5．合同委員会の協議結果

5－1　経緯と概要

本調査団派遣に先立ち、合同委員会の協議結果としてミニッツに盛り込むべき内容等について、7月23日の国内委員会や調査団員による派遣前団内打合せ等を通じ十分検討し、調査団対処方針案を策定した。

本合同委員会は、平成2年8月3日午後14時より、国立アスンシオン大学保健科学研究所において友好裡に行われ、8月6日我が方仙道富士郎、先方Dr. Luis Berganzaアスンシオン大学長及びDr. Ricardo Moreno Azorero同大学保健科学研究所長との間で、ほぼ我が方案の通りでミニッツに署名を了した。（ミニッツの概要については第2章要約を参照）

なお、本合同委員会の席で、仙道団長及びDr. Ricardo Moreno Azorero所長により本プロジェクトの進捗状況及び今後の活動計画等につき説明がなされた。

附　属　資　料

① 協議議事録（ミニッツ）

② ＩＩＣＳシャガス病等寄生虫研究プロジェクト
パラグアイ側Ｃ／Ｐ研究報告要約

③ 寄生虫学・循環器学セミナーに係る調査団員講演原稿

④ パラグアイ原子力委員会作成のＩＩＣＳ放射性廃棄物管理調査報告及び処理に係るガイドライン

⑤ 平成２年度供与機材リスト

①　協議議事録（ミニッツ）

THE MINUTES OF DISCUSSIONS BETWEEN THE JAPANESE ADVISORY SURVEY TEAM AND THE AUTHORITIES CONCERNED OF THE GOVERNMENT OF THE REPUBLIC OF PARAGUAY ON THE JAPANESE TECHNICAL COOPERATION FOR THE RESEARCH PROJECT ON CHAGAS' DISEASE AND OTHER PARASITIC DISEASES

The Japanese Advisory Survey Team (hereinafter referred to as "the Team") organized by the Japan International Cooperation Agency (hereinafter referred to as "JICA") and headed by Prof. Dr. FUJIRO SENDO visited the Republic of Paraguay from July 28th to August 10th, 1990 for the purpose of making technical guidance and working out the details of the technical cooperation programme concerning the Research Project on Chagas' Disease and other Parasitic Diseases(hereinafter referred to as "the Project").

During its stay in the Republic of Paraguay, the Team observed the overall progress, exchanged views and had a series of discussions with the Paraguayan authorities concerned regarding the activities and implementation of the Project.

As a result of the discussions, the Team and Paraguayan authorities agreed upon the matters referred to in the document attached hereto.

Asuncion, August 6th, 1990

Prof. Dr. Fujiro Sendo
Leader,
Japanese Advisory Survey Team
Japan International
Cooperation Agency,
Japan

Dr. Luis Berganza
Rector,
National University of Asuncion,
The Republic of Paraguay

Dr. Ricardo Moreno Azorero
Director,
The Institute of Research for
Health Sciences,
The Republic of Paraguay

ATTACHED DOCUMENT

I . GENERAL REVIEW

This project started on March 4, 1988, for the purpose of establishing research activities upon Chagas' disease and other parasitic diseases, and thus contributing to the promotion of public health in the Republic of Paraguay as a five-year project. In order to achieve the purpose set above, Japanese technical cooperation was initiated in the fields of 1) Parasitology, 2) Pathology, 3) Immunology, and 4) Others mutually agreed upon as necessary. The Project was expected to be implemented in accordance with the Master Plan attached in the Record of Discussion and the Tentative Schedule of Implementation signed on March 4th, 1988.

Regarding to the project activity in the Japanese Fiscal Year (starting from April 1st, to March 31st, of the next year) 1989 (FY 1989) and the first half of FY 1990, JICA has dispatched 5 long-term experts and 19 short-term experts, and has accepted 6 Paraguayan counterpart personnel for technical training in Japan. And JICA has preceded to provide and will provide the machinery and equiupment necessary for the Project. On the other hand, the Paraguayan side has provided the facilities necessary for the Project and has secured the budgetary allocation and Paraguayan counterpart personnel required for smooth implementation of the Project.

Both sides reviewed the activities of the Project and the achievement made so far with regard to the implementation of the Project. Thus, based on the common recognition of the present state of the Project, both sides confirmed the continuous cooperation between the Japanese and Paraguayan governments for the further progress of the Project.

II. ACHIEVEMENT OF TENTATIVE SCHEDULE OF IMPLEMENTATION

The technical cooperation activities under the Project in FY 1989 and the first half of FY 1990 have been carried out as follows:

1. Dispatch of Japanese experts to the Project

a. Long-term experts (field, name, term)

1)	Team Leader	Dr. Masato Kawabata 1988. 4/12 - 1990. 4/ 9
2)	Coordinator	Ms. Masayo Kondo 1988. 5/24 - 1990. 5/21
3)	Parasitology	Dr. Tadashi Watanabe 1988. 10/21 - 1990. 3/20
4)	Team Leader	Dr. Makoto Sakamoto 1990. 3/24 - 1991. 3/23
5)	Coordinator	Ms. Yoko Akimoto 1990. 4/20 - 1992. 4/19

b. Short-term experts (field, name, term)

1)	Parasitology	Dr. Yoshihisa Hashiguchi 1989. 5/19 - 1989. 9/18
2)	Immunology	Dr. Susumu Nishinarita 1989. 6/ 1 - 1989. 8/31
3)	Immunology	Dr. Mitsuhiro Numata 1989. 6/ 1 - 1989. 8/31
4)	Pathology	Dr. Hitoshi Suzuki 1989. 8/ 2 - 1989. 9/ 1
5)	Engineering (Setup and Maintenance of the FPLC System)	Mr. Hiromichi Ogawa 1989. 11/24 - 1989. 12/ 3
6)	Architecture (Consulting and Supervision of the Laboratory Constrution matters)	Mr. Kisen Misawa 1989. 11/24 - 1989. 12/ 3
7)	Technical Cooperation	Mr. Koichi Noguchi 1989. 11/24 - 1989. 12/ 3
8)	Engineering (Mainttenance of the Millipore Water Distiller Apparatus)	Mr. Hiromasa Yamamoto 1990. 2/24 - 1990. 3/ 5
9)	Architecture (Consulting and Supervision of the Laboratory Constrution matters)	Mr. Kisen Misawa 1990. 2/24 - 1990. 3/ 5
10)	Parasitology	Dr. Tatsuyuki Mimori 1990. 4/16 - 1990. 9/28
11)	Clinical Laboratory	Mr. Tetsuo Miyashita 1990. 4/16 - 1990. 10/15
12)	Molecular Biology	Dr. Yoshio Ichinose 1990. 5/15 - 1990. 8/ 1

13) Architecture Mr. Kisen Misawa
(Consulting and Supervision of the Laboratory Constrution matters) 1990. 6/ 9 - 1990. 6/18

14) Radiology Dr. Akio Komatani
(Administration of Isotope Waste)
1990. 6/ 9 - 1990. 6/18

15) Equipment Mainttenance Dr. Fumihisa Sakuma
1990. 6/ 9 - 1990. 6/18

16) Parasitology Dr. Susumu Saito
1990. 7/16 - 1990. 10/15

17) Pathology Dr. Takeshi Shozawa
1990. 7/16 - 1990. 8/15

18) Parasitology Dr. Shozo Matsuo
1990. 7/28 - 1990. 8/ 8

19) Parasitology Dr. Hiroji Kanbara
1990. 7/28 - 1990. 8/ 8

2. Training of Paraguayan counterpart personnel in Japan
(field, name, term)

1) Molecular Biology Dr. Edgar Modesto Villagra Vera
1989. 6/20 - 1990. 9/20

2) Biochemistry Dra. Maria Elena Zorrilla Gadea
1989. 9/ 4 - 1990. 9/ 4

3) Entomology Dra. Gladys Antonieta Rojas De Arias
1989. 9/ 4 - 1989. 10/ 1

4) Inmunogenetica Dr. Ricardo Moreno Azorero
1990. 2/13 - 1990. 3/11

5) Immunology Dra. Estela Picagua
1990. 5/14 - 1991. 5/14

6) Cardiology Dr. Jorge Ernesto Martinez Espinola
1990. 5/14 - 1989. 5/14

3. Provision of Machinery and Equipment

Machinery, equipment and other materials (hereinafter referred to as "the Equipment") necessary for the implementation of the Project have been provided in FY 1989 and FY 1990.

The following is the list of main Equiupment provided to the Project.

FY 1989

1) Trinocular Microscope
2) -1. Inverted Research Microscope
-2. Automatic Photomicrographic App.
3) Inverted Microscope

4) Table Top Centrifuge
5) Clean Bench
6) Co2 Incubator
7) Universal Refrierated Centrifuge
8) Electrophoresis Apparatus
9) Hot Plate and Stirrer
10) Polariod Multi-Purpose Camara
11) -1. Multi-Mixer
    -2. Microplate Holder for Above
12) Tubing Roller Pump
13) Aseptic Box
14) -1. Heating Mantle
    -2. Glass Ware for Above
15) Eppendorf Pipette
16) Standard Tip for Above
17) Densitometer
18) Dissecting Instrument Set
19) Incinerator
20) Ultrasound Tomographic
21) Incubator, Digital Type
22) Incubator, Circulating Typa
23) Automatic Balance
24) UV Transilluminator
25) Bio-Dot Apparatus
26) Aspirator

FY 1990

1) Diagnostic Ultrasound Scanner
2) Endoscopic Equipment
   -1. OES Gastrointestinal Fiberscope GIF Type XP-20
   -2. OES Colonofiberscope CF Type 20
   -3. OES Halogen Light Source CLE-10
   -4. OES Gastrointestinal Fiberscope GIF Type P-20
   -5. Colonofiberscope CF Type 1T20I
   -6. 35mm Medical SLR Camara
   -7. OM Halogen Adapter A10-M1
3) Electrophoresis and Blotting Apparatus
   -1. DNA Sequencing Electrophoresis Unit
   -2. Vertical Slab Gel Electrophoresis System
   -3. Portable Electrophoresis Unit
   -4. Semi-Drive Lotter
   -5. Mini-Slab Gel Electrophoresis Unit
   -6. Power Supply
   -7. Destaining Shaker
4) Hibridization Oven
5) Pathology Equipment
   -1. Microtome Knife Sharper
   -2. Microtome Knife
   -3. Iwaoka's Strop
6) Binocular Microscope
7) Freezers
   -1. Ultra Low Temperature Freezer
   -2. Medical Freezer
8) Vacumm Freezer Dryer
9) Polarizing Binocular Microscope
10) Catheter Introducer

III. ACHIEVEMENT OF THE TECHNICAL COOPERATION IN EACH FIELDS

According to the Tentative Schedule of Implementation (TSI), we have acquired the following results and achievement:

1. Establishment of the clean room cell culture laboratory
2. Production of monoclonal antibodies
3. Formation of E-rosette by the lymphocyte of Cebus apella in search of animal models
4. Establishment of lines of T. cruzi isolated from Chagasic patients in Paraguay and their characterization
5. Immunological studies of T. cruzi-infected mice from the standpoint of cytokine production
6. Chromosomal studies on Chagas' vectors
7. Studies on humoral immune response in acute Chagasic patients by Western blotting
8. A pilot study in the search of a vaccine using a mouse model

Besides the TSI, we have done the following additional research programs:

1. HLA typing of the Chagasic patients (Epidemiological-immunological study)
2. Research on lymphocyte function of the Chagasic patients (Immnunological study)
3. Standardization of biochemical and immunological values in C. apella

Establishment of the clean room for cell culture

Owing to the delay of the new laboratory construction and the supply of equipment clean bench), we had to utilize the laboratory for cell culture set in the second floor of the IICS main building. For obtaining the super distilled water, Milli-Q system and Milli-RO system were set up. For a few months, the problems of supply for electricity were happening one after another, but at the moment, the systems are working enough for supplying the necessary super distilled water for the cell culture experiments.

Production of monoclonal antibodies against Trypanosoma cruzi

Monoclonal antibodies against T. cruzi were produced for diagnosis of Trypanosoma infection and for finding out the possibility of vaccine development.

Six clones were obtainied by the cell fusion with the mouse spleen cells infected by T. cruzi and murine myeloma cells. (Dillution method was

employed for the cloning.)

The immunoglobulin isotypes of antibodies produced by the each clone are;

2 clones of Ig]g 2b specific for low molocule of protein of T. cruzi antigen, and 4 clones of IgM specific for low to high molecule protein of T. cruzi antigens respectively.

As an antigen, y-epimastigote strain is utilized in this experiment and the cross-reactivity of other strains or the cross-reactivities for Trypamastigotes and amastigotes are under discussion.

Inhibition of E rosette formation of Cebus apella lymphocytes with monoclonal antibodies CD2

E rosette formation of Cebus apella lymphocytes is well known and its confirmed by ourseleves. By the treatment with FITC labeled monoclonal antibody CD2, 50-80% of monkey lymphosyte and 50-60% of human lymphocyte are estimated for positive respectively. We have studied the E rosette formation inhibition with monoclonal antibody CD2 to know the cross-reactivity of CD2 receptor on between human lymphocytes and monkey lymphocytes.

It is determined that there is common sheep blood cell receptor structures on the surface of both lymphocyte (Cebus apella) and human lymphocytes resulting from the inhibition assay of E rosette formation by monoclonal antibody CD2.

Establishment of lines of T. cruzi isolated from Chagasic patients in Paraguay and their characterization

Heterogencity of T. cruzi has been demonstrated by many workers.

Inasmuch as that the main final goal in this Project is to control Chagas' disease in Paraguay, it seems necessary to clarify the nature of T. cruzi in Paraguay. We, therefore, tried to isolated T. cruzi from Chagasic patients in Paraguay. With an improvement of method, we established many lines up to now. With DNA analysis of kynetoplast of established T. cruzi lines, it has been found that blotting pattern from one established line differed completely from those of the other already established lines, meaning that we established a novel line of T. cruzi.

Immunological studies of T. cruzi-infected mice from the standpoint of cytokine production

In order to perform immunological therapy of Chagas' disease, it seems necessary to understand immunological status of T. cruzi infected

hosts. As a preliminary work to this end, we examined production of tumor necrosis factor (INF), which is now recognized as an important cytokine in a serial respose from inflammation to immunity, in T. cruzi infected mice. We found that intraperitoneal macrophages in acute phage of infection produced a large amount of TNF, and thereafter its production gradually decreased. This result seems to suggest that TNF may play an important role in acute phase response to T. cruzi.

Chromosomal studies on Chagas' vectors

In order to establish : 1. The chromosomic information of species R. neglectus and T. intestans and 2. Analize intraespecifically the T. infestans in population from different geographical area. This work strats with the purpose of using it as a background for future researchs concerning the biology and control mechanism of the T. infestans.

Twenty insects were processed and 250 slides were perform. At least 10 mitotic metaphases were observed and photographied. Both species present a kariotype of 22 chromosome (2N:22). In R. neglectus in some cases the number of them varied between 23 and 27. The X chromosome with its medial constriction has a tendency to separate itself in this region during prophase and early metaphase in mitosis.

T. infestans from Brazilian and Argentinian strain show a polymorphism in variation of C bands. Three cytotypes had been identified for Brazilian strain. Paraguayan strain from three rural localities had presented similar cytotypes as described originally for Brazilian and Argentinian strains.

This finding can have a great interest in the study of the evolution of this group of insects and the knowledge of alternative mechanism to control Chagas' disease.

Studies on humoral immune response in acute Chagasic patients by Western blotting

It seems important to know what kind of epitopes in T. cruzi antigen is recognized in Chagas' disease for final goal of vaccine establishment. We checked the Western blotting pattern using sera serially obtained from acute Chagasic patients as antibody sources. Interestingly, it was found that epitopes recognized by patients sera obtained from different time after the infection differed each other. T. cruzi antigens of certain molecular weight were recognized at early phage of infection, but they were hardly recognized with sera obtained at relatively later phase of infection. This result suggests that antigen recognition by antibodies produced in Chagasic patients is a quite complexed phenomenon.

A pilot study in the search of a vaccine using a mouse model

The starting point in vaccine experiment should be to elucidate whether T. cruzi antigen is antigenic to the host or not. To this end, we immunized mice with UV irradiated whole tripomastigotes or each fraction of sonicated tripomastigotes obtained from sucrose gradient centrifugation. Thereafter, mice were challenged with live tripomastigote, and observed their survival. The mice immunized with UV irradiated whole tripomastigotes or a fraction obtained from the layer of 40% sucrose survived the challenge in constrast to other groups. This result indicates that T. cruzi antigen is immunogenic for induction of protective immunity. It was further suggested that antigens obtained from the layer of 40% sucrose may be a candidate as a source of vaccine.

HLA typing in the Chagas' disease patients

Chronic Chagas' disease of human is classified into two distinctive types, i.e., cardiomegaly type and megacolon/mega esophagus type. It is speculated that the differences of T. cruzi strain may cause a different clinical course in human.
Because the expression of HLA is strongly related to the susceptibility of human diseases, we have examined an HLA antigen typing in the group of Chagas' disease patients.

20 patients are checked on both HLA class I (HLA-A, B, C) and class II (HLA-DR, DQ). Between the cardiomegaly group in high frequency. We concluded that this has some meaning on the pathogenesis of cardiomegaly in this disease.

Research on lymphocyte function of the Chagas' disease patients

It is known that lymphocyte function of T cells play an important role in the prevention of infection and in pathogenesis in Chagas' disease. It was necessary to establish the assay system for the blastogenesis of T cells against T. cruzi antigen or mitogens.

Therefore, we have studied on the blastogenesis of T cells from Chagas' disease patients for T. cruzi antigens preliminary to determine the optimal condition for the incubation periods, concentration of antigen and its specificity.

We concluded that the optimal antigen concentration is 5μg/ml, culture period is 7 days. Blastic transformation of T cells did not appear in lymphocytes from the healthy group but appeared in T cells from each Chagas' disease patient at the different reaction level. In series of experiments, the epimastigote antigen was used, and the blast formation

was expressed with blastgenesis score, but not with 3H-thymidine incorporation, and there are considered to be solved in the future time.

Standardization of biochemical and immunological values in C. apella

One of the main projects in this research is to use Cebus apella for characterization of T. cruzi infected hosts from immunological, biochemical, and other view points. Thus, as a first step, we examined serum enzymes, each class of immunoglobulins and complement titers of Cebus apella which are looked like normal by macroscopical obsevations. To our surprise, in some monkeies, although not so many, the values of each observed items were in abnormal range. And standerd deviation of each item was rather broad.

These results gave us a suggestion that Cebus apella looked like normal is not in a completely healty condition, and that we should be quite carefull for selection monkeies at the begining of T. cruzi infection experiment.

Anyhow, these results seem to give us very important basic information for monkey study.

IV. SUMMARY OF THE DISCUSSIONS

1. Research activities

The Paraguayan side explained to the Japanese side that the study of Cebus apella monkey as an animal model for Chagas' disease is of high interest and considered to be one of the most important areas in the Project. Therefore, they requested that a higher priority may be given to it in the future

The Japanese side mentioned that the IICS had established the fundamental studies on primates at the time of the Tentative Schedule of Implementation of the Project. The Japanese side recommended that a continuous supply of monkey may be secured for the sake of the success of the Project.

The Paraguayan side proposed the necessity to reinforce the study on clinical aspects of Chagas' disease in the coming period of the Project. The Japanese side mentioned that clinical studies is an area predetermined to be performed in the later part of the Project. The Japanese side mentioned that these studies are considered to be one of the most important areas of research in the later period of the Project.

The Paraguayan side suggested that biochemical studies should be strengthened for the purification of T. cruzi antigens. The Japanese side replied that this subject might be critical for the final purpose of this Project.

The Japanese side, therefore, agreed with the proposal of the Paraguayan counterpart.

2. Supply of reagents

The Paraguayan side mentioned that there have been some troubles with the supply of reagents. The Japanese side answered that this would be improved as long as the fault falls on the Japanese side. Furthermore, the Japanese side suggested that it could be necessary to find a way of getting those items around the local area. The Paraguayan side expressed their agreement on this problem.

3. Dispatch of Japanese experts to Paraguay

The Paraguayan side requested that the Japanese side to informe early in advence on the specialties of the Japanese experts who would be coming to Paraguay every time. In respose to the above statement, the Japanese side expressed their willingness to make all efforts in this regard.

4. Technical training of the Paraguayan counterpart

The Paraguayan side explained that the award of scholarships is of paramount importance in order to guarantee the continuity of long term activities.

The Paraguayan side asked about the possibility of getting financial

support for training in neighboring countries such as Brazil and Argentina.

In respose to the above statement, the Japanese side explained the system of training in a third country. The counterpart at IICS may utilize this system.

5. Assignment of Counterpart personnel

The Japanese side suggested that it is necessary to assign more full-time counterparts to the Project, in order to ensure smoothness in the development of the planned research activities. The Paraguayan side expressed their willingness to make all possible effort in order to overcome this problem.

V. TENTATIVE SCHEDULE OF IMPLEMENTATION

According to the present state of progress and other conditions of the Project, both sides jointly formulated workable Annual Implementation Plan.

The outline of the Annual Implementation Plan is as follows:

1. Japanese side

1.1. Dispatch of Japanese experts to the Project (the latter half of FY 1990 and FY 1991)

a. Long-term experts

1) Team Leader
2) Coordinator
3) Parasitology

b. Short-term experts

1) Pathology
2) Epidemiology
3) Maintenance of the equipment
4) Biochemstry
5) Epidemilolgy
6) Cardiology & gastroenterology
7) Others according to the mutual aggreement if necessary

1.2. Training of Paraguayan counterpart personnel in Japan (the latter half of FY 1989 and FY 1990)

1) Electron Microscopy and Inmuno Histo Chemistry
Dra. Elena Satiko Kasamatu
1990, 8/20 - 1990, 11/19

2) Genetics
Dra. Marta Ascurra De Duarte
1991, 2/ 5 - 1991, 6/ 4

3)

4)

5)

6)

1.3. Provision of the Equipment

Equipments necessary for the Project will be provided within the limit of the budgetary allocation of the Japanese side.

1.4. Both sides agreed that the Project activities such as experts dispatch, counterpart training and provision of equipments mentioned aboved, would be implemented through procedures under the Colombo Plan Technical Cooperation Scheme.

2. Paraguayan side

2.1. Securing the budgetary allocation in accordance with implementation of the Project.

2.2. Appropriate provision of Paraguayan counterpart personnel in accordance with the implementation of the Project.

②　ＩＩＣＳシャガス病等寄生虫症研究プロジェクト
パラグアイ側Ｃ／Ｐ研究報告要約

レスーメン

MOLECULAR BIOLOGY

SUMMARY OF ACTIVITIES

FROM FEBRUARY TO JULY 1990

1.- February, March, April

° Training on basic techniques in molecular biology

- Transformation of E. coli strains.
- Plasmid DNA isolation, miniprep-maxiprep
- Restriction of plasmid DNA
- Agarose-gel electroforesis

2.- May, July:

° Screening of a T. cruzi cDNA expression library using sera of chagasic patients.
Objetive: To identify interesting antigens which could be used for diagnostic purposes. To clone genes producing these antigens.

3.- Jun 25 - July 6:

° Graduate-level course on Genetics of Saccharomyces cerevisiae.

FUTURE PLANS

1.- To continue inmunoscreening of cDNA library

2.- To perform PCR using T. cruzi template and T. cruzi DNA primers. Tentative use in diagnosis.

a. Perform PCR using blood of infected mice.

b. To try technique using blood of infected humans.

PARASITOLOGICAL AND EPIDEMIOLOGICAL STUDIES AND ESTABLISHMENT OF A PRIMATE ANIMAL MODEL.

LIC. ANTONIETA ROJAS DE ARIAS

SUMMARY OF ACTIVITIES

1. Parasitological and epidemiological studies:
Since 1989, epidemiological studies have been performed with the main objective of isolating parasites from chagas patients through hemoculture. When this protocol was presented last year, some concerns were mentioned about the few slim probability of isolating parasites from chronic patients. However we have had now obtained a 27 % positive hemoculture from patients with different types of lesions.
The percentage has been increased as a result of somes introduced in our techniques, which has alowed the successful isolation of parasites.
Details about patients and hemoculture will be explained by Ma Elena Ferreira.
Simultaneously, parasitological studies for characterization of strains are being performed. In this case, our purpose is. first, to establish the biological behavior of isolated paraguayan strains such as: virulence, infectivity, pathogenicity, morphology and tissue tropism.
Five strains were inoculated into Balb/c mice; three of them isolated from acute patients and two from chronic patients. The protocol of maintainance will be presented by Nelson Fleitas.
Our second purpose is the characterization of *T. cruzi* strains by Schizodeme analysis which is underway. Ten paraguayan strains are being studied where six restriction enzymes are being employed. This work will be explained by Marisel Maldonado.

Our final purpose is to perform analytical and epidemiological studies by correlating the biological and biochemical characteristics of the strains with both the types of lesions and the geographical origin of the patients.
Some other techniques are being performed to help other research areas. In other to collaborate with being perform the production of parasites in large amounts, the metacyclogenesis technique has been set up. This group obtains parasites to be processed in antigen fractions which has been used in the animal model assays; Ma. Elena Ferreira wil explain the details about this.
In the epidemiological aspect a few activities have been carry out. About 100 patients were attended in our department during this year.
Epidemiological and personal data were recorded such as, serum test results, EKG, X rays, hemoculture and HLA.
Rural patients have been studied where acute patients are being sought for the eventual detection of early infections.
Epidemiological survey about chagas and leishmaniasis and comparisons of double infection rates and some considerations about

diagnosis will be presented by Margarita Samudio.
Chromosomal studies in chagas'vectors were planned after my visit to Parasitic Disease Departament in Kumamoto University. Interesting results were obtained in this area.
Currently, studies on *T. infestans* obtained from three different towns are been carried out. Detailed aspects of this work I will present later on.

Animal colonies:
Mice breeding colony was set up during this period. Currently an average of 550 mice are maintained and two strains are being bred namely Balb/c and C3H/HeJ.
An early breeding of rats has been started. The animall room has an average of 150 births and 25 mice are requested per month. Mice are used to maintain *T. cruzi* strains, to obtain parasite for celular infections and for immunological assays.

Monkey colony: Some modifications were iintroduced in the colony 3 months ago. Water heater and air heater room, wil be installed. Thirty cages were built, the capacity of the colony is being 68 monkeys distributed in 4 experimental rooms and one quarantaine room. Now, 60 monkeys are living in the colony which has an estimated cost of 15 DS/S per month per monkey.

Insect colony
Two species of insects are being bred in our insectary *T. infestans*, principal vector of chagas disease in Paraguay. These have been obtained from from different places. *R. neglectus* which is not a local insect, is used in experimental assays with natural products and residual effect of insecticides.

We still have a small colony but we hope to get more facilities to increase our breeding for studies on control of vector and laboratory assays.

2. Animal model
The activities for establishing the immunological profile started on June 18 th.1990.
Selection of primate was made according to sex, weight and age, a basaline data about, bllood picture, cardiac enzymes, complement, proteinogram, and total immunoglobulins.
Other basaline data will be eventually obtained during this month and coming september. After evaluation of the results, monkeys will be selected to start the project of biological protection.
The baseline data will be explained by Alicia Schinini and Rosana Galeano.

3. Teaching activities

In the two areas described, we are preparing a teaching program for follows who are working in our deparment. The project activities has been developed with three scholars: Nelson Fleitas,

(Parasitology) Gloria Yaluff (Chromosomal studies of vectors) and Carmen Vitale (Monkey colony).

Equipment and reagents requested:

Parasitological and epidemiological studies:

- electrophoresis chambers
- freezer for serum bank
- air condicioner with controlled temperature for mice colony
- mice cages with water bottles
- shelves
- incubator 28° C.
- Lab benches for Parasitology area.
- supplies: Photograph films for chromosonal studies
- pombo colony (set up)

Animal model studies:

- Cleanning table for necropsy
  with runnig water.
- Reparir of observation room, monkey colony
- Automatic water connection for monkey cages

CHRONOGRAM

AREAS/YEARS	1989	1990	1991
CHARACTERIZATION OF T. CRUZI STRAIN:			
X STUDIES OF PATIENTS	----------	----------	
X ISOLATION OF T. CRUZI	----------	----------	
X SCHIZODEME ANALYSIS		----------	
X MORPHOLOGICAL STUDIES		----------	
CHROMOSOMAL STUDIES OF VECTORS		---------- - - - -	
ANIMAL MODEL			
IMMUNOLOGICAL PROFILE		----------	
BASELINE DATA		--------	
BIOLOGICAL PROTECTION			----------
OTHER ACTIVITIES		---------- - - - - - - - - - -	

MORPHOLOGY UNIT - IICS

SUMMARY OF ACTIVITIES - OBJETIVES - PROJECTS

INTRODUCTION

The Morphology Unit will actually enter the Chagas Project in 1991. Currently, the monkeys are being evaluated epidemiologically and inmunologically. The pathological studies will be performed later in order to investigate the histological changes due to Chagas'disease. Biopsy and autopsy will be used for this purpose. The Morphology Unit has been involved in previous works on chagasic cardiopathy in monkeys, using autopsy, as well as cytotoxicity and HLA studies in Chagasic patients.

During the first year, the Morphology Unit was mainly devoted to consolidate its internal organization and to establish new techniques in Light Microscopy, Electron microscopy, Inmunopathology, Cytogenetic and Experimental Pathology. In fact, we have truly benefited by the Japanese Cooperation through JICA, for the setting up of new techniques in Pathological diagnosis. The development of new techniques are useful not only to our Institute, but also to other Health Institutions of Paraguay. Thus, we have cooperated to the improvement of medical diagnosis and research at national level.

SUMMARY OF ACTIVITIES (April 1989- July 1990)

Until March 1989, the Morphology Unit was just a Department of Electron Microscopy. Since April 1989, a new administration took over, which iniciated a program of activities divided in two stages:

A) ORGANIZATION STAGE: (April-August 1989). New areas were created, such as Inmunopathology, light microscopy and experimental pathology. The Department of Cytogenetic joined the Morphology Unit. The personnel increased from 4 to 13 members. The lab space was expanded and new equipment ,donated by JICA was incorporated. A JICA Expert helped to set up the new facilities.

B) PRODUCTION STAGE (September 1989-July 1990):

1- RESEARCH: a) Chagas'disease projects underway: The following research works have been performed by members of our Unit:

-Cytotoxicity in Chagas (Dra.G.Russomando)

-Production of trypomastigotes in cell culture and in axenic cultures (Dra. G.Russomando)

-Inmunohistological defense detected in mice using trypomastigotes from cell and UV treated (Dra. G.Russomando)

-Histocompatibility antigens in Chagasic patients (Dra.M.Ascurra)

-Chromosomal studies in Chagas vectors (Dra.M.Ascurra)

b) Future Projects in Chagas'disease:

-Endomiocardial and colonic biopsies in Chagasic patients

-Endomiocardial and colonic biopsies in T.Cruzi infected C.Apella monkeys.

-Autopsy studies of T.Cruzi infected Cebus Apella Monkeys.

These works will be done by Dra. E.Kasamatsu and collaborators. Dr. Shozawa from Akita University, who is a specialist in cardiac pathology, is currently giving direction to lab members, in this regard. The same group will be involved in endomiocardial and colonic biopsies in chagasic patients .

c) Other projects: Among other works, no related to Chagas, can be mencioned:

- Preneoplastic lesions of gastric cancer in Paraguay
- Study of malignant solid tumors in children by electronmicroscopy, inmunohistochemistry and cytogenetic.
- Phenotipic and cytogenetic characterization of leukemias and lymphomas in children

-Ultrastructural and direct inmunofluorescence aspects of glomerulonefritis

-Chromosomal studies in spontaneous abortion

2) SPECIAL DIAGNOSIS SERVICE AND TEACHING:

Special diagnosis methods is being provided to all Health Institution of the country in renal, muscular, nervous, tumoral pathology.

Our lab members participate in several scientific events such as seminar, symposia, congress, courses,etc. They are also involved in teaching. Lectures are given to first and third year medical students in area of Histology and Pathology.

Dra. Elena Kasamatsu de Balbontin
Chief of Morphology Unit-IICS

CHAGAS PROJECT

IMMUNOLOGY

SUMMARY OF ACTIVITIES. Period: July 1.989- July 1990.

Staff members:

1. María Agueda Cabello.
2. María Idalia M. de Calabró.
3. Margarita B. de Cabral.
4. María Elisa Vera.
5. Estela Picagué.
6. María Mercedes de Tomasone.
7. Cristina Rovira.
8. Rosa Jimenez.
9. María Angelica Leguizamón.
10. Blasia Cabral. Técnicos.
11. Lucila Pervín.

We continue the following activities:

1. Plaque forming cells: We had standerized the PFC's assay, then we tried to study the humoral immune response in infected T. cruzi mice. Those mice were immunized with sheep red blood cells. The second part of this work (February 90 - June90), the assay was performed with infected T. cruzi mice . These mice were not immunized with Sheep red blood cells .

2. Tumor Necrosis Factor: The porpuose of this study was to measure the production of TNF by splen and peritoneal cells from infected T. cruzi mice.

3. Rheumatoid Factor in Chagas Disease: June 89 - Dec. 89. We had evaluated the presence or the absence of this factor in Chagas Disease's patients. They were in different infections stages. 59 cases had been studied . We are going to continu this work as soon we get the reagents.

4. The Basal Immunological Profile in Cebus Apella Monkeys:

Cebus Apella Monkeys with out T. cruzi infection were studied.

The study included the following points:

a. Immunoglobulin's dosages.

b. Complement's fractions. Part of Humoral Immunity.

c. CH 50

We also assayed:

a. Ig S (surface immunoglobulin)

b. Rosettes Forming Cells

c. PNM's Fagocite activity

d. Reductión of NBT.

It is important to emphasized that most of the monkeys were infected with Filaria.

5. HTLV-1 Infection Prevalence in Paraguay.

This study included people from High Risk Groups for HIV1 infection and japanese immigrants.

The next step for this study is to complete 500 samples from differents groups.

IICS - JICA PROJECT ON CHAGAS' DISEASE

DEPARTMENT OF BIOCHEMISTRY
SUMMARY OF ACTIVITIES
SETEMBER 1989-JUNE 1990

The department of Biochemstry was started in the late 1989. The present staff of the department is as follows:

Dr. Esteban A. Ferro
Dr. Rolando Oddone (IICS scholar)
Dr. Dolly Nuñez (trainee)

* Dr. Elena Zorrilla will be incorporated to the department upon her return from Japan.

1.Production of ITFC-anti-monkey gamma globulin conjugate.
This was made in order to obtain a high quality fluorescent conjugate, using a previously prepared anti-serum in goat. This conjugate was chromatographicaly purified through DEAE-cellulose getting a F/P of 5.4.

2.Preparation of F(ab)2 from goat IgG anti-monkey-gamma globulin.
Papain digestion of immune goat IgG was performed, but the results were rather poor due to insufficient chromatographic facilities at that time.

3.Set-up and training on FPLC.
The Pharmacia FPLC equipment was set-up by a technician who offered the IICS personnel a brief training. The training has been carried out following the intruction manuals and supplementary information, but would be very important to get expert assistance so as to take more advantages from this fine equipment.

4.Isolation of *Cebus apella* IgG.
This was performed by salting-out with 35% ammonium sulphate saturation (following Dr. Nishinarita's suggestion) followed by FPLC analysis and purification using ion-exchange chromatography. The IgG obtained was tested by immunodifussion, electrophoresis and analytical FPLC, proving to be a pure sample.

5.Preparation of anti *C. apella*-IgG goat antiserum.
C. apella IgG was inoculated with complete Freund adjuvant to a healthy goat. This was followed by 2 reinoculations with incomplete Freund adjuvant. This work could not be carried out to completion due to the accidental death of the goat.

6.Isolation of *Cebus apella* IgM.
C. apella IgM was precipitated from serum of healthy monkeys with boric acid. This precipitate has been purified using gel filtration chromatography.

7.Training.

Two memebers of the staff are been trained on protein purification and chromatographic techniques

Future Plans

1. To continue *C. apella* serum proteins purification.
2. To inoculate either goats or rabbits with the isolated proteins to get antisera.
3. To purify *T. cruzi* antigenic fractions useful in diagnosis, protection or skin tests.

4. To search for bioactive compounds from medicinal plants using *in vitro* bioassays (Research proposal presented together with Molecular Biology Department)

Needs

1. Six goats.
2. Facilities for 10 rabbits.
3. Ten rabbits.
4. Glassware for the depatment.
5. Chemicals and reagents.
6. Supplementary furniture.
7. Books and manuals on biochemical techniques.

③　寄生虫学・循環器学セミナーに係る
調査団員講演原稿

JORNADAS PARAGUAYO - JAPONESAS de PARASITOLOGIA y CARDIOLOGIA

PROGRAMA

1-3 DE AGOSTO 1990

JORNADA PARAGUAYO JAPONESA DE PARASITOLOGIA

"ENFOQUE BASICO Y CLINICO PARA EL CONTROL DE LA ENFERMEDAD DE CHAGAS"

Miércoles 1 de Agosto:

Palabras de apertura a cargo del Prof. Dr. Ricardo Moreno Azorero

Coordinador: Prof. Dr. Ricardo Moreno Azorero.

8:30 - 9:30

Caracterización de los tripomastigotes del T. cruzi por medio es estudios experimentales.

Dr. Hiroji Kambara. Prof. del Instituto de Medicina Tropical, Universidad de Nagasaki, Nakasaki, Japón.

9:30 - 9:35

Preguntas

9:35 - 10:35

Recomendaciones al Proyecto Chagas, JICA desde el punto de vista parasitológico, entomológico y epidemiológico.

Dr. Isao Tada, Prof. del Departamento de Parasitología de la Universidad de Kumamoto, Kumamoto, Japón.

10:35 - 10:40

Preguntas

10:40 - 10:50

RECESO

10:50 - 11:50

¿Cómo pueden los investigadores paraguayos y japonese colaborar con el control de la enfermedad de Chagas?

Dr. F. Sendo, Prof. del Departamento de Parasitología de la Universidad de Yamagata, Escuela de Medicina. Yamagata, Japón.

ORGANIZA: IICS - PROYECTO CHAGAS JICA

LOCAL: SALON AUDITORIO DEL INSTITUTO DE INVESTIGACIONES EN CIENCIAS DE LA SALUD

JORNADA PARAGUAYO JAPONESA DE CARDIOLOGIA

Jueves 2 de Agosto:

COORDINADOR: Prof. Dr. Ricardo Moreno Azorero

9:00 - 10:00

Patología de las miocarditis y de enfermedades cardiacas relacionadas - Infecciones bacterianas, virales y protozoarios.

Dr. Takeshi Shozawa, Prof. del Departamento de Patología, Unviersidad de Akita, Escuela de Medicina, Akita, Japón.

10:00 - 10:15

Café

10:15 – 10:45 hs. Diagnóstico de las miocardiopatías
Dr. Laurentino Barrios, Cardiólogo Ecocardiografista
Dr. Rómulo Caffarena, Hemodinamista, Jefe del Dpto. de Hemodinamia I.P.S.
10:45 - 11:45 hs. Estudio Clínico sobre Miocardiopatías
Dr. Shuzo Matsuo, Prof. del Dpto. de Medicina Interna
Colegio Médico Saga, SAGA JAPON

11:45 - 12:30 hs.

Mesa Redonda

Prof. Dr. Carlos Velazquez *Presidente de Mesa*
Prof. Dr. Néstor Arrua T.
Dra. Mirian Ayala
Dra. Elena Kasamatsu
Dr. José Bellasai

Organizado por: Instituto de Investigaciones en Ciencias de la Salud.

Auspiciado por: Sociedad Paraguaya de Cardiología.

Local: Aula Magna - Facultad de Ciencias Médicas.

JORNADA PARAGUAYO JAPONESA DE CARDIOLOGIA

Viernes 3 de Agosto:

COORDINADOR: Dr. Enrique Courcelles.

9:00 - 10:00

Patología de la Cardiología isqémica
Dr. Takeshi Shozawa, Prof. del Departamento de Patología, Universidad de Akita, Escuela de Medicina, Akita, Japón.

10:00 - 10:10:15

Café

10:15 - 10:45

Diágnostico y Tratamiento de la Cardiopatia Isqémica.
Dr. Enrique Courcelles, Jefe del Servicio de Cardiología I.P.S.
Dr. José Corvalán, Jefe de Cirugía Cardiovascular Sala X,
Dr. Miguel Adorno, Jefe del Servicio de Cardiología HUNSA

10:45 - 11:30 hs.

Mesa Redonda. *Cardiopatía Isquémica.*
Prof. Dr. Omar Sosa *Presidente de Mesa*
Prof. Dr. Oscar Lovera
Dr. Rubén Diaz Juré
Dra. Elena Kasamatsu
Dr. José Bellasai

Organizado por: Instituto de Investigaciones en Ciencias de la Salud.

Auspiciado por: Sociedad Paraguaya de Cardiología

Local: Aula Magna - Facultad de Ciencias Médicas

IICS－JICAプロジェクトセミナー報告書
（平成2年8月1日～3日）

於　IICS，パラグアイ

山形大学医学部寄生虫学講座
仙道 富士郎

パラグアイIICSでのセミナーで「いかにして、パラグアイと日本の研究者はシャガス病克服のために協力できるか」というタイトルで、下記の内容の講演を行ったので報告する。

(添付資料：セミナープログラム)

再びここで皆さんにお話出来ることを非常にうれしく思います。今日お話することは、私の専門に関することではなく、JICAのプロジェクト全体についての一般的なお話ですので、私の英語力ではとてもお話できませんので、日本語でお話させていただくことをお許しください。

プログラムに載せましたタイトルで今日お話しようと思った理由は、プロジェクトもすでに2年半が経過し、折り返し地点にきておりますので、この辺で皆さんと一緒にこれまでのプロジェクトを振り返り、それをもとにしてこれからの後半部分を成功裡に行っていくことが、現在時点で非常に大事な問題ではないかと思ったからであります。あくまでも私個人の意見であり、いろいろ足りない点、あるいは誤解している部分があるかもしれませんが、そのような点に関しては皆さんから後で補ってもらえれば有難いと思います。またこのプロジェクトに深く関わってきた人達には、わかりきったことを申し上げるようなことになるかもしれませんが、ここに参加に皆様方全体の理解を得るために、若干繰り返しになるかもしれませんが、その辺は御容赦下さい。

まず、プロジェクトを行うにあたっての基本的な事項について説明いたします。第一にプロジェクトを行うにあたって、日本の政府とパラグアイの政府が協力しあうというのが基本的なポイントであります。その協力関係の上にたって日本側としては、技術協力、技術移転に必要な専門家を派遣いたします。また、その技術移転に必要な機械・器具などを用意することがうたわれております。さらには、IICSの研究者を日本で技術研修を行うということが第三点としてあげられます。

それでは、プロジェクトを開始する際に決められた暫定的な計画の各項目について、今までどのようなことが行われてきたか、またこれからどのようにしていったらいいのか、私の考えを述べていきたいと思います。まず第一点目としては、組織培養システムの確立と単クローン抗体の産生についてであります。最初のスライド

にも書かれてありますように、このプロジェクトは研究プロジェクトでありますので、研究を行うのに十分な設備を確立することが必要であると考えられました。ご存知のように現代の生物学においては、細胞を試験管内で培養することが必須の条件であります。そこで、まず培養システムを確立することを最初のポイントと考えました。組織培養には、ご存知のように非常に純度のいい蒸留水を用意することが、必須であります。そこで、まず純水装置のセットアップをいたしました。セットアップの後2年半の間にいろいろなトラブルもありましたが、現在は、IICSの研究者の方々の手で立派にメンテナンスされていると思います。この純水装置の維持は、このプロジェクトにとって一つの生命線でありますので、今後も十分な管理をしていくことが必要かと思われます。次に単クローン抗体の確立があります。この研究プロジェクトの一つの最終的な目的は、免疫学的な手法によってシャガス病を克服するということでありますので、その免疫学的な研究にとって現在においては、一つの重要な道具となっております単クローン抗体の樹立を試みたわけであります。その結果いくつかのトリパノゾーマクルーザイに対する単クローン抗体を樹立することができました。単クローン抗体が出来るための条件としては、非常にいい質の水がどうしても必要だということになっておりまして、そういった意味からも、IICSで現在使われている純水装置が、十分に作動しているということを証明しているかと思います。ただこの単クローン抗体については、当初の我々の仮説が若干間違っていたこともありまして、ワクチンを作るのには非常に大事だと考えられますトリポマスティゴートに特異的な抗体はまだ得られておりません。といいますのは、私たちが最初考えましたのは、トリパノゾーマクルーザイに感染したマウスを用いた方が、感染状態で認識される抗原に対する抗体をよく反映するのではないかというふうに考えたわけですけれども、最近の私たちの研究室で明かになったことでありますけれども、感染動物から得られた単クローン抗体は、多くの場合はいわゆる自己抗体を含む、交差反応性の高い抗体であることがわかってきております。そこで今後の問題としては、トリポマスティゴートそのもので免疫したマウスの脾臓細胞から単クローン抗体を確立する必要があるというふうに考えられます。この点は、最終的なワクチンの確立に向けて非常に大事なポイントではないかと思います。

以上のような研究成果をふまえてまして、次に行われました免疫学的な研究について説明いたします。まず最初にあげなければならないのは、すでにこのプロジェクトが始まる前にDr.カベーリョとDr.カブラールらによって、おおよそのことはわかっていたのでありますけれども、Cebus appelaのリンパ球が羊の赤血球とロゼットを作るという現象の解析があります。この現象をさらに詳しく解析するために、人のリンパ球に対する単クローン抗体でこの羊の赤血球に対するロゼットが抑制されるかどうかをみてみたわけでありますけれども、見事に抑制されるということが、判明いたしました。また、シャガス病の患者さんのリンパ球が、一定のHLA抗原を持

っているかどうかを調べたわけでありますけれども、まだ例数がそれほど多くありませんけれども、やはり慢性のシャガス病の患者のHLAには一定の傾向があるという結果が得られております。最終的にはワクチンでシャガス病を予防、あるいはできれば治療するといたしますと、シャガス病患者がどのような免疫学的な状態にあるかということを知る必要があるわけでして、その一つの試みとして重要なサイトカインの一つでありますtumor necrosis factorの産生が、トリパノゾーマクルーザイに感染したマウスでどのようになるかを調べております。これから研究を更に進めていく必要がありますが、感染後の経過によってTNFが非常に強く産生される時期があることがわかっております。また、同じ様な研究としては、シャガス病の患者には、いろいろな自己抗体が産生されることがわかっておりますが、その一つと考えられますリウマチ因子についても検討しておりまして、興味ある結果を得ております。今後更に症例を増やしていく必要があるだろうと思います。また、その再現性に若干問題があると聞いておりますけれども、トリパノゾーマクルーザイで免疫したマウスのリンパ球がトリポマスティゴートを試験管内で殺害するという予備的な結果が得られております。以上説明いたしました研究は学問的にも非常に大事な結果を含んでいると思われますが、最初に述べましたサルのリンパ球のロゼット形成以外の実験については、まだ完成されているとはいえないと思います。そこで、ぜひこれらの研究を完成させまして、しかるべきジャーナルに投稿してただきたいと思います。免疫学的な研究については、ジャーナルに投稿するということが現在残されている一番大事な問題ではないかと思います。

次に寄生虫学的な研究についてでありますが、当初免疫学的研究が先行したために、この分野の研究はプロジェクトでまだ十分進んでいるとは思われません。しかしながら、シャガス病の本体を解明するには、寄生虫学的研究は基本的な分野でありますので、今後更に大きなスケールで研究を進めていく必要があると思います。これまでにやられました研究としては、シャガス病の患者さんからのトリパノゾーマクルーザイを分離して、いろいろな株を確立して、その性状を調べるということをしております。またさきほど申しあげましたワクチンの研究にはどうしても大量のトリポマスティゴートを産生する必要がありますが、試験管の中でそれが可能であるという結果が得られております。さらにはこの問題は、後で述べます分子遺伝学的な研究とも関連ありますけれど、トリパノゾーマクルーザイのキネトプラストのDNA解析も行われております。また、媒介体でありますさしがめのクロムゾームの解析も開始されております。以上の検索は、いずれも始まったばかりでありますので、手掛けた研究を更に深めることと、それからどのような寄生虫学的な研究が今後必要なのかを、皆さんで大いに討議してもらいたいと思うわけであります。

4番目のプロジェクトのタイトルといたしましては、サルを用いたシャガス病の実験モデルの確立及びそれを用いたワクチンの研究があります。申すまでもなくこのプロジェクトの最終的な研究目標は、人のシャガス病を免疫学的に克服するということにあるわけですけれども、ネズミをいつまでも用いていたのでは、それに近づくことは出来ません。すでにIICSではCebus appelaを用いまして、シャガス病のモデルが作られておりました。そこでこのモデルを用いまして、シャガス病本体をさらに研究することにいたしました。またこのCebus appelaを用いて、できれば最終的には、ワクチンでシャガス病を予防、治療できるシステムを作り、それをもとに人のシャガス病の研究へ持っていこうというのが基本線であります。このサルのモデル実験についてはまだ多くのやらなければならないことが残っております。まずは、実験に用いますサルの供給をより確立していく必要があります。これなくしては、このプロジェクトは進んでいかないというふうにも考えられます。そうした十分なサルの供給に基づきまして、まず調べなければならないことは、トリパノゾーマクルーザイに感染したサルがどのような免疫学的な状態にあるかを十分に検討することであります。そうしたことをしなければワクチンの研究にはつながっていかないと思われるからであります。しかるのちにあるいはそれと平行して、まずマウスを用いた実験で本当にワクチンをつくることが可能であるかどうかということを検討することが必要であります。これも非常に大事な基礎的研究だと思われます。さらにはそのようなマウスのモデルを使いまして、トリパノゾーマの抗原をワクチンに用いるようにするには、どのようにしたらいいかということを検討する必要があります。つまり抗原の精製であります。これには免疫学、生化学さらには分子遺伝学などの各分野の協力が必要であると思われます。

ここにあげました4つの項目につきましては、まだ研究はほとんど進んでおりませんで、これからの問題であります。つまり後半部分のプロジェクトをいかに実りあるものにするかは、ここにあげました4つの点についていかにいい結果が得られるかにかかっていると思われます。そこで大事なことは、どのようにしてこれらの点を研究していくかについて十分なdiscussionが必要であるという点であります。

以上、プロジェクト開始時に決められました全暫定的な研究計画の各ポイントにつきまして、私なりに今までどういうことが行われてきたか、またそれに基づいて今後どのようなことが必要であるかという点について考えを述べさせていただきました。次に以上のような研究を行うにあたっての基本的な視点について私の考えを述べさせていただきます。まず、最初にも説明いたしましたように、このプロジェクトは研究プロジェクトなわけでありますけれども、パラグアイのような発展途上

国において基礎自然科学を確立することは、一体可能であるのかどうかまたそれは必要であるのかどうかという点について考えてみたいと思います。このプロジェクトの最終的な目的は、いうまでもなくまた何回も私の話のなかでましたように、シャガス病を免疫学的に克服するということにあるわけであります。けれども、それならば自然科学の非常に進んだ国の成果をふまえてそれを応用していくのが妥当ではないかという考え方があるかと思います。この考え方は一見妥当であるように思いますが、私は必ずしもそうではないと考えております。と申しますのは、パラグアイの人々の健康を守っていくのは、パラグアイの人たちの手によるほかないわけでありますけれども、そうしますと現在の非常に早いスピードで進んでいます自然科学に対する深い洞察を持った上でなければ、それは可能でないと思われます。そのような現在どんどん進んでいきます自然科学の情報を得れば、それでことは足りるのではないかという考えもあるかと思いますけれども、これまで自然科学に関わってきた人間の一人としていえますことは、自然科学の理解といったものはそういった単なる情報の受渡しによるだけでは可能ではないというふうに思うわけであります。つまり自分の手でいろいろなことを体験してはじめてその中から自然科学に対する深い造詣というものが培われていくというふうに思われるのであります。そのような立脚点にたちますと、いろいろな点からパラグアイにおいて基礎自然科学を樹立することは、非常に困難であるということが予想されますけれども、やはりどうしても避けて通ることの出来ない大事な道の一つであると、私は考えるわけであります。そのようなことが必要だとするならば、どのようにしてそれをやっていくことが出来るのか。まず第一にそれを可能にしていくのは、優秀な自然科学者を育てていくことであります。自然科学者を育てるということは、言葉でいうのはに簡単でありますけれども、なかなか大変なことでありまして、やはり彼らが育っていく場を作らなければならないと思います。そういった意味で、このプロジェクトがお役にたてば非常にうれしいと言うふうに思うわけであります。何回も申しますけれど、このことは簡単な道のりではないということはよくわかりますけれども、私たちが協力して通り抜けていかなければならない道であると言うふうに確信いたします。

次に問題になりますのは、このような基礎的な結果をどのように臨床に応用していくことが出来るだろうかということであります。第一点目としては、臨床と基礎科学のちょうど中間に位置いたします臨床病理学の研究を強化していくことがあげられます。すでにこのプロジェクトでこの研究に必要な機材はじょじょに設備されつつあると思っております。次に問題になりますのは、パラグアイにおけるシャガス病の実態を知った上で事にあたるのでなければ、机上の空論になりますので、やはりその実態を知るためには疫学的な調査研究が必要になりまして、この点も非常に大事な問題であると思います。第三番目としては、シャガス病患者の病態を正確

にとらえるためにはその臨床像を詳細に検討していく必要があります。その点で生化学的な検査あるいは心エコーを消化管カメラなどを用いましたシャガス病患者の十分な検索が必要になってくると思います。このような病理学的あるいは臨床学的なシャガス病の研究は、このプロジェクトの後半部分においてに大事なものであるということは先ほどから何回も私が申し上げている通りでございます。

さて、最後にこのプロジェクトをさらに発展するためのいくつかの問題点について触れてみたいと思います。まず第一点目としては、日本の専門家とパラグアイのカウンターパートとの間にヒューマニズムに基づいたよりよい関係を確立することであります。私も含めまして日本の専門家は、パラグアイの母国語であるスペイン語をほとんど理解することが出来ません。この言葉の壁が大きな問題にであることは、よく承知しておりまして、我々が今後努力していかなければいけない問題でありますけれども、そういった言葉の壁を乗越えるような関係を私たちは作っていかなければこのプロジェクトが成功裡に終わることはないだろうと言うふうに思います。また、何回もパラグアイ側から言われていることでありますけれども、日本人専門家の専門分野を、早く知らせてほしいということでありますけれども、この点については私自身非常に反省しているわけでありますが、今後十分注意して出来る限り早くにIICSに来る専門家の分野についてお知らせするように努力したいと思います。次の問題としては、IICSのカウンターパートの勤務時間の問題があります。多くの方がハーフタイムでありますけれども、細胞を扱う仕事には非常にこの点は致命的な点であるとも思います。これもどのようにして解決していったらいいか、十分な討論をお願いしたいと思います。また以上の問題とは若干問題点を異にいたしますけれども、JICAから搬入されました多くの機材があり、これをいかに有益に使っていくかというその方法を確立するのも大事なことではないかと思います。そのほか私が気が付かない多くの点があるかと思いますが、今後お互いに十分なdiscussionを通して新しい道を見つけたいと思っております。

以上、私の考えを述べましたが、私たちの行く手に多くの困難があることを否定いたしません。しかし、太陽がのぼっていきやがてはさんさんと照り輝くように、このプロジェクトが必ず大きな成功をあげることを信じて疑いません。また、私たちはその努力をしていかなければならないと確信するものであります。

〔資 料 1.〕

SUGGESTIONS FOR JICA CHAGAS' DISEASE PROJECT FROM PARASITOLOGICAL, ENTOMOLOGICAL AND EPIDEMIOLOGICAL VIEWPOINTS.

Dr. I. Tada, M.D.
Kumamoto University
Medical School, Kumamoto
JAPAN

The major purposes of our technical collaboration projects in the field of medical science are : a) Assistance for the research/control activities in public health of the countries, and b) Development of human resources concerned in the above activities. From this viewpoint, I would like to present some suggestions for the project in IICS of the National Asuncion University, at its third year of collaboration program since 1988.

Based on the establishment of cultivation of Trypanosoma cruzi, various immunological techniques aiming at the studies of the protective immunity have been established in the laboratory so far. And now encouraged are the molecular biology.

My suggestion is to introduce multidisciplinary technologies in the project not to disperse the focus, but to encourage entire activities in the research.

1. Parasitological view:

With regard to the identification of T.cruzi strains, various techniques should be introduced, such as kDNA fragment patterns, isozyme techniques and specific antigens etc. The former technique is on the way by Dr. Mimori with Lic. Samudio. Recently, Breniere et al (1989) examined separated parasite strains from 495 Bolivian patients by 12 isozyme loci and made ECG diagnosis of the patients. They found zymodeme diversity in the endemic area and less enteric form of the diseases in their

territory. Thus, they concluded necessity of examining the association between paathology and zymodeme. This is an example to apply isozyme technology and shows its significance in the analysis of clinical pathology, a more practical view of the disease.

TDR news of June 1990 reported a blood-screening project of the defined recombinant peptides (produced by each of 10 laboratories from 4 countries) for diagnosis of Chagas' disease supported by a reference laboratory in the Federal University of Goias, Brazil. Participation to such an international project will be stimulating in the present project.

From the clinical view, the most important field would be the chemotherapy of the disease. Nifurtimox and Benznidazol have been traditionally used so far, while they reveal serious accesory reactions frequently. Various chemicals are now being tested recently, such as Verapamil (Tanovitz et al., 1989), alopurinol (apt et al., 1987) etc. Are there no indigenous herbal components which are effective to T.cruzi ? I would like to encourage any intention to find active traditional crude drugs for treatment of Chagas' disease.

2. Entomological view:

Shenone (1987) describes that the most important vector in Paraguay is Triatoma infestans and 18.2 % of them are infected with T.cruzi. Recently Rojas (1990) reported a similar and detailed evidence on the vector triatomine bug. On this species, Hirai et al. (unpublished) clarified cytogenetically the presence of three cytotypes in a T.infestans colony. They

estimated that the colony does not perform random mating and that incompatibility-like regulation is probably occurring. This eidence, if it is frequently occurring in the natural condition, shows a possible genetic control of the bug. The on-going residual spraying/fumigation of any insecticides is causing various problems in the world. Any possible biological control measures should be encouraged including the genetic analysis of the bug.

As is stated by Rojas (1990), T.infestans and T.sordida are the number 1 and 2 vectors in Paraguay. Schoffield (1987) points that they compete same ecotope of the dwellings, so that studies on the ecology of triatomine bugs should be enforced for the control trial.

3. Epidemiological view:

Recently, Mota et al. (1990) reported a prospective study of Chagas' disease in Castro Alves of Bahia, Brazil by examining seroconversion rate and development of abnormal ECG in the population. They assessed the association between epidemiological findices and risk of cardiological lesions. A similar sero-epidemiological study was reported by Rojas (1990) dealing with the populations in Departments of Boqueron and Paraguari, Paraguay. In order to clarify the pathogenic feature of Paraguayan disease, and to give guide line for clinicians, the epidemiological analysis is essential. On this context, epidemiologists should be trained who collect data and analyze them by the computer processing. For this purpose, Dr. Nozaki of Nippon University is certainly helpful in organizing the system

as a member of the steering committe in JICA.

In the former portion of my presentation, I proposed some suggestions from the technical viepoints. However, in order to develop the present collaboration project, successfully, I would like to show some suggestions on the Paraguayan counterparts.

1. Counterpart-expert relationship.

In order to maintain and develop any scientific techniques, qualified counterparts with enough time for experiments is essential. In order to avoid inconveniences in the custom and economy, I plea the authorities of university to give them some administrative relief. Arrangements will also be necessary in the assignment of counterparts among several aid projects by foreign countries. For this purpose, a steering committee of the IICS should be responsible including representatives of foreign aid projects.

As is required by Dr. Moreno when he visited Kumamoto University this March, a good relation should be maintained in counterpart-experts combination even when the collaboration period in Paraguay finished. I recommend Japanese experts to continue communication back-up by sending references etc.

2. Seminar and field-training as lessons.

In the project, in order to give younger counterparts the entire figure of Chagas' disease, seminar and field training will be quite useful by offering knowledge obtained from basic to sophiscated techniques. I would like to require the authorites of IICS to arrange such courses. The virtual experiences obtained will encourage motivations in the individual rsearch field.

Collections of triatomine bugs, bed-side teaching on patients, reading of ECG etc will stimulate younger researchers.

3. Publishment of results.

Scientific reports are the final productions which shows that the writer(s) have engaged in research. As researchers are evaluated primarily by their papers, even the short communications are very important in any scientific journals. From this viewpoint, younger counterparts should learn how to write scientific papers based on the order and moral of the scientific publishment. At the present situations, English is the cosmopolitan language to be used, while Spanish papers are also important, particularly when the papers are considered useful for domestic readers.

4. Application of research grants.

In order to have own grant for the research, application to some domestic and international agencies such as TDR (Special Programme for Research and Training in Tropical Diseases) is quite desirable. Research grants will undoubtedly improve research condition and enhance activities. In order to get informations on this, TDR news are available by the request.

Finally, please remind again the fact that we are already in the thrid year of the 5-year program of this collaboration project. For the continuous development and maintenance of the research facilities, it will be necessary to encourage younger researchers. I sincerely hope the high-ranked staffs of the IICS and the University of Asuncion to give them favorable conditions in the position and salary.

④　パラグアイ原子力委員会作成のＩＩＣＳ放射性廃棄物管理調査報告及び処理に係るガイドライン

Ministerio de Relaciones Exteriores
COMISION NACIONAL DE ENERGIA ATOMICA

— * —

Asunción, Cs de 19

FORMULARIO PARA SOLICITUD DEL SERVICIO DE DOSIMETRIA PERSONAL

AL QUE SUSCRIBE . . . . . .

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CON DOMICILIO PROFESIONAL EN . . . . . .

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DE ACTIVIDAD . . . . . .

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SOLICITA EL SUMINISTRO DE DOSIMETRO CON FRECUENCIA . . . . . .

. . . . . .

(MENSUAL, SEMANAL, ETC.) . . . . . .

FUENTE DE RADIACION UTILIZADA:

RADIOISOTOPOS ____ RAYOS X DE USO MEDICO ___

RAYOS X DE USO INDUSTRIAL ___ RAYOS X DE INVESTIGACION ___

FAVOR MARCAR LO QUE CORRESPONDA.

ENTIENDO QUE EL DOSIMETRO DEBE SER PORTADO EXCLUSIVAMENTE EN EL LOCAL O LOCALES DE TRABAJO.

. . . . . .

FIRMA

Ministerio de Relaciones Exteriores
COMISION NACIONAL DE ENERGIA ATOMICA

— * —

DIRECCION GENERAL DE CIENCIA Y TECNOLOGIA NUCLEAR

DEPARTAMENTO DE PROTECCION RADIOLOGICA

Servicio de Dosimetría Personal

El Departamento de Protección Radiológica ofrece el Servicio de Dosimetría de Personal utilizando Dosimetría por Termoluminiscencia.

El mecanismo empleado es el siguiente:

La Comisión Nacional de Energía Atómica entrega al usuario, debidamente registrado un porta-dosímetro con las pastillas-dosímetro. Una vez al mes en fecha pre-establecida se presentan a la Comisión Nacional de Energía Atómica, Edificio Paraguaí en Chiraby y Estrella Primer Piso Oficina 102, los porta-dosímetros para el canje de las pastillas, por personal técnico de esta Comisión Nacional.

Mensualmente se presenta el informe al usuario del Servicio.

El costo del servicio es de 1.000 ₲. (un mil ochocientos guaraníes), por cada cambio de dosímetro (generalmente mensual; si algún usuario requiere un servicio más frecuente, también es posible).

Por el porta-dosímetro se entrega una garantía de 5.000 ₲. (cinco mil guaraníes) por cada uno, suma que se devuelve al fin del servicio.

Se debe llenar el formulario correspondiente.

Ministerio de Relaciones Exteriores
COMISION NACIONAL DE ENERGIA ATOMICA

D.G.C.T. N°: 041/90

MEMORANDUM

A: Prof. Dr. Ricardo Moreno Azorero

DE: Prof. Dr. Juan F. Facetti

ASUNTO: Su pedido sobre: Eliminación de desechos radiactivos.

1. Existen 3 pasos básicos en su manejo.
   A) Recolección
   B) Tratamiento: necesario para altas actividades.
   C) Eliminación.

En todos estos casos, el personal de operación debe estar adecuadamente protegido.

2. Para desechos sólidos

Se guardan en cajas de cartón.
Conviene encerarlas o parafinarlas por dentro.
Se recolectan las cajas una vez por semana o en lapso mayor, según sea la actividad.

a) Muy baja actividad:
Se puede agrupar los sólidos en paquetes con intensidad de exposición en contacto no mayor que 6 mR/hora y "tirar a la basura" no más de uno por recipiente de colección de basura, ni más de 3 por día. Cuidar detalles que tengan efectos sicológicos infundados pero indeseables en la población.

b) Actividades medias
Si la exposición en contacto es mayor que 6mR/h pero menor que 200mR/h, se necesita una persona que proceda al retiro periódico.
b.1. En este caso, el material se incinera, cuidando de retener la emisión de polvo; las cenizas se depositan en calicatas y se cubre con tierra gorda.
b.2. Cuando el material no fuere incinerable por sus efectos poluentes, se prensa y se entierra "in totum".

3. Para desechos líquidos:

a.1. Para actividades no mayores que 5 uCi/ml, se diluye con abundante agua hasta una actividad menor que 10^{-2}uCi/ml. y se elimina por las cañerias sanitarias.
Se repite esta operación las veces que sea necesario.
a.2. Para líquidos con actividad mayor que 5 uCi/ml, pero menor que que 10^{2} uCi/ml. el personal técnico del laboratorio, puede fraccionarlo, depositando en recipientes con A < 5uCi/ml, para después individualmente someterloa dilución como en a.1. En este caso, conviene que la operación la hagan 2 personas, (el técnico y un personal de protección radiológica).
a.3. Para actividades del orden de 50 a 100 uCi/ml, ^{137}Cs o ^{90}Sr de vida media larga, conviene construir una calicata con tosca por piso, cubriendo de arena y pedregullo, y allí verter los desechos. (El Cs y el Sr, son totalmente retenidos en los primeros 7,5 cm de la tosca en el campus de San Lorenzo).

4. Relación entre Almacenamiento y Período"

a.1. Debe recordarse que radioisótopos con T 1/2 corto, se pueden dejar decaer y luego eliminarlos. Siempre que se pueda es apropiado un lapso de decaimiento. Una regla útil es considerar 5 períodos como un buen tiempo de decaimiento.
Por ejemplo: I-131 con 40 dias es suficiente.

5. Relación entre " aislamiento" del material y la energía del radioisótopo.

a.1. Debe recordarse que los emisores Beta puros (sin gamma) quedan totalmente blindados por su propio contenedor.
a.2. Cuando existen gammas debe proveerse blindaje; recordar que gamas de baja energía requieren pocos espesores.
a.3. Debe recordarse también que el agua funciona como blindaje.

6. Almacenamiento de desechos.

- Las bajas actividades se pueden almacenar en las campanas , con protección de plomo (ladrillos o "chanchitos") hasta su recolección períodica.
- Hasta su disposición final, cuando se requiere mantener grandes volumenes de líquidos, puede construirse si se desea un pequeño bunker con paredes de 45 cm de espesor donde tambien podrá guardarse (siempre en blindajes) actividades altas de radioisótopos que vayan a ser posteriormente fraccionados.

Las dimensiones de la sala se debe definir de acuerdo al volumen previsto de material radiactivo, así como la zona de separación entre desechos y radioisótopos utilizables.

Es conveniente tener chimeneas con estractores para ventilación.

La altura de la chimenea debe culminar a unos 2 ms. por encima de los techos de las edificaciones cercanas.

El nivel de radiación en el local debe controlarse periodicamente, y nunca entrar en el mismo sin la asistencia de un profesional de Protección Radiológica para los controles previos.

Periodicamente se debe controlar el nivel externo de radiación en contacto con la pared.

Nunca debe ser mayor que 2,5 mR/h.

7. Dosimetría de personal

De acuerdo a las reglamentaciones vigentes de uso internacional y nacional el personal que trabaja con radiación debe ser monitoreado mensualmente respecto de las dosis que recibe ocupacionalmente.

Los sitemas vigentes son a) por Film-monitor b) Termoluminiscencia.

La Comisión Nacional de Energía Atómica ofrece ambos servicios , pero recomienda para el caso de esa Institución el de Termoluminiscencia.
Adjunto una hoja explicativa así como la solicitud para el efecto.

Estos son principios y normas generales que podrán guiarles en la solución del problema que plantean . Como puede notarse todo depende del o los radioisótopos a utilizar, de sus vidas medias y de su energía.

Cordiales saludos.

Asunción, 3 de agosto de 1990.

⑤　平成２年度供与機材リスト

番号	品名及び仕様	メーカー名	数量	単価	金額
1.	複合電子走査形超音波診断装置	東芝			
	1. 複合電子走査形超音波装置, 220V. 50Hz 構成 1) 本体 (1) 2) ドップラーユニット (1) 3) 電子セクタープローブ(ドップラ) (1) 4) 電子セクタープローブ 3.75MHz(1) 5) 参考信号ユニット (1) 6) ノンフェードユニット (1) 7) アクセサリー (1)	SSA-250A (システム19) SDS-250A PSF-25R PSE-37H UJUR250A UINF250A	1式		13,800,000
	2. 電子セクタープローブ (小児心臓) 5.0MHz	PSF-50SS	1ケ		530,000
	3. フットスイッチ	UZFS001A	1ケ		40,000
	4. 電源安定装置		1式		130,000
	(関連附属品) 1. ソノプリンター 2. ソノペーパー (6巻/箱) 3. 超音波ゼリー、5ℓ 4. V.C.R. 1/2″、VHS (リモコンユニット付)	 TP-8300 B-310 BR-6400	 1ケ 10箱 2ケ 1式	 8,200 13,700	 220,000 82,000 27,400 380,000
2.	内視鏡機器				
	1)OES上部消化管汎用ファイバースコープ、GIP TYPE XP-20	オリンパス	1式		1,250,000
	2)OES下部消化管ファイバースコープ PCF TYPE20	〃	1式		1,330,000
	3)OES光源装置、CLE-10 150Wハロゲンランプ6ヶ付、220V, 50Hz	〃	1式		388,000
	4)ガストロファイバースコープ GIF TYPE P-20	〃	1式		1,250,000
	5)OES大腸ファイバースコープ CF TYPE 1T20I	〃	1式		1,560,000
	6)35m/mカメラ、OM-IN	〃	1ケ		44,000
	7)OMアダプター、A10-M1	〃	1ケ		37,000
3.	泳動及びブロッティング装置				
	1)核酸シーケンシングシステム166-1012 (内訳) ①核酸シーケンシング電気泳動槽 (1) ②モデル 583ゲルドライヤー (1) ③モデル 3,000Xiパワーサプライ (1) ④アクリルアミド99.9%, 100g入 (1)	バイオラッド	1式		985,000
	2) 2-D電気泳動システム (内訳) ①プロティアンII, 2-Dセル20cm No. 165-1933 ②モデル225チューブゲルキャスティングスタンド No.165-2020 ③モデル556ゲル脱色装置No.165-2010 ④モデル385グラジェントフォーマー	バイオラッド	 1式 1式 1式 1式		 282,000 47,800 106,500 106,500

番号	品 名 及 び 仕 様	ﾒｰｶｰ名	数量	単 価	金 額
	3)小型電気泳動システム MUPID-3 AC100Vトランスフォーマー付	ｱﾄﾞﾊﾞﾝｽ	3式	48,700	146,100
	4)セミドライブロッター (内 訳) ①SM17556,ザルトブロットⅡ ②SM11306/SM15906/SM12753 各1式 ③SM11307/SM15906/SM12753 各1式	ｻﾞﾙﾄﾘｳｽ	 1式 1式 1式	 	 132,000 47,600 47,600
	5)マイクロスラブ電気泳動装置 KS-8020	マリソル	1式		71,000
	6)高精度安定比電源 MP-7655, AC220V用	マリソル	1式		152,200
	7)シーソーシェーカー BC-700,AC220V用	ﾊﾞｲｵｸﾗｯﾄ	1式		117,700
4.	インキュベーター付回転培養器HB-OVI型 AC220V,50Hz ﾎﾞﾄﾙ35x300m/m 12本付	HYBAID	1式		680,000
5.	病理組織標本用機器				
	1)ミクロトーム刃自動研磨器 MN-72型, 220V, 50Hz	サ ク ラ	1式		1,120,000
	2)ミクロトーム刃, B17cm 08-720-2	エ ル マ	3本	26,400	79,200
	3)岩岡式なじみ革砥 08-955-0, 2本／組	〃	1本		36,500
6.	臨床検査用双眼顕微鏡 BHTU-112, 220V, 50Hz 本体部：(BHTU, F-set) 鏡 筒：双眼 (BH_2-Bi30) ステージ：共軸右下ﾊﾝﾄﾞﾙ (BH_2-SVR2) コンデンサー：ﾊﾈﾉｹ式 (BH_2-SC) 対物レンズ：DPLAN 4X,10X,20X,40X, 100X (各1) 接眼レンズ：WK10X (2) スペアーハロゲンランプ (6V,20WHAL) 6ケ付	ｵﾘﾝﾊﾟｽ	2式	446,000	892,000
7.	冷 凍 庫				
	1)超低温フリーザー MDF-292AT 温度範囲：−85℃ 有効内容積：180ℓ 外寸法：1500(W)x700(D)x945(H)m/m 内寸法：760(W)x420(D)x565(H)m/m 220V, 50Hz, 単相 (特別付属品) 貯蔵ケース, MDF-39SC	サンヨー	1台 4ケ	 15,200	1,540,000 60,800
	2)メディカルフリーザー MDF-230 温度範囲：−30℃ 有効内容積：222ℓ 外寸法：924(W)x743(D)x883(H)m/m 内寸法：790(W)x440(D)x715(H)m/m 220V, 50Hz, 単相	サンヨー	2台	253,700	507,400

番号	品名及び仕様	ﾒｰｶｰ名	数量	単価	金額
8.	真空凍結乾燥機	ラブコンコ			
	構成：				
	1) 77530 LL-6本体床置型		1台		1,345,000
	2) 77560 ｽﾄｯﾊﾟﾘﾝｸﾞﾄﾚｲﾄﾞﾗｲﾔｰ		1台		2,120,000
	3) 77716 特注サポートスタンド		1台		241,000
	4) 0076 ALSCO-150 真空ポンプ		1台		226,000
	5) 75406 300mℓ FF フラスコ		5ケ	28,400	142,000
	6) 75408 600mℓ FF フラスコ		5ケ	29,800	149,000
	7) 75409 900mℓ FF フラスコ		6ヶ	30,800	184,800
	8) 75458 3/4"-3/4" 曲管アダプター		16ケ	2,890	46,240
	9) トランス・スタビライザー		1台		289,000
	10) 真空ポンプオイル（18ℓ缶）		2ケ	28,900	57,800
9.	偏光双眼顕微鏡 BHS-651P(220V用)	オリンパス	1式		856,860
	本体部：(BHS-P-Set)回転芯出しｽﾃｰｼﾞ(BH_2-SRP)付 検板：(AH-TP530-2)(AH-TP147-2) 鏡筒：双眼(BH_2-Bi30) 中間鏡筒(BH_2-PA)付 レボルバー：(BH-PRE) コンデンサー：偏光用(BH_2-Poe) 対物レンズ：PODACH 4x, 10x, 20x, 40x, 100x（各1） 接眼レンズ：WHK10x(1), WHK10x-CROSS(1), WHK10x-MICRO(1) スペアーハロゲンランプ（12V 100W HAL-L）6ケ付				
10.	心臓カテーテル生検装置				
	1)ｶﾃｰﾃﾙｲﾝﾄﾛﾃﾞｭｰｻｰ C155P11TSM	ﾒﾃﾞｨｷｯﾄ	20ケ	7,900	158,000
	2)ｶﾃｰﾃﾙｲﾝﾄﾛﾃﾞｭｰｻｰ C170P11TSM	ﾒﾃﾞｨｷｯﾄ	20ケ	7,900	158,000
	3)バイオプシー鉗子 502-302	トノクラ	20ケ	55,000	1,100,000
	4)バイオプシー鉗子 502-300L	トノクラ	20ケ	55,000	1,100,000
	合計				¥ 36,400,000
	消費税				¥ 1,092,000
	総合計				¥ 37,492,000

現地調達機材リスト

No.	機材名	仕様（参考銘柄）	数量	単価	計
I	医療機材				
1	レントゲン現像機	QX-130（KONICA）	1	US$ 10,198	Gs12,207,006
2	バックアップ装置	FAI（TRAFOPAR）	4	Gs 1,580,000	Gs 6,320,000
3	スタビライザー	SET（TRAFOPAR）	2	Gs 402,000	Gs 804,000
4	オートクレーブ	14-487-1（FISHER）	1	US$ 7,492.70	Gs 8,968,761.90
II	試薬、医療器具（別添リスト、見積書参照）				
1	試薬	CIENTIFICA PARAGUAYA（SIGMA）		US$ 15,410.32	Gs18,446,153
2	医療器具	CIENTIFICA PARAGUAYA（CMS）		US$ 7,537.70	Gs 9,022,626.90
III	事務備品				
1	事務机（大）	107 (SILVESTRI)	2	Gs 330,400	Gs 660,800
2	事務机（小）	104 〃	10	Gs 228,000	Gs 2,280,000
3	椅子（大）	205 〃	2	Gs 345,000	Gs 690,000
4	椅子（小）	206 〃	10	Gs 219,500	Gs 2,195,000
5	収納棚（引戸）	120A 〃	5	Gs 192,500	Gs 962,500
6	本棚	123 〃	5	Gs 87,300	Gs 436,500
7	キャビネット	119A 〃	5	Gs 175,000	Gs 875,000
8	本棚（ガラス戸）	126 〃	5	Gs 141,000	Gs 705,000
9	本棚（開き戸）	125 〃	5	Gs 162,000	Gs 810,000
10	実験台	(3.84x1.20x0.90) 〃	1	Gs 1,870,500	Gs 1,870,500
11	収納棚	(3.84x0.32x0.90) 〃	2	Gs 196,300	Gs 392,600
12	L字型実験台	(1.45x2.44x0.90) 〃	1	Gs 388,500	Gs 388,500
13	L字型試薬棚	(1.45x2.44x0.60) 〃	1	Gs 262,000	Gs 262,000
14	キャビネット	(0.50x0.50x0.84) 〃	4	Gs 304,000	Gs 1,216,000
15	実験台用椅子	〃	8	Gs 175,000	Gs 1,400,000
16	収納棚	(1.10x1.80x0.42) 〃	1	Gs 195,000	Gs 195,000
17	実験動物用棚	(1.00x0.32x1.80) 〃	3	Gs 92,500	Gs 277,500

合 計 Gs71,385,447.80

＝ ¥ 9,243,729.70

（8月統制レート 1US$ ＝ Gs1.197 ＝ ¥ 155）

試薬リスト

No.	品番	品　　名	数量	単　価	計
1	R2627	Restriction Endonuclease Ecor I	4	100.48	491.92
2	R5628	Restriction Endonuclease HAE Ⅲ	2	64.79	129.58
3	R6003	Restriction Endonuclease HINF I	4	80.41	321.64
4	R0629	Restriction Endonuclease HPA Ⅱ	4	89.32	357.28
5	R4506	Restriction Endonuclease MSP I	2	111.65	223.30
6	R5378	Restriction Endonuclease BAMH I	2	261.25	522.50
7	R6377	Restriction Endonuclease BGL Ⅱ	2	113.90	227.80
8	R4253	Restriction Endonuclease BSTE Ⅱ	2	69.24	138.48
9	R1882	Restriction Endonuclease HIND Ⅲ	2	185.35	370.70
10	R8507	Restriction Endonuclease HPA I	2	125.07	250.14
11	R1258	Restriction Endonuclease KPN I	2	129.58	259.16
12	R2007	Restriction Endonuclease PST I	2	131.78	263.56
13	R2631	Restriction Endonuclease PVU Ⅱ	2	105.05	210.10
14	R0754	Restriction Endonuclease SAL I	4	102.74	410.96
15	R4503	Restriction Endonuclease SMA I	4	91.63	366.52
16	R6379	Restriction Endonuclease XHO I	2	100.64	201.28
17	D2164	DNA Polymerase I	2	169.73	339.46
18	R9005	Ribonuclease A	2	29.04	58.08
19	D2886	T4 DNA Ligase	2	247.83	495.66
20	D4527	Deoxyribonuclease I	2	129.58	259.16
21	D6500	Deoxyadenoside 5-Triphosphate	2	200.97	401.94
22	D4760	Deoxycytidine 5-Triphosphate	2	277.81	555.62
23	D4135	Deoxyguanoside 5-Triphosphate	2	267.96	535.92
24	T8635	Thymidine 5-Triphosphate	4	133.98	535.92
25	A5394	Adenosine 5-Triphosphate	10	35.75	357.50
26	L6876	Lysozime Grade I	2	484.55	969.10
27	P4914	Protease Type XXV Ⅲ	2	80.41	160.82
28	D9780	Lamb DNA HIND Ⅲ Digest	2	55.88	111.76
29	A3284	Adenosine 5-Triphosphate	2	49.17	98.34
30	A5132	Ammonium Sulfate Grade I	4	42.46	169.84
31	A6387	Ammonium Sulfate Grade Ⅲ	2	79.75	159.50
32	B9508	Ethanolamine Free Base	4	40.26	161.04
33	G7126	Glycine Free Base	6	44.66	267.96
34	S8875	Sodium Bicarbonate	4	44.66	178.64
35	G7757	Glycerol	2	31.35	62.70
36	G9012	Glycerol Sigma Grade	2	62.59	125.18
37	P1386	Propionic Acid	4	22.33	89.32
38	T1503	Trizma Base Reagent Grade	4	113.96	455.84
39	U1250	Urea	4	26.84	107.36
40	D5029	P-Dioxane-D8	10	53.57	535.70
41	P1644	Percoll	4	453.31	1,813.24
42	1077	Histopaque	4	145.20	580.80
43	E9385	Exposure Cass St/St	2	255.90	511.80
44	F9252	Folin Ciocal 500ml	2	77.50	155.00
45	T0656	1,3,4,6 Tet 500mg	2	89.70	179.40
46	P7626	Phenylmeth, 1gr.	2	24.50	49.00
47	T7254	NA-P-TOSYL 100mgr.	2	59.20	118.40
48	N3516	Nonidet P40 100ml.	2	32.70	65.40
			TOTAL :	US$	15,410.32

医療器具リスト

No.	数量	品番	品　　名	単　価	計
1	2	107-169	Cylinder 100ml. OB-550E	12.80	25.6
2	2	107-219	Cylinder 250ml. OB-550F	24.50	49.00
3	2	101-243	Cylinder 1000ml. OB-550H	65.30	130.6
4	2	097-15A	Flask 25ml. paq. x12 10-040B	50.80	101.6
5	2	097-162	Flask 50ml. paq. x12 10-040D	50.80	101.6
6	2	097-170	Flask 125ml. paq. x12 10-040D	50.80	101.6
7	2	097-212	Flask 500ml. paq. x6 10-040H	31.00	62.00
8	2	097-238	Flask 1000ml. paq. x6 10-040K	51.00	102.00
9	1	097-914	Stirrer Hot plate	403.00	403.00
10	1	342-550	Mixer	281.00	281.00
11	1	059-753	Red Blood Pipet	106.00	106.00
12	1	059-717	White Blood Pipet	106.00	106.00
13	3	235-259	Volumetric Pipet 1ml.	5.00	15.00
14	3	235-267	Volumetric Pipet 2ml.	5.00	15.00
15	3	235-291	Volumetric Pipet 5ml.	5.00	15.00
16	3	235-333	Volumetric Pipet 10ml.	5.00	15.00
17	5	265-94A	Serological Pipet 1ml.	8.40	42.00
18	5	265-945	Serological Pipet 2ml.	10.50	52.5
19	2	268-285	Serological Pipet 25ml.	19.40	38.8
20	5	265-947	Serological Pipet 10ml.	13.40	67.00
21	2	194-142	Safety pipet 13-631-51	12.30	24.6
22	2	274-332	Timer 06-659	34.70	69.4
23	2	234-526	Timer/Stopwatch 14-649-15	48.90	97.80
24	2	219-527	Stopwatch 14-647-50	114.30	228.60
25	5	102-19A	Counter 07-905	18.40	92.00
26	1	CAI/9	Nine Couting unit 02-670-1A	450.00	450.00
27	2	117-036	Tubos 15ml. (cja. x500)05-538-53D	290.00	580.00
28	2	198-879	Flask 250ml. (cja. x50)10-126-5	254.00	508.00
29	1	10-126-1B	50ml. Canted Neck Flask	496.00	496.00
30	2	08-757-16A	Corning Cell Wells Tissue	110.00	220.00
31	2	08-757-16B	Corning Cell Wells Tissue	122.00	244.00
32	2	07-757-156	Corning Cell Wells Tissue	134.00	268.00
33	2	05-538-53D	Sterile Polypropilene Tube 15ml.	246.00	492.00
34	2	13-671-108A	Large Tip Open. Serolog. Pip. 1ml.	82.00	164.00
35	2	13-671-108C	Large Tip Open. Serolog. Pip. 5ml.	96.00	192.00
36	2	13-671-108D	Large Tip Open. Serol. Pip. 10ml.	114.00	228.00
37	5	09-730-130	Acro LC Dispors. Filt. Assembly	166.00	830.00
38	5	05-529-1A	Nalgene Brand-Dak Ridge 10ml.	34.00	170.00
39	5	05-529-1D	Nalgene Brand-Oak Ridge 50ml.	48.00	240.00
40	5	11-405-1B	UV-Absorbing Goggle	22.00	113.00

TOTAL : US$ 7,537.70