	
3. Connecting Pipe	...10 pcs.	
4. Compressor	... 5 pcs.	
5. Thermostat	... 5 pcs.	
Spare parts for Low Temperature Room consisting of;		1 set
1. Dryer	... 1 pc.	
2. Drift Solenoid Wire	... 1 pc.	
3. Defrost Solenoid	... 1 pc.	
4. Side Glass	... 1 pc.	
5. Expansion Valve	... 1 pc.	
6. Three Way Valve	... 1 pc.	
7. High Pressure Control Valve	... 1 pc.	
8. Operating Valve	... 1 pc.	
9. Thermostat	... 1 pc.	
10. Pressure Controller	... 1 pc.	
Spare parts for Low Temperature Room consisting of;		1 set
1. Dryer	... 1 pc.	
2. Drift Solenoid Wire	... 1 pc.	
3. Defrost Solenoid	... 1 pc.	
4. Side Glass	... 1 pc.	
5. Expansion Valve	... 1 pc.	
6. Three Way Valve	... 2 pcs.	
7. High Pressure Control Valve	... 1 pc.	
8. Operating Valve	... 3 pcs.	
9. Thermostat	... 1 pc.	
10. Pressure Controller	... 1 pc.	
11. Dryer	... 1 pc.	
Spare parts for Centrifugal Pump consisting of;		1 set
1. Packing	...20 pcs.	
2. Bolt	...20 pcs.	
3. Nut	...20 pcs.	
4. Water Stinger	...20 pcs.	
5. Packing	...20 pcs.	
6. O-Ring	...20 pcs.	
7. Liner Ring	...20 pcs.	
8. Impeller	... 5 pcs.	
9. Ball Bearing	...20 pcs.	
10. Liner Ring	...20 pcs.	
11. Coupling Bush	...20 pcs.	
12. Pump without motor	... 3 pcs.	

Description of Goods	Quantity
Spare parts for Constant Flow Pump Model BXD-23D consisting of; 1. Board ...8 pcs. 2. Piston Plate ...5 pcs. 3. Needle Bearing ...5 pcs. 4. Check Ball ...5 pcs. 5. Check Valve ...5 pcs.	1 set
Spare parts for Roller Filter 1. Main Timer ...1 pc. Cat. No. ATM-2NE 65H 2. Pressure Gauge ...1 pc. 3. Filter ...3 pcs. 4. Filter Media ...1 pc. 5. Relay ...1 pc.	1 set
Commuter, Model 3A for use on 400V, 50Hz, three-phase	1 set
Spare parts for Auto-Still Model WAR-30 consisting of; 1. Thermostat ...1 pc. part no. 253143-229 2. Filter ...2 pcs. part no. 253143-523 3. Resin Cartridge ...2 pcs. part no. 253143-616	1 set
Spare parts for Auto-Still Model WAR-560 consisting of; 1. Thermostat for Biler ...2 pcs. part no. 253104-330 2. Thermostat for condenser ...2 pcs. part no. 253104-516 3. Heater ...4 pcs. part no. 263104-311 4. Filter for raw water ...4 pcs. part no. 253104-521 5. Filter for pure water ...4 pcs. part no. 253104-519 6. Housing ...4 pcs. part no. 253104-516	1 set

Description of Goods		Quantity
Spare parts for Clean Bench Model PCV-841AL consisting of;		1 set
1. Hepa-Filter	... 6 pcs.	
2. Pre-Filter	... 6 pcs.	
Spare parts for Clean Bench Model CCV-1311 consisting of;		1 set
1. Hepa-Filter	... 6 pcs.	
Spare parts for Clean Bench Model CCV-811		1 set
1. Hepa-Filter	... 6 pcs.	
Spare parts for Cryostat Model AC2W consisting of;		1 set
1. Disposable Knife	... 3 pcs.	
2. Belt	... 2 pcs.	
3. Acryl Board	... 1 pc.	
4. Oil	... 3 pcs.	
Spare parts for Low Temperature Refrigerator Model SRF-910 consisting of;		1 set
1. Specimen Box, with cover	... 6 pcs.	
Spare parts for Gas Chromatographic Equipment, Model 163 consisting of;		1 set
1. Recording Paper	...51 rolls	
2. Micro Syringe, 1 ul	... 6 pcs.	
3. Micro Syringe, 10 ul	... 6 pcs.	
4. Pure Water Apparatus	... 1 pc.	
5. Empty Gas Cylinder	... 2 pcs.	
Spare parts for Spectrophotometer Model 100-50 consisting of;		1 set
1. Glass Cell, set of 2 pcs.	... 4 sets	
2. Filter, graduated	... 3 pcs.	
3. Cell Holder	... 3 pcs.	
4. Micro Cell Attachment	... 3 sets	
5. Quartz Micro Cell	... 4 pcs.	

Description of Goods	Quantity
Spare parts for Spectrophotometer Model 200-20 consisting of; 1. Micro Cell Attachment ... 1 set	1 set
Spare parts for Infrared Spectrophotometer Model 260-50 consisting of; 1. Liquid Cell, IRL-2 ... 1 set	1 set
Spare parts for Ultra Low Temperature Refrigerator Model MDF-390AT consisting of; 1. Recording Paper, box of 100 pcs. ... 3 boxes 2. Specimen Box, with cover size: 350 x 250 x 65 mm ...75 pcs. 3. Black Rubber Stopper ... 4 pcs.	1 set
Spare parts for Anaerobic Culture Jar Model JK-1 consisting of; 1. Empty Gas Cylinder with gauge and trolley ... 1 set 2. Catalyzer ... 2 tins 3. Glass Jar ... 6 pcs.	1 set

TOTAL: 10 wooden cases	20 sets	¥15,743,940
	Shipping Charge	¥156,866
	Ocean Freight	¥539,859
	Insurance	¥105,487
	C.I.F.	¥16,546,152

Description of Goods	Quantity
AIR CONDITIONER Spare Parts for Air Conditioning Device consiting of: 1. a-Indoor Unit for SRK-45K2 ... 8 sets b-Outdoor Unit for SRK-45K2 ... 8 sets 2. a-Indoor Unit for SRK-45K2 ... 8 sets b-Outdoor Unit for SRK-45K2 ... 8 sets 3. Freezing Mixture, R-22 (20 kgs Cylinders) (20 kgs Cylinders) ...20 pcs. 4. Connecting Pipe ...24 pcs.	1 lot

TOTAL:	1 lot	¥5,200,000
	Shipping Charge	¥170,900
	Ocean Freight	¥402,833
	Insurance	¥37,045
	C.I.F.	¥5,810,778

Description of Goods	Quantity
JEM-100S Electron Microscope	1 set
Special Attachments EM-SPI Field Position Indicator EM-A25NP Goass Plate Exposure Device EM-AVP 5 Vibration-Proof Mount JEE-4X Vacuum Evaporator JUM-7 Ultra-Microtome SVC-2025 Automatic Voltage Regulator HX-50 Cooling Water Circulator 2SP 2 Years use spare parts	1 set

TOTAL: Two (2) Cases only		
	2 sets	¥25,000,000
	Shipping Charge	¥108,704
	Ocean Freight	¥271,916
	Insurance Prem.	¥134,597
	C.I.F. RANGOON	¥25,515,217

Description of Goods		Quantity
Stop-Watch, Digital, 1/10 "SEIKO" TYJ016		2 pcs.
Hair-Dryer Model HD-1651 "HITACHI"		1 set
"IKA-ULTRA-TURRAX" Dispensers		1 unit
Contents of:		
Drive motor T 18/10	1	
Thyristor speed controller TR 50	1	
Laboratory stand R 1821	1	
Boss head R 181	1	
Clamp support RH 1	1	
Screw cap B 29	1	
Rubber cap	3	
Shaft 8N	1	
Shaft 10N	1	
Shaft 18KG	1	
Dehumidifier Model RD-900L "HITACHI"		2 sets
Pressure Pump XX 61,000.00 (115V 60Hz) "MILLIPORE"		1 set
"TABAI" PLATINOUS HUMIDOR Model PH-3E Temperature & Humidity Chambers Complete with standard accessories and extra spare parts (200V AC 3-phase 50Hz)		1 set
Pipettes Multi-channel "TITERTEK"		
77-858-00.5-50		12 pcs.
77-888-00.5-50		12 "
77-896-05 4-ch.(Tips)		500 "
Pipettes Multi-channel "TITERTEK"		
77-858-00 with tips (1 box)		2 sets
DIAFLO Ultra Filteration Membrames, 10's		
ϕ62 XM-100A		6 boxes
ϕ62 XM-50		6 "
ϕ62 PM-10		6 "
S.S. Reservoirs RS-12		1 box
Balance for Rabbit T-703A "TOKIWA"		2 sets
- do - for Rat T-701B "		2 "
- do - for Mouse T-701A "		2 "
Counter Balance, 4 kgs T-702B "		1 set

Description of Goods	Quantity
U.V. Monitor "TOYO" UVICON Model UV-750L Complete with standard accessories and 10 rolls recording paper.	1 set
Safety Cabinet 5-row "IUCHI" No. 3-1038	1 set
"HITACHI" Double-Beam Spectrophotometer Model 100-50 Complete with standard accessories, Micro Flow Cell Attachment, Recorder and recording papers (5 rolls) For operation on 100V, 50Hz, AC.	1 unit
"TOYO" U.V. Filter, ϕ150mm 5's	1 box
"TAIYO" Unit-type Constant Temperature Water Bath Model C-600 with Circle Shaker and Plastic Bath For operation on 100V, 50Hz, AC.	1 unit
Descicators, Electric Parfection Descicator Auto Dry System "IUCHI" Model O-H For 100V, 50Hz, AC.	1 set
"CHYO" Electronic Balance Model PD2-3000 Capacity 300g/0.1g For 100V, 50Hz, AC.	1 set
Freezing & Preservation Container using Liquid Nitrogen (-196°C) Model DALIC-35 Complete with 6-spare canister and 1-Measuring rod.	1 unit
"SAKURA" Ethylene Oxide Gas Sterilizer Model EOA-50 Inside chamber dimension: 310ϕ x 380mm For 100V, 50Hz, AC.	1 set
Slide Projector "ERMO" Model AS-3000A For 100V, 50Hz, AC. Complete with standard accessories.	1 set
Variable Volume Multichannel Pipette, 0.05~0.2ml 4-ch. 77-829-00	2 pcs.
- do - 8-ch. 77-859-00	2 "
- do - 12-ch. 77-889-00	2 "
Microtiter Plate, U type No. 220-24A 50's "DYNATECH"	10 boxes

Description of Goods	Quantity
Microtiter Plate, U type 96-well 100's with cover "LINBRO" No. 76-213-05	2 boxes
Microtiter Plate Cover, 100's "LINBRO" No. 76-205-05	1 box
Pipette Tip for Multichannel Pipettes 500's "TITERTEK"	2 boxes
"WHEATON" Vials with screw cap, 15 x 48mm 144's	10 boxes
- do - 17 x 63mm	10 boxes
"B.D. CORNWALL" Continuous Pipetting Outfit,	
1ml	4 sets
- do - 2ml	2 "
- do - 5ml	2 "
- do - 10ml	2 "
Stirrer Bar Set, 6.5 x 15mm 2 "R.K.I."	2 sets
6.5 x 20mm 2	
7.5 x 30mm 2	
10.0 x 40mm 2	
9.0 x 28mm 2	
15.0 x 52mm 2	
2.0 x 7mm 2	
3.0 x 10mm 2	
Rod for Stirrer Bar "	2 pcs.
Freeze Box V-6253A for 100 vials "	6 pcs.
Test Tube with Morton's Cap B-13 "	500 pcs.
Vistking Tubing 20ϕmm 24/32 100ft "SANKO"	2 pcs.
- do - 49ϕmm 1-8/7 50ft "	2 "
- do - 16ϕmm 20/32 50ft "	2 "
Petri's Dishes, plastic, 35 x 10mm 20's "FALCON"	25 boxes
Petri's Dishes, glass, 90 x 20mm "TYSTONE"	500 pcs.
Film, B&W NEOPAN SS 36EX	50 pcs.
Film, color F2 "	50 "
"PYREX" Flasks, glass, with screw-cap, 300ml 4985FK	72 pcs.

Description of Goods	Quantity
"BBL" Gas Pak 100 Systems "SANKO"	10 sets
" CO_2 Gas Bag No. 70304 "	131 boxes
" Gas Pak Catalist No. 70303 "	100 "
" Gas Pak Anaerobic Indicator No. 70504 "	100 "
"ERMA" Thoma's Haemacytometer JHS	1 set
Lab-Tek Tissue Culture Chamber, 8-chamber, 16 slides	100 pcs.
Labels, size: 25 x 40mm "IUCHI"	50 pcs.
Microscopic Slide Glass, S-1111 50's "MATSUNAMI"	30 boxes
Pipette Tips "PIPETTEMAN" P-20 or P-100 1,000's	10 boxes
Plate 96-well PS 100's No. IS-FB-96 clean "RKI"	2 boxes
Multi Dish, 24-well N-1483 75's "RKI"	10 boxes
Interchangeable Syringe, glass, 2, 5, 10ml "MS"	15 doz.
Disposable Syringe with needle, 1ml "MS" 100's	36 boxes
- do - 2ml " "	36 "
- do - (Tuberculin) 1ml " ""	36 "
"PYREX" Centrifugal Tube, 10ml 8080CTF	36 pcs.
- do - 50ml 8424CTF	36 "
Aluminium Foils, 25cm x 20M	50 pcs.
Aluminium Lunch Box, 10 x 18 x 7cm	12 pcs.
Ampoule LABCONCO No. 65/502 1ml 144's	5 boxes
- do - No. 65/505 2ml "	5 "
- do - No. 65/509 5ml "	5 "
Beaker Griffin Low-form, polypropilene 500ml "IUCHI"	12 pcs.
- do - 1,000ml "	12 "
- do - 2,000ml "	12 "
- do - 4,000ml "	4 "

Description of Goods	Quantity
"FEATHER" Operating Surgical Blades, 100's	5 boxes
Tissue Culture Flasks, disposable, 70ml with screw-cap 20's "RKI"	70 boxes
Tissue Culture Bottle, 200ml with screw cap, 48's No.222247	30 boxes (1,440pcs.)
Silicon Rubber Cap for Pipettes, 2ml	10 pcs.
- do - 5ml	10 "
- do - 10ml	10 "
Pipette Sterilizing Container, RKI 355 size: 75 x 65 x 395mm	5 pcs.
Test Tube Sterlizing Container, RKI 363 size: 480 x 200 x 250mm	5 pcs.
Microscopic Cover Glass, 22 x 22mm 1,00's "MATSUNAMI"	50 boxes
- do - 22 x 50mm 2,000's "	5 "
Visking Tubing 20/32 100ft "SANKO"	1 pc.
Disposable Petri's Dishes, 60mmφ N-1401 10's "RKI"	30 boxes
- do - 35mmφ N-14201 " "	30 "
- do - 35mmφ N-14201 " "	30 "
Stirring Bar, Teflon, 3.5cmφ "	6 pcs.
- do - 2.0cmφ "	6 "
Microscopic Slide Glass, S-1111 50's "MATSUNAMI"	60 boxes
- do - S-3313 50's "	60 boxes
"MILIPORE" Filters HAWP04700 100's	4 boxes
" Filter Holder SX 001300 12's	5 boxes
" Filters HAWP01300 100's	4 boxes
Disposable Syringe Needle, 27G 100's "MS"	20 boxes
- do - 25G " "	20 "
Holder with nichrome-wire	2 pcs.
Vacuum Pump RKI No.4111-A, Capacity: 100L/min.	1 set

Description of Goods	Quantity
Bunsen Supports, Ring(3) Clamp(1) RKI No. 7049	4 sets
Three-Way Stopcock, Luer-Lock Tip	8 pcs.
Pasteur Pipettes, capillary tubes, 9 inches, 250's "PYREX"	2 boxes
Centrifugal Tubes, 10ml RKI No. 8991	20 pcs.
- do - graduated, 10ml "	20 "
- do - round bottom, 50ml "	10 "
- do - 15ml graduated, with lim "PYREX"	50 "
Leiton's Tubes, square tube, RKI No.142 size: 15x15x50mm x 150mm long	50 pcs.
For High Speed Autoclave SD-30-ND Silicone Gasket "TOMY"	4 pcs.
Heating Element 2KW 100V "	2 pcs.
For Refrigerated Centrifuge RS-20-II Centrifuge tube glass 10ml "	50 pcs.
Centrifuge tube glass 50ml "	24 "
Centrifuge tube glass 15ml "	24 "
For CO_2 Incubator Model CO-2 TOKIWA Thermostat (Main)	1 set
Plastic Tubing 2m each " 6 x 12mm 4 x 7mm	2 pcs.
Lamp Neon "	2 pcs.
Plastic Drain Bottles Capacity 2L. "	2 pcs.
Rubber Gasket "	4 pcs.
For Refrigerated Centrifuge "TOMY" Model RS-20-II Compressor, hermetic type 3-phase AC 200V	1 set
Evaporator "	1 set
Temperature Control "	1 set

Description of Goods		Quantity
Capillary Tube and Dride Strainer	"TOMY"	3 sets
Carbon Brush	"	10 sets
Multimeter Model SP-10D	"SANWA"	2 sets
- do - Model YX390TR	"	2 sets
LAB Supplies Glassware etc. to be Pathology		
Sample stage for cryostat Model AC-2W	"TRIO"	6 pcs.
Sectioning Knife, 15cm	"	2 pcs.
Slide Holder (20 slides)	"MATSUYOSHI"	5 pcs.
Container for 50-slides	"	5 pcs.
Decapitating Apparatus Model CL-855, large	Clea	1 set
Ear Punch, 120mm Model -856	"	1 set
Forceps, 30cm long	Mysco	5 pcs.
Glove Kitchen	"	20 pcs.
Parasitology		
Lamp ultraviolet GL-30	"MATSUYOSHI"	2 pcs.
Lamp ultraviolet set 30W 100V GL-30W-A	"	2 sets
Microscopic Slide Cavity 76 x 26mm box of 50 pcs.	"	20 boxes
Microscopic Slide Cavity 76 x 40 mm box of 12	"	1 boxes

TOTAL:	2,414 pcs. 73 sets 4 units 15 doz. 1,032 boxes	¥23,315,570
Shipping Charge		¥159,378
Ocean Freight		¥448,392
Insurance Prem.		¥126,869
C.I.F. RANGOON		¥24,050,209

Description of Goods	Quantity
"OLYMPUS" FLUORESCENCE MICROSCOPE Model BHF-342 Trinocular head, High pressure mercury burner, Barrier filter attachment (BH-FA) Objectives: F10x, Ach.20x, SFL40x, SFL100x. Eyepieces : HEPBiWF10x (paired) Photo eyepieces: FK3.3x, FK5x. Complete with standard accessories and 2-spare mercury burner (USH-200W/2), 6-spare 30W tungsten bulb (LS-30). For operation on 100V AC.	1 set
"CALIFORNIA" LABORATORY PELLET MILLS Model CL Type 3 Dimensions: L45.5"x W34.25"x H63" Main Drive Motor: 2HP 1800RPM Feeder Drive : 1/4HP 1800RPM Feeder Hopper : .9514 Feed Control : Variable vibrating, electrically driven Feeder Screw : Constant speed, variable pitch Die Speeds : Replaceable sheaves for four die speeds Electrical : 3 phase 50 Hz 220 volt current. Complete with standard accessories and special accessories; 3 pcs. L-8 9 1/8" Dies, 3 pcs. L-8 9 1/2" Dies, 3 pcs. Belts, 6 pcs. Roller	1 unit
"OLYMPUS" INVERTED TISSUE CULTURE MICROSCOPE Model IMT-313 Trinocular head (BH-TR30) Objectives : Ach.4x, F-10x, LWD-C20x, LWD-CPL40x Eyepieces : BiCK-10x (paired) FK3.3x for photo. Complete with standard accessories. For operation on 100V AC.	1 set
"OLYMPUS" AUTOMATIC PHOTOMICROGRAPHIC SYSTEM CAMERA Model PM-10-35AD-2 Complete with standard accessories. For operation on 100V AC.	1 set

Description of Goods	Quantity
DYNADROP SR-I "SANKO JUNYAKU" 8-tip manifold 2-325-01	1 set
FOR AUTOSTILL WAR-30 "YAMATO" Thermostat EA 3-L 15 120°C 253143-229 Robert shaw	1 pc.
Reverse osmosis membrance SR-6 253143-523 Tokyo filtration	1 "
Ion-exchange resin cartridge type B-5Y 253143-616 Japan organo	2 pcs.
FOR AUTOSTILL WAR-560 "YAMATO" Thermostat (for boiler) 2455 RBu 103°C ON 253164-330 EL Wood	1 pc.
Thermostat (for condenser) CS-7 70°C 253104516 Fukushima dengyo	1 "
Heater 1.4KW AC 220V "YAMATO" 253104-311	4 pcs.
Filter for raw water, SWPPO 10-1 10u 253104-519	1 pc.
Filter for pure water H.C-40u 0.14u 253104-519	1 "
Reverse osmosis filter SRV-19 spiral type 253104-516	1 "
Ion exchange resin B-10Y IR-120B IRA-41 253104-801 Organo	2 pcs.
FOR INFRATED SPECTROPHTOMETER 260-50 "HITACHI"	
Chart paper A4 size 260-1305	2 pcs.
Chart paper double A4 size 260-1306	3 "
Polystyrene film (standard sample)	1 pc.
Ink red 50cc 963-5022	1 "
Chart paper 033-0010	20 pcs.
Kbr random crystal 100gm B142031	1 pc.
Record pen S250260	1 "
SPARE PARTS FOR ICE MAKING MACHINE Model SIM-60A "SANYO" Compressor, hermetic type (100V 50Hz.1Ø)	1 set

Description of Goods	Quantity
SPARE PARTS FOR REFRIGERATOR Model SR-493F "SANYO" Compressor, hermetic type (100V 50Hz.1Ø)	1 set
SPARE PARTS FOR ULTRA DEEP FREEZER Model MDF -390AT "SANYO" Temperature control	5 pcs.
Temperature recorder	5 "
Compressor, hermetic type (100V 50Hz.1Ø)	2 sets
SPARE PARTS FOR DEEP FREEZER Model SRF-910 "SANYO" Compressor, hermetic type (200V 50Hz.3Ø)	1 set
Temperature control	1 pc.
Thermometer with remote sensor	1 "
SPARE PARTS FOR FLAKE ICE MACHINE Model SIM-F-120 "SANYO" Bin thermostat	1 pc.
E-Thermostat	1 "
LABORATORY ANIMAL SUPPLY CENTRE Bio-sticker, "Aron-alpha A" Sankyo	10 pcs.

10 sets, 1 unit & 68 pcs.	¥10,204,430
Shipping Charge	¥44,436
Ocean Freight	¥119,203
Insurance Prem.	¥54,987
C.I.F. RANGOON	¥10,423,046

Description of Goods		Quantity
Blood Agar Base		7 pcs.
Tc Medium 199		2 "
Fetal Bovine Serum		2 "
Bovine Serum		2 "
Yersinia Enterocolitica Antisera		1 pc.
Shigella Antisera		1 "
Salmonella Antisera		1 "
Vibrio Parahaemolyticus Antisera		1 "
Vibrio Paranaemolyticus Antisera O-Grouping		1 "
Escherichia Coli Antisera		1 "
Vibrio Cholerae Antisera		1 "
Tridisk Cephaloridine	50 Sheets	3 pcs.
Tridisk Aminobenzyl Penicillin	50 Sheets	3 "
Cycloheximide	5 g	5 "
Sensitivity Disk Cepazolin	50 Sheets	3 "
Vancomycin	1 g	1 pc.
Trimethoprim	5 g	5 pcs.
Erythritol	25 g	1 pc.
Bacitracin	5 g	1 "
Colchicine	1 g	1 "
Cycloheximide	5 g	1 "
Nalidixic Acid	5 g	1 "
Novobiocin	1 g	5 pcs.
Polymixin B Sulfate		1 pc.
Trimethoprim	5 g	1 "
ε-(Hymotrypsinogen-A)	1 g	1 "
Coomassie Brilliant Bluer	25 g	1 "
Cytochrome-C		1 "
Eagle Mem	100 g	5 pcs.
Leibovitz's L-15 Medium		1 pc.
Calf Serum		5 pcs.
Calf Serum		5 "
Albumin Bovine	25 g	4 "
Bovine Albumin Fraction	50 g	5 "
Heparin Sodium Salt	5 g	3 "
Hanks Balanced Salt Solution		1 pc.

Description of Goods		Quantity
Tryptose Broth	100 g	1 pc.
Medium 199	100 g	2 pcs.
Phosphatase Alkaline		3 "
P-Nitorophenylphate Disodium Salt	25 g	5 "
Peroxidase from Horseradish	1 g	5 "
£-Chymotrypsin	1 g	3 "
Anti Mouse	1 gm	1 pc.
Anti Mouse	1 gA	1 "
Anti Mouse	1 gG	1 "
Diluent for Lyophilized Complement	1 ml	3 pcs.
5-Bromodeoxyuridine Anhyd Cryst	100 ml	5 "
Eagle Medium	500 ml	1 pc.
Fetal Bovine Serum		2 pcs.
Nicotinamide-Adenine Dinucleotide	1 g	1 pc.
Nicotinamide-Adenine Dinucleotide Phosphate		5 pcs.
P-Nitrophenylphosphate Disodium Salt	1 g	1 pc.
Tetrahydrolic Acid		1 "
Lecithin Egg		2 pcs.
Deoxyribonuclease I		1 pc.
L-£-Lysophosphatidylcholine		2 pcs.
Piruvate Kinase		1 pc.
Ribonuclease-A		1 "
Lactoperoxidase		5 pcs.
Peroxidase		1 pc.
dl-L-Tetrahydrofolicacid		3 pcs.
Dihydrofolic Acid		1 pc.
Albumin	1 g	1 "
Catalase	1 g	1 "
Dithiothreitol		1 "
Human	1 gG	1 "
Thrombin		20 pcs.
Lipopolysaccharide		2 "
Giemsas-Losung		3 "
Mit Tetrahelium		5 "
Nicotinamide-Adenine Dinucleotide Phosphate		5 "
Nicotinamide-Adenine Dinucleotide	1 g	5 "

Description of Goods		Quantity	
Xanihine Oxidase		5 pcs.	
Pyridoxal Phosphate Monohydrate	1 g	1 pc.	
Calcium Chloride, Hexahydrate	500 g	1 "	
Glutaldehyde	500 g	1 "	
N-Acetyl-L-Cysteine	25 g	2 pcs.	
Adenosine-5-Triphosphate Disodium Salt	1 g	1 pc.	
Z-Ketoglutaric Acid	25 g	1 "	
TOTAL:		198 pcs.	¥2,134,860