



सत्यमेव जयते

1957

HAFFKINE INSTITUTE

ANNUAL REPORT
GOVERNMENT OF BOMBAY

THE HAFFKINE INSTITUTE, BOMBAY

MEMBERS OF THE STAFF

(As on 31st December 1957)

Director

Dr. H. I. Jhala, M.D. (Bom.), F.C.P.S. (Bom.), D.T.M. (Cal.).

Assistant Directors

Dr. A. K. Hazra, M.B.B.S. (Bom.), D.T.M. (Cal.), Assistant Director in-charge, department of Antitoxins and Sera.

Dr. C. V. Deliwala, M.Sc. (Bom.), Ph.D. (Bom.), Assistant Director in-charge, Department of Chemotherapy.

Dr. M. V. Sant, M.D. (Bom.), F.C.P.S. (Bom.), Assistant Director in-charge, Department of Clinical Pathology and Diagnostic Reagents.

Dr. P. J. Deoras, M.Sc. (Nagpur), Ph.D. (Durham), F.E.S.I., Assistant Director in-charge, Department of Entomology.

Dr. N. K. Dutta, M.B. (Cal.), D.Phil. (Oxon.), Assistant Director in-charge, Department of Pharmacology.

Assistant Director (post vacant)—Charge held by Dr. A. K. Hazra, M.B.B.S. (Bom.), D.T.M. (Cal.), Department of Vaccines.

Assistant Director (post vacant)—Charge held by Dr. H. I. Jhala, M.D. (Bom.), F.C.P.S. (Bom.), D.T.M. (Cal.), Department of Virus Diseases.

Dr. (Shrimati) Alamela Venkataraman, B.A. (Madras), M.Sc. (Madras), Ph.D. (Madras), Senior Biochemist, Biochemistry Section.

Dr. K. V. Kulkarni, M.B.B.S. (Bom.), Medical Officer, Blood Bank Section.

Dr. A. E. Kulkarni, G.B.V.C. (Bom.), Senior Veterinary Surgeon, Animal Laboratory.

Administrative and Auxilliary Sections

Shri P. K. Sholapurwalla, B.Sc. (Bom.), LL.B. (Bom.), Business Manager.

Shri Shankar Rao, Personal Assistant to the Director, Haffkine Institute.

Shri G. I. Vaishnav, B.A. (Bom.), Office Superintendent.

Shri B. O. Shah, B.Com. (Bom.), Cost Accounts Officer.

Shri T. V. Ranganathan, B.A. (Madras), Accounts Superintendent.

Shri N. Lakshminarayana, L.M.E. (Mysore), L.E.E. (Mysore), Maintenance Engineer.

Library

Shri K. N. Pai, B.Sc. (Bom.), B.T. (Bom.), S.T.C. (Bom.), Dip. Lib. (Bom.), Librarian.

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REPORT OF THE HAFFKINE INSTITUTE FOR THE YEAR 1957

I. GENERAL REVIEW

In general the activities of the Institute have increased in every direction. The Departments of the Institute are now recognised as independent teaching departments under the Bombay University Act and are now represented on University Boards of Studies in Chemistry, Zoology, Microbiology, Pathology, Pharmacology, Bio-chemistry in Science and Medical Faculties of the University.

The Institute was faced with an emergency by the outbreak of epidemic of Influenza in the month of May 1957. Production of Influenza vaccine from the virus of Asian Influenza strains was undertaken at the Instance of Government of India. The usual epidemics of cholera and sudden increase of demand for sulfa group of drugs during the influenza season caused heavy strain on the activities of the Haffkine Institute.

The demand for plague vaccine dwindled down; demand for sera, anti-toxins, sulpha drugs and vitamins was on increase. Full demand of sera was not possible to meet with. The Institute has been able to manufacture Hyperimmune Serum for prophylaxis of rabies, Triple Vaccine for Prophylaxis of Diphtheria, Whooping Cough and Tetanus, and the Influenza Vaccine. It is proposed to put these products on commercial lines in near future. The expansion of Pimpri Stud Farm has been sanctioned, yet, it will be sometime before it may be possible to catch up with the demands made on the Institute for supplies of biologicals.

The Chemotherapy Department filed a patent for methods of synthesis of N^1 -p-methylbenzenesulphonyl- N^3 -n-butyl-urea, a compound which is effective in the treatment of diabetes. Two processes have also been evolved for the synthesis of p-nitroacetophenone which promise to be of industrial application. Applications have been filed for taking patents for both these processes also.

The Institute is feeling short of space in several departments. Our activities are not regulated according to our own desire. They have to be regulated in response to the needs that arise elsewhere and many of which are unforeseen. This reference is both to the indents made for supplies to be effected and also to the tests that have to be carried out due to expansion of hospital or drug control programmes in the State. A saturation limit has been reached and only expansion will be possible by adoption of shift system in some sections. In case of some of the supplies it is found very difficult to cope up with total orders in the time limit of 24 to 48 hours from

receipt of the indent. It is, therefore, necessary that there should now be expansion of the laboratories and plants at this Institute. Too much of routine interferes with development of research plans and projects. Preoccupation of the staff with routine leaves little room for expanding research project. A solution may lie in the employment of more staff in regulating the production schedules by removing bottlenecks of physical limitations of storage and plants and by increasing the fellowship and exchange programmes, especially at post-doctoral level. Since the financial condition of the Institute is extremely sound and since the Institute can pay for its expansion itself, it is hoped that such expansion will be possible in a very short time.

Col. Amirchand Prize was awarded to Dr. N. K. Dutta and Shri M. K. Habbu for their joint work on "Experimental Cholera in infant rabbit; a method for chemotherapeutic investigations."

Dr. N. K. Dutta was asked to participate in the symposium on Cholera organised by the Indian Association of Pathologists at Lucknow and Director participated in the symposium on Influenza organised by Indian Medical Association at Bangalore. The Second Analysts' Conference was held at the Haffkine Institute and the Institute took prominent part both in playing as host as well as in organisation and deliberations of the conference. Institute collaborated in deliberation on Kysanur forest disease and the influenza epidemic. Institute was visited by some distinguished visitors from abroad during the year.

Dr. D. W. Soman, M.D., D.T.M.&H., D.P.H., retired on 5th November 1957 from Directorship after a long merited service at the Institute. Earlier he was in charge of Clinical Pathology and Virus Departments. He had an important role in development of the later departments and his contributions to research are outstanding in many fields of his work at the Institute. On his retirement, Dr. H. I. JHALA, M.D., F.C.P.S., D.T.M., took over the charge of the Institute from the same day.

The departmental reports will show that Institute fulfilled its functions of research, training, supplies and tests to its very best. The facilities and benefits of service of the Institute to the public and the state medically and monsterily, have continued with increasing tempo. The report has been modified to incorporate research notes and the division of the report into Part I and Part II has been discontinued.

II. DEPARTMENT OF ANTITOXINS AND SERA

GENERAL

During the year under review increasing demands for the products, mainly Tetanus Antitoxins and Antivenene were in evidence. The Health Insurance Scheme has been increasing its demands as anticipated. The department has gone ahead with the plan to produce Triple Vaccine for combined immunization against Tetanus, Diphtheria and Whooping Cough. A limited supply

of this would be available soon. Efforts to implement improved methods for concentration of sera are also being made, the progress being limited in view of heavy increase in routine production activities.

Academic Activities.—The department took part in the second analytical conference held at the Institute in April 1957. Kumari Kadaba, a post-graduate student in Science, continued her work on biochemical studies on venoms.

Training of Workers.—Dr. Sutas Guptarak of Thailand, a W. H. O. Fellow, was provided with facilities for training in large scale production of toxins, toxoids and antitoxins.

RESEARCH ACTIVITIES

The proteolytic activity of concentrated antiserum was determined using haemoglobin substrate as well as when it acted on its own serum proteins. With haemoglobin substrate, highest activity was found at pH 3.0 and at 8.5. When acting on serum proteins the optimum pHs were at pH 4.0 and pH 9.0.

The proteases in Krait venom were determined using denatured haemoglobin and gelatin as substrates. The activity on haemoglobin substrate was measured according to the procedure of Anson. The pH activity curve showed three peaks at pH 3.5, 5.2 and at 8.4. The pH activity curve with gelatin substrate using viscometric methods, showed peaks at pH 1.85, 4.3 and 7.7, thus indicating the presence of three proteases in Krait venom.

The action of EDTA, Heparin and Trypsin inhibitors on blood coagulating action of Russell's Viper venom was determined. With EDTA, it was found that it prevented in a concentration of 2 mg/ml. the coagulation of recalcified plasma in presence of Russell's Viper venom. Under the same conditions, Heparin in a concentration of 1 unit per ml. prevented coagulation. Trypsin inhibitors prepared from four different pulses had no action on blood coagulation in presence of Russell's Viper venom.

Effect of different ions on the proteolytic activity of Russell's Viper venom at pH 3.6 and pH 9.0 have been determined. Protease at pH 3.6 was inhibited by Ferric and Cupric ions while Cyanide activated it. At pH 9.0, Cadmium, Barium, Lead, Cyanide and Cupric ions inhibited activity while Nickel and Cobalt activated the protease.

A number of experiments were carried out to determine the antigenic response in animals to different components of a mixed antigen consisting of Diphtheria and Tetanus Toxoid and Pertussis Vaccine. For preparation of a mixed antigen, crude Diphtheria-Toxoid was purified by fractionation with Ammonia Alum and Ammonium Sulphate while Tetanus Toxoid was purified by Levin's method of Ammonium Sulphate fractionation. Pertussis Vaccine was prepared on experimental scale in fluid Casein Hydrolysate medium of Cohen and Wheeler. The medium was slightly modified. The

yeast dialysate which was not easily available was replaced by Bacto yeast extract. Standard strains of *H. pertussis* 41405 and 40103 were used for production of the vaccine. The immunogenic response to Tetanus and Diphtheria Toxoids was found to be satisfactory with purified toxoids. The response to tetanus toxoid when used in crude form was found to be lower and the mixed antigen failed to pass the minimum requirements for tetanus.

Various methods using different enzymes under different conditions of pH, temperature, etc., and thermo-coagulation under different conditions are recommended for the purification of antitoxic sera on a large scale. With a view to work out a suitable method for purification in our laboratory, work was undertaken. Preliminary experiments using methods of J. L. Delsal, *et al*, Harms, and Pope *et al*, and modifying some of these methods, it was observed that satisfactory recovery of diphtheria antitoxin could be achieved by using plasma of some individual horses as starting material. However, the yields were low with some lots of plasma pooled from different horses. Work is in progress with a view to overcome these difficulties and evolving a satisfactory method which would give consistent results in large scale production.

ROUTINE

The quantities of toxins and toxoids produced is given below :

Types of Toxins and Toxoids				Quantity in ml.
Tetanus Toxin	...	...	...	5,03,000
Diphtheria Toxin	...	...	...	1,90,700
Welchii Toxin	...	...	...	1,35,500
Oedematiens Toxin	...	...	...	19,000
Septique Toxin	...	...	...	25,000
Shiga Toxin	...	...	...	1,000
Tetanus Toxin converted to Toxoid	...	...	...	1,08,500
Diphtheria Toxin converted to Toxoid	...	...	...	93,200

The total volume of concentrated serum produced was 17,17,600 ml. as against 14,75,400 ml. in 1956. The quantities of the various sera produced are given below :

Types of Sera.				Quantity in ml.
1. Concentrated Sera—				
Tetanus Antitoxin	...	...	...	7,92,100
Diphtheria Antitoxin	...	...	...	3,78,900
Antivenene	...	...	...	2,93,200
Gas gangrene Antitoxin	...	...	...	2,46,400
Anti-rabic Sera	...	...	...	7,000
Total				17,17,600

Types of Sera.				Quantity in ml.
2. <i>Natural Sera</i> —				
Normal Horse Serum	...	...	...	23,500
Anti-plague Serum	...	...	...	35,700
Total				59,200

During the year, the department supplied 12,27,980 ml. of Tetanus Antitoxin, 1,79,872 ml. of Diphtheria Antitoxin, 2,92,960 ml. of Antivenene, 85,600 ml. of Gas gangrene Antitoxin and 2,060 ml. of Normal Horse Serum. Seventy-two thousand, four hundred and forty-five ml. of Tetanus Toxoid and 7,698 ml. of Diphtheria Toxoid were also issued.

III. DEPARTMENT OF CHEMOTHERAPY

GENERAL

Academic Activities.—Shri D. V. Mehta was enrolled as a research student for the post-graduate degree of the M.Sc. in Organic Chemistry.

Shri S. E. Mhasalkar, a technician in the department, who had registered as post-graduate student for M.Sc. degree in Organic Chemistry, continued the work on the synthesis of substituted propanediols and related compounds.

Shri C. K. Kamath, a research chemist for Messrs. Unichem Laboratories who was deputed to get training in the synthetic organic chemistry and general laboratory practice completed his training.

RESEARCH ACTIVITIES

The study of various alternative routes for the synthesis of Chloramphenicol and its intermediates resulted in finding out two new methods for the synthesis of p-nitroacetophenone, which promise to be of industrial importance. Applications have been filed for taking patents for both these processes. The first process has already been accepted. The work in connection with the synthesis of analogues and derivatives of Chloramphenicol is being continued.

Studies were undertaken to find out practical methods for the manufacture of N¹-p-methylbenzenesulphonyl-N³-n-butyl-urea, a compound that is effective (by oral route) in the treatment of diabetes. A new process for the preparation of this compound has been worked out which is economical and easy to operate as compared to the processes already described in the literature. A provisional patent specification has been filed for this process too.

A number of substituted propanediols have been synthesised for evaluating their tranquilizing properties.

ROUTINE

To increase the shelf-life of the tablets, sugar-coating was undertaken on an experimental basis. The regular supply of Vitamin A-D forte as well as Vitamin A forte tablets in coated form has been started during the year.

The department continued to manufacture and supply vitamin, sulpha drugs, quinine preparations and other drugs to the Government Hospitals, Dispensaries, Municipalities and Employees State Insurance Scheme. All the demands were fully met with.

The table below shows the products manufactured and issued :—

Serial No.	Products.	No. of Tablets issued.
1	Vitamin AD Tablets	55,67,250
2	Vitamin AD (Spl.) Tablets	1,36,000
3	Vitamin AD Forte Tablets	7,67,172
4	Vitamin A Forte Tablets	74,600
5	Vitamin BC Tablets	1,04,74,150
6	Vitamin BC-3 Tablets	9,49,450
7	Vitamin B ₁ Tablets	7,15,950
8	Vitamin B ₂ Tablets	1,14,900
9	Vitamin C Tablets	11,10,800
10	Acid Nicotinic Tablets	3,07,950
11	Nicotinamide Tablets	8,850
12	Multivitamin Tablets	59,22,134
13	Vitamin K Tablets	8,250
Total ...		2,61,57,456
1	Sulphathiazole Tablets	29,48,325
2	Sulphadiazine Tablets	58,98,975
3	Sulphaguanidine Tablets	13,57,575
4	Quinine Sulphate Tablets	5,24,650
5	Magnesium Trisilicate Tablets	42,900
Total ...		1,07,72,425
1	Sulpha Injectable	29 × 12 boxes.
2	Solapson Strong B. P. C. solution	59,850 ml.
3	Pemon	11,123 lbs.
4	Sulphathiazole Powder	45.5 lbs.

Preliminary investigations on the recovery of Quinine Sulphate from the sub-standard Quinine Tablets that was started on an experimental basis, has been continued.

The Analytical Section of the department carried out nearly 950 analyses during the year. This included the testing of raw materials, intermediates, and finished products. Micro-analyses of compounds synthesised for research were also undertaken.

IV. DEPARTMENT OF CLINICAL PATHOLOGY AND DIAGNOSTIC REAGENTS

GENERAL

Staff.—Kumari S. H. Aswani, B.Sc., joined the department as a Technician. Shri L. S. Krishnan, M.Sc., Technician, relinquished his post at the end of the year.

Academic Activities.—Kumari I. R. Ovalekar and Kumari M. C. Patel to work for research degree.

Training of Workers.—Dr. Isabel Ng from Phillipines worked for a short term as an observer.

Among those who took training in the department were: Shri B. Choudhary of Central Railway, Dr. M. K. Tambwekar, M.B.B.S. of D. G. Hospital, Sholapur, Shri K. Pillay of Messrs. Hindustan Lever (Private) Ltd., Shri H. V. Davey of Western Railway, Shri Narayanaswami Naidu of Southern Railway, Shri K. Subramaniam of Railway Hospital, Perambur and Kumari Piroja Gai, Bombay.

RESEARCH ACTIVITIES

Attempts to prepare a refined and stable antigen with a Nichol's stain of *Treponema pallidum* have been encouraging. Studies are in progress to improve the antigen and to carry out comparative studies with other S.T.S.

A study has been started on the various epidemiological factors of Salmonella infection in the white mice.

Since the problem of Salmonella food poisoning has attracted considerable attention lately, a study has been commenced to find out the state of Salmonella infection in Bombay City and the various Salmonella types involved in the same. Sixty-five human stools were examined. Most of these samples showed the presence of *B. proteus*. No Salmonella organism could be isolated.

ROUTINE

The department continued to (i) conduct bacteriological, serological, microscopic and biological tests on clinical materials obtained from government hospitals, private practitioners, and institutions; (ii) prepare, stock and issue diagnostic reagents and stains; (iii) carry out histological examinations of the biopsy material and report on pathological tissues; (iv) prepare autogenous vaccines and (v) conduct special tests such as Rideal Walker test, pregnancy test, etc.

General Bacteriological Work.—All essential tests for the diagnosis of infectious diseases such as Diphtheria, Cerebro-spinal fever, Enteric group of fevers, Typhus, Plague, Cholera, Tuberculosis (pulmonary), etc., were carried out free of charge.

The number of routine examinations carried out during the year are as shown under :—

I. *Serological Examinations*—

(i) Wassermann Test	...	...	...	3,071
(ii) Kahn Test	...	...	...	2,183
(iii) Triple Widal Test	...	...	...	2,728
(iv) Agglutination Test for—				
(a) Typhus	...	...	...	2,720
(b) Brucellosis	...	...	...	2,720
(c) Leptospirosis	...	...	...	43
(d) Paul-Bunnell Test	...	...	...	7

II. *Bacteriological and Biological Tests*—

(a) Clot culture for enteric group	...	...	2,713
(ii) Urine culture for pathogenic organisms	...	...	74
(iii) Throat Swabs for Diphtheria	...	...	693
(iv) Stools and rectal swabs for culture	...	...	199
(v) Sputum and body fluids for AFB by smear culture and animal inoculation tests.	...	...	870
(vi) Blood culture routine	...	...	31
(vii) Preparation of autogenous vaccine	...	...	40

III. *General Microscopic Examinations*—

(a) Blood films	...	...	...	153
(ii) Blood cell counts—				
(a) RBC	...	...	...	834
(b) WBC	...	...	...	1,194
(c) Differential count	...	...	...	1,523
(d) Haemoglobin	...	...	...	844
(iii) Bone Marrow	...	...	...	3
(iv) Semen	...	...	...	88
(v) Material for smear and examination (presence of pathogenic organisms like G. C., etc.)	...	...	...	23
(vi) Stool	...	...	...	1,116
(vii) Urine	...	...	...	797

IV. *Prothrombin Times Estimation* 12

V. *Pregnancy Test* 996

VI. *Rideal Walker Test* 55

The Government of Bombay Typhoid Scheme.—This was continued for the tenth year. Six hundred sixty-eight calls from private practitioners were attended to. As before, blood samples were collected for culture and triple Widal tests. All positive cases of typhoid group were reported to the

Executive Health Officer, Bombay Municipality, to enable him to take the necessary preventive measures at the earliest opportunity. Thirty-three per cent of the cases were positive. The largest number of calls were during the months of January, May, June and December. The higher incidence of the disease during December was corroborated by evidence of pollution of water supply in the City in Ward C.

The total number of samples examined for Widal test and clot culture were 2,728. Of these, Widal test was positive in 870 and culture in 135 (*B. typhosus* 123 and *B. paratyphosus* A-12).

Diagnostic Reagents Section.—The supply of various reagents to laboratories all over Bombay State and outside was continued. The demand for these reagents had been more.

The issues for the year are as under :—

(i) Killed suspensions for Widal and Weil-Felix Tests...	1,82,110 ml.
(ii) Blood Grouping Sera	15,498 ml.
(iii) High Titre Sera and Amboceptor	1,061 ml.
(iv) Kahn Antigen	8,590 ml.
(v) W. R. Antigen	400 ml.
(vi) Russell's Viper and Cobra venom solutions	3,312 ml.
(vii) Biological stains	2,71,750 ml.

Salmonella Reference Work begun on a small scale in 1956 has been developed further. A few samples of sera received for Salmonella typing (agglutination) were tested and reported.

Histopathological Section.—During the year under review, 2,659 pathological specimens, received from different hospitals in the State of Bombay and a few from outside the State were examined and reported upon. These included brain tissues from suspected rabid animals, neoplasms, gynaecological conditions and some rare and interesting pathological specimens. The total number of blocks cut were 4,246.

The interesting specimens received during the year were as follows :—

(i) The diagnosis of Ewing's Sarcoma (Ewing's Tumour) was suggested in a specimen of bone biopsy of a vertebra.

(ii) In one lymph node specimen "secondary deposits from papillary carcinoma" from thyroid was reported.

(iii) One tumour from sacral region showed the features of a "Teratoma".

(iv) A specimen of a cyst from the eyelid was diagnosed as "Sweet gland carcinoma". Another cyst from the eye was reported as "Sweet gland carcinoma".

(v) One specimen of a nodule from anterior vaginal wall showed the presence of metastatic deposit of "Ochorio Carcinoma." The chest röntgenogram of this patient also showed the presence of metastases.

(vi) A biopsy specimen from a laryngeal growth from a boy aged 9 years showed the characters of "An Epidermoid Carcinoma".

(vii) In a specimen of total hysterectomy from a female aged 25 years, complaining of menorrhagia, the section of the ovary revealed the histological characters of "Struma Ovarii" (Thyroid tumour of ovary).

(viii) Specimen of a skin biopsy from right elbow of a male aged 42 years, with a history of recurring neuralgic pain, showed the features of an "Intradermal naevus".

V. DEPARTMENT OF ENTOMOLOGY

GENERAL

Academic Activities.—The department took part in a Symposium on "The Significance of Zoological Studies in Medical Research Programme of National Development" during the Indian Science Congress meetings at Calcutta.

The University of Bombay admitted Shri M. S. Gokhale to the Ph.D. degree on his work on "The Studies on Bombay rats". Shri N. E. Vad, Kumari M. Savant and Kumari K. Patel continued studies for a M. Sc. degree. Shri A. K. Joshi continued to work for a Ph.D. degree.

Nearly 5,273 persons belonging to various medical, veterinary, science, arts and agriculture colleges, secondary schools and private individuals were given instructive lectures on Snakes with demonstrations. 31 officers of the department of Public Health, Government of Bombay and Bombay Municipality attended Classes on Plague preventive measures.

Training of Workers.—Mr. Wang Lu-chi was deputed by the Chinese Government for training on Snakes. Mr. Khin Manrg Gyi of Burma and Mr. Abdul Karim Nayil of Afganistan, W. H. O. Fellows, were given training in Medical Entomology.

Research activities.—The proventricular teeth of *X. cheopis*, *X. astia*, *C. Canis*, *C. Felis* and *E. gallinacea* was worked out. It was noted that the females of all the fleas have nearly double the number of teeth as compared to the males. There was a certain amount of correlation between the density of the teeth in the proventriculus and the size of the papillae on the rectum. There were in all 4 types of teeth laid in varying number of rows, the outer-most being curved and pointed while the inner-most were blunt with flat raised sides.

The extensive bridding of *X. cheopis* and *X. astia* in the laboratory has shown that the sex ratio on emergence every month bore more or less the same pattern as was available in the collection of fleas for rats brought to this Institute in cages during different months of the year.

Observations conducted for a period of six months to know whether the rodents bred for studies in the animal house harboured any fleas, showed only one siphonaptera but revealed a large number of mites, lice and a few earwigs. The mites and lice have been identified by the British Museum as *Chirodiscoides caviae*, *Cyropus ovalis*, and *Gliricola porcelli*.

Studies were started in the snake farm moat to find out whether there were any insect predators of anophelene larvae that might supplement the gambusia fishes in controlling them. It was observed that apart from a number of insects in the moat, there were Dragon fly nymphs which kept the anophelene population down. This Dragon fly was identified as *Bradinaphyga geminata*.

The work on the morphology of *Echis carinatus* was completed. These studies were confined to the external morphology, digestive, circulatory, excretory, reproductive and nervous systems of the reptile.

Observations were made on the output of venom in snakes kept in the room as compared to those in the Snake Farm. The results show that in Cobra and Krait it was the males that gave, on an average, more venom when kept in the room and in the Farm, while in the case of Russell's vipers it was the female.

The copulation in the case of *Natrix piscator* was observed in the month of September. Cloacal smears of the females showed the presence of spermatozoa in the months of September, February, May and June. The egg laying was done in March and the hatching by the end of April. It was concluded that there might be some hibernation of spermatozoa in the female snakes.

Experiments on the susceptibility of Bombay City rats to Plague infection by injecting a standard test infective dose showed the following results :—

Species of rats.				No. of rats used in experiments.	No. of rats dead.	Mortality per cent.
R. rattus	...	...	...	83	9	10.8
R. norvegicus	...	...	...	64	3	4.6
B. bengalensis	...	...	...	82	64	78.0

ROUTINE

This department rendered services to Government, Local Bodies and private individuals in advising them on rats, rat control measures, rat trap designs, insecticides, supply and identification of insects of medical importance with particular reference to fleas.

Thirty cases were referred to this department for identification of snakes. Total number of snakes handled for collection of venom during the year was Cobra 33, Russell's viper 39, Krait 34 and *Echis* 2,111. The following snakes were recorded *Boiga trigonata* from Kutch, *Typhlops* sp. from Bombay, *Vropeltis* sp. from Lonavala and a subspecies of *Echis carinatus* from Kutch.

Venom collection during the year showed that the average yield per snake for Cobra was 0·1,782 g; Russell's viper 0·1,149 g.; Krait 0·0,153 g.; and Echis 0·0,024 g. The total venom produced and supplied during the year is given below :—

					Venom produced in g.	Venom supplied in g.
Cobra	...	...	...	...	83·9513	40·1328
R. Viper	...	...	...	...	41·4934	40·7019
Krait	...	...	...	...	6·1253	16·0763
Echis	...	...	...	...	13·3535	15·7690
Total					144·9235	112·6800

During the year 10,17,922 rats trapped alive and 62 dead rats were received at this Institute from the various Wards of Bombay City. 29 dead rats, 34,539 live rats from the City and 8,538 live rats from the Bombay Port Trust area were examined during the year. These rats were examined for plague infection. No such infection was detected. The detailed classification of all these rats according to species during the year is given below :—

Name of rat.		Rats trapped alive.	Rats found dead.	Rats from Bombay Port Trust Docks.
R. Rattus rufescens	...	2,23,068	15	3,896
R. norvegicus	...	1,76,095		1,538
B. bengalensis	...	4,78,736	12	2,744
B. malabarica	...	4,325		26
Plus dubius	...	37,933	2	356
Suncus caeruleus	...	91,904		407
Unidentified	...	861	33	
Total		10,17,922	63	8,967

VI. DEPARTMENT OF PHARMACOLOGY

GENERAL

Academic Activities.—The Indian Council of Medical Research established as Indigenous Drug Research Unit under Dr. N. K. Dutta. Dr. S. D. Vaidya, M.B.B.S., joined as a Unichem Research Scholar. Shri M. S. Sastri, M.Sc. continued as a Unichem Research Fellow.

The Second Analysts' Conference was held at this Institute from the 16th to 18th April 1957. This conference was sponsored by the Government of India to discuss on problems arising out of drug analysis. Shri M. S. Kannamwar, Minister for Health, Government of Bombay inaugurated the Conference. About one hundred delegates representing Central and State Governments, University Laboratories, private pharmaceutical firms and analytical laboratories, attended the Conference on invitation. There were five sectional meetings, e.g. pharmacology, pharmacognosy, pharmaceutical chemistry, biochemistry and bacteriology. The committee besides other, adopted several resolutions related to the procedure to be adopted for collaborative studies for fixing standards for crude drugs, spirituous medicinal preparations, shelf-life of complex vitamin preparations, toxicity of certain drugs, methods of standardization of vaccines, antitoxins, etc., and on matters relating to the uniformity of the methods to be adopted and in the manner of reporting.

Training of Workers.—Shri A. D. Deodhar and Shri G. K. Kamat representing Messrs. Pharma Search Laboratory, Bombay, Shri M. S. Jain of the University of Saugar, and Kumari Suman G. Kundaji were trained in the methods of microbiological assay of vitamins. Kumari I. F. Cooper took training in pharmacology and biological assay of drugs. Shri A. V. Jayakar of Messrs. Idomax Chemical, South Satara, undertook training on the assay of antibiotics and other preparations. Shri V. C. Khanderia of Messrs. International Franchise Ltd., spent some time at this department in learning the technique of bacteriological testing of parenteral solutions. Shri Shivaram, Sanitex Chemicals, Baroda, took training in the microbiological testing of drugs. Dr. S. W. Bible, W. H. O., Fellow in Pharmacology from Ceylon spent a short time in the department.

RESEARCH ACTIVITIES

Using the method of Dutta and Habbu (1955) for evaluation of drugs against cholera, it was shown that neomycin, one of the recently discovered antibiotics could prevent the death of infant rabbits suffering from cholera, if the drug is given orally at a fairly early stage of the infection.

Preliminary study of the stems and roots of *Aristolochia bracteata* showed that the aqueous and alcoholic extracts exhibited considerable uterine contractions. This property is not due to the alkaloid present in the plant. The defatted roots and stems on extraction with solvent ether yielded iso-Aristochoic acid, $C_{17}H_7O_7N$.

The optimum conditions for the development and measurement of colour using Mandelin's reaction for strychnine have been worked out. The Beer-Lambert's law is obeyed over a wide range for mixtures of pure quinine and strychnine (100 : 1) at the optimum wave length of 500 mμ and not at 530 mμ as reported earlier. This method was applied satisfactorily for the estimation of strychnine in Easton's Syrup samples, which had been stored for some time.

Teniospora Cordifolia, one of the indigenous drugs was studied pharmacognostically; microscopical examinations showed that septate lignified fibres from the pericycle and bordered pitted vessels and tracheids from xylem form the important diagnostic characters. Fluorescence of ether extract of the drug also helped rapid identification.

Comparative chemical studies on the two varieties of *Alpinia galanga* namely China and Malabar varieties were undertaken. With regard to the total ash, presence of essential oil, petroleum ether and hot water extractives, the chemical constituents are more or less identical, but it was significant to note that the Malabar variety of *Alpinia galanga* showed the complete absence of flavone bodies, namely campheride, galangin and alpinin whereas the China variety contained 0.3 per cent. of the flavone bodies.

ROUTINE

During the year 3,735 samples were tested. This covered a wide variety of drugs such as pharmacopoeial preparations, patent and proprietary medicines, indigenous drugs, fine chemicals, surgical dressings, glass wares, etc. Samples were received under the Drugs Act from Government departments, other State Governments and Commercial firms.

Out of the 3,735 samples tested, 907 were found to be not of standard quality which worked out to be about 24 per cent of the total. This is much below the figure of the previous year which stood at about 37 per cent.

Under the Drugs Act, 2,233 samples were analysed. Out of these, 699 were declared to be not of standard quality which is 31 per cent of the total samples tested. During the corresponding period of the previous year the percentage of the non-standard drugs amounted to about 39 per cent. The quality of the products showed an improvement.

"Biological and special products" and "other special products" tested during the year showed a slight rise, the number being 1,753, as compared to 1,315 during 1956. It nearly formed 47 per cent of the total samples examined. The percentages of the nonstandard drugs falling within these categories in 1957 and 1956 were 21 and 30 per cent. respectively.

Pharmacology section.—Twenty six samples, mostly from private manufacturing firms, of Crude Liver Extract were tested for the presence of histamine or histamine-like substances.

The depressor substances present in the various samples were between 13 ug to 300 ug in normal and about 3 ug to 260 ug per ml. in the atropinised cats when estimated in terms of histamine base. It may be possible that the reactions encountered in clinical cases following the Injection of Liver Extracts are due to the excessive amount of histamine or histamine-like substances present in some of the preparations.

Out of 15 samples of Tr. Scillae and a few samples of Tr. Urginea biologically assayed using the guinea-pigs method, 5 did not show any glyco-cidal activity.

Out of the 29 samples of Adrenaline examined only one was found to be not of standard quality.

One hundred and three samples were examined for their abnormal side effects or "safety tests". They included products like Chloramphenicol, Penicillin, Streptomycin, Dihydrostreptomycin, Liver Extract, Protein Hydrolysate, Neoarsphenamine and Vaccines, etc.

It was observed that the speed of injection in determining the presence or absence of undue toxicity in mice in the case of Procaine Penicillin is an important factor. It may be that a sample may not pass U. S. P. safety test but it may conform to B. P. as the former specifies that injections must be completed within five seconds.

Five hundred and thirty two samples of parenteral solutions were examined for presence of pyrogenic substances. Only 4 samples showed positive pyrogenic reaction. It is interesting to note that 8 samples stated to be Ayurvedic preparations were examined for the presence of pyrogenic substances. We are not aware that there are injectables in Ayurvedic System of Medicine.

Chemistry Section.—There are several substances which have proved to be unstable. The most notable one is the Liq. Ext. of Ergot. One should seriously consider whether any advantage is gained by keeping such preparations in the market where suitable facilities for storage do not exist.

There are several instances where genuine ingredients in a preparation were missing or present in lesser quantities. For instance, out of the 62 samples of aspirin dispensed in combination with other ingredients, 24 were found to be of not of standard quality. In some of them only acetylsalicylic acid was present while the other ingredients like caffeine, phenacetin or quinine were absent.

In the case of "Gripe Mixtures" it was observed that about one-third of the total samples tested, failed for one reason or the other. In some cases saccharin was substituted for sugar though not mentioned on the label. In others, one or more labelled ingredients like sodium bicarbonate was either more or less than the stated quantities.

It is not out of place to mention here that 8 samples out of 15 of para-amino salicylic acid or its salts failed to comply with B. P. C. limits for the content of meta-amino phenol, clarity and colour of the solution.

Forty-six samples of "Water for Injection" were tested, 15 of them were declared to be not of standard quality. The most important cause of failure was the presence of foreign matter, though a few samples failed on account of sterility and limit for pyrogenic substances.

Out of 12 samples of Chlorodyne, 6 did not comply with B. P. C. requirements for "weight per ml." or "content of chloroform", or both.

Seven samples of Tinct. of Rauwolfia Serpentina were tested for their alkaloidal content, but surprisingly none of them gave test for the presence of the same.

About 50 per cent of these appeared to be alcoholic preparations containing lower percentages of active ingredients or simple alcohol to which a little colour or a flavouring agent had been added. For instance, about half the number of samples of Tinct. Iodi Mitis, did not contain either iodine or potassium iodide or their contents were much less than the prescribed limit. Only 1 out of the 12 samples of Tinct. Senega tested could be considered to be of standard quality.

Eleven samples of "Ganja" (*Cannabis Sativa*) were tested pharmacognostically. They satisfied the I. P. requirements with the exception of "ash" and "alcohol soluble extractives".

Twelve samples of "Balamrit" were analysed for their total hypophosphite content and calcium hypophosphite content. Twenty five per cent of these samples showed the absence of hypophosphite.

Out of 29 samples of absorbent cotton, 13 failed to comply with pharmacopoeial requirements for absorbency, water soluble extractive or/and ash.

Biochemical Section.—Out of the total 943 vitamin preparations examined as many as 451, i.e., 48 per cent of them failed to confirm to the claims made on the label.

One hundred and twenty-five preparations containing vitamin A were analysed. These preparations consisted of Cod-Liver oil, Shark-liver oil, and water soluble vitamin-A. In 50 samples, the vitamin contents did not agree with the claim made. In many of them the potency was low.

Out of the 541 samples which were examined for vitamin B₁, 175 were of non-standard quality. In many of the failed samples the vitamin content was as low as 50 per cent. and in a few cases it was even less.

As many as 220 samples containing vitamin-C were examined. Eighty-three samples were of non-standard quality. In most of them the potency had deteriorated. About 43 per cent. of the samples of Folic Acid showed considerable deterioration. In nearly 65 per cent of the samples containing Pantothenic acid (or its salts), the amount found did not comply with the stated quantities. Thus, it will be observed that the vitamins B₁, C, Folic and Pantothenic acid do not remain stable in oral preparations. It is necessary that these vitamins should be fortified by adding excess quantities in order to counteract the loss of potency due to ageing.

One hundred and ninety-two samples were tested for vitamin B₁₂ activity. Sixty-six per cent of these retained their full potency while in others deterioration was evident. Some samples showed higher potency and in these, no doubt, excess amount of vitamin was added to counteract the loss of potency.

Eleven samples were tested for the proteolytic activity of Pepsin. In most of the cases the enzyme was mixed with other vitamins. Fifty per cent of them showed low proteolytic activity.

Six samples of Insulin were biologically standardised using rabbit sugar method. All of them were found to be of standard quality.

During the year, as many as 250 antibiotic preparations were tested. This is more than double the number examined in 1956. Both bacteriological and chemical methods were followed depending on the nature of the substances examined. The samples included Procaine Penicillin G., Penicillin G Sodium, Benzathine Penicillin, Streptomycin, Penicillin and Streptomycin Dihydrostreptomycin, Tetracycline, Chloramphenicol, Chloramphenicol Palmitate, Oxytetracycline, etc.

Out of the 148 penicillin preparations examined, 24 failed to comply with the required standards. It was observed that Penicillin Ointments, Lozenges and Tablets suffered deterioration. For example, nearly 45 per cent of Penicillin Lozenges were found not to comply with the claimed potency. One sample was devoid of any activity. In the case of Penicillin Ointments, about 32 per cent of the samples showed lower potency. It was interesting to note that in a few samples, the potency of Penicillin in terms of the total powder contained in a vial conformed to the labelled amount, but when it was calculated on the basis of units per milligram of the powder, the potency was found to be lower. The potency of penicillin in combination with streptomycin or dihydrostreptomycin was found to be fairly stable.

Six hundred and sixty-seven parenteral solutions were examined for sterility out of which only 4 were found to be non-sterile.

VII. DEPARTMENT OF VACCINES

GENERAL

Academic Activities.—The department took part in discussions on the standardization of Cholera vaccine in the Second Analysts' Conference held at the Institute.

Two members of the staff of this department obtained the M.Sc. Degree in Biochemistry by thesis.

Training of Workers.—Dr. Sutas Guptarak, W.H.O. Fellow from the Department of Medical Sciences of Thailand received training in the preparation and standardization of vaccines. Dr. Isabel Ng of Phillipines spent a short time in the department.

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RESEARCH ACTIVITIES

The antigenic complexity of the typhoid antigens and the antigenic relationship between various strains of *Salm. typhi*, *Salm. paratyphi* A, B and C, *Salm. enteritidis* and *Salm. ballerup* were studied by gel-diffusion technique in addition to the usual agglutination and precipitin tests. Results have shown that there were as many as 5 common components, between typhoid and paratyphoid organisms. The Vi antigens of *Salm. typhi*, *Salm. paratyphi* C and *Salm. ballerup* were found to be identical.

The effects of heat, dilute acid and dilute alkali on the Vi antigen of typhoid were studied. It was observed that heating at 65°C and at lower temperature liberated a large amount of Vi antigen in soluble form into the menstrum. At least four of the seven components of Vi antigen were found to be destroyed by heating at 80°C and 100°C for an hour. The acid treatment could inactivate the Vi antigens only partially but the alkali treatment inactivated them almost completely.

Extraction and fractionation of the Vi antigen of typhoid by various methods were studied. It could be seen from the results that freezing and thawing in the presence of 0.9 per cent. saline extracted in largest amount and the greatest number of components of Vi antigen. Diethylene glycol extracted a highly protective and almost homogeneous component of the Vi antigen.

The effect of heat on the antigenic complexity of the Cholera organism was studied by Ouchterlony's gel-diffusion technique in addition to precipitation and agglutination methods. The formation of a new antigenic component probably by degradation during heating was observed by gel-diffusion technique. This component remains in traces even when the suspension of the organism is heated to boiling under reflux.

Polysaccharide fractions were prepared from the strain 569B Inaba by phenol extraction, papain digestion and also from the growth in casein hydrolysate. The fractions were studied for their protective power; their antigenic complexity was studied by gel-diffusion method. It was observed that the fraction prepared by phenol extraction was more protective and antigenically more homogeneous as shown by gel-diffusion method.

Some of the common methods used in the extraction of antigens from the organisms such as extraction with saline, buffers, enzyme digestion and alternate freezing and thawing, were applied to extract antigens from *V. cholerae*. The extracts were studied by gel-diffusion technique and it was noticed that the freeze-thaw extract gave the maximum number of antigens.

H. pertussis medium (both solid and liquid) was prepared for the production of Triple Vaccine. The formulae was modified. The casein Hydrolysate was introduced as the base, instead of Bacto Casamino Acid. The medium was found to be satisfactory. Further work is under progress.

Several media recommended for leptospira were prepared Schuffner modification of Verwoort's medium was found most suitable. The phosphate precipitates interfered with growth; when these were eliminated there was considerable success.

ROUTINE

During the year under review, the demands for Plague, T.A.B. vaccines fell to a slightly lower level than that of the last year. However, there was a very heavy demand for Cholera vaccine due to the outbreak of epidemics all over the country.

9,40,645 ml. of plague vaccine, 170,54,514 ml. of Cholera vaccine and 2,21,795 ml. of T.A.B. vaccine were issued while 182,76,432 ml. of Cholera vaccine and 2,29,026 ml. of T.A.B. vaccine were produced during the year.

The total quantity of vaccine issued during the year was 182,17,354 ml. The demands for Plague vaccine were received mostly from Assam, Uttar Pradesh and Burma, those for cholera vaccine from Bombay, Madhya Pradesh, Mysore, Uttar Pradesh and Bihar and those for T.A.B. vaccines were mostly from Bombay State only.

Section of Public Health Chemistry and Bacteriology.—One hundred and six samples of water were examined bacteriologically and eighteen samples chemically. One sample of sewage effluent and one sample of milk powder were also examined bacteriologically.

Media Section.—During the year 21,560 litres of Casein Hydrolysate medium for Cholera vaccine and 1,767 litres of Casein Hydrolysate medium for Plague vaccine were prepared.

For T.A.B. vaccine, 1,693 Roux Bottles of 200 ml. agar medium each were supplied. Fifty thousand seven hundred eighty-nine bottles of 80 ml. each "sterility test" medium (aerobic and anaerobic) were supplied.

Parenteral Solution Section.—The parenteral solution section has continued to produce more and more and the production for this year excelled that of the previous ones. Below is the quantities of the injectables issued:—

Parenteral Solution.				Issued in Litres.
Glucose Saline	...	...	...	33,857
5 per cent. Glucose Solution	...	...	...	18,863
25 per cent. Glucose Solution	...	...	...	1,474
50 per cent. Glucose Solution	...	...	...	50
Normal Saline	...	...	...	4,888
Hypertonic Saline	...	...	...	7,977
Protein Hydrolysate	...	...	...	1,241
Sodium Sulphate	...	...	...	259
Sodium Citrate	...	...	...	249
Anti-Coagulant Solution	...	...	...	891
Distilled Water	...	...	...	1,557
Total				71,309

VIII. DEPARTMENT OF VIRUS DISEASES

GENERAL

Staff.—The charge of the vacant post of Assistant Director in charge Department of Virus Diseases was taken over by Dr. H. I. Jhala, Director, from Dr. D. W. Soman who retired with effect from 6th November 1957.

Academic Activities.—The department took part in the Symposium on Influenza organised by the Indian Medical Association at their annual conference held in Bangalore and in the one organised by the Bombay Hospital in Bombay, during the year.

Three students registered for Ph.D. degree in Microbiology and three were registered for the M.Sc. degree in the same subject.

One student obtained his M.Sc. degree in Microbiology. University of Bombay recognised this department as a separate independent teaching department in the subjects at this Institute under the Bombay University Act.

Kumari M. Savitri continued to work under Research Training Scholarship Scheme of Government of India on problems of APC viruses. Kumari R. Mendes continued to work as Government of Bombay Research Scholar on Influenza virus, and Kumari B. A. Ver continued to work as Donation Research Fellow on C. F. Test for the Rabies. Dr. B. K. Kathuria, Veterinary Institute, Mukteshwar, spent some time in the department.

Training of Workers.—Dr. M. V. Bapat continued to work at this Institute under ICMR/Rockefeller Foundation Fellowship during the year. Dr. Isabel Ng from Phillipines worked as an observer for a short term.

RESEARCH ACTIVITIES

During the year the department was granted a research inquiry by the Indian Council of Medical Research on the "Adaptation of Street Virus to developing chick embryos". This work was started in 1953 and was also continued in 1956. The method followed is one advocated by Komorov from Israel. Six strains of rabies "street virus" were isolated in mice; the strains are being serially passaged in mice prior to inoculation in eggs.

Studies on salivary glands of rabid animals were continued and out of 16 rabid animals whose brains and glands were made available by Sakarbai Veterinary Hospital, Bombay, there were only 3 cases where the salivary glands were negative though the brain was positive. The total number of salivary glands and brains tested during the year were 23.

The studies of C. F. Test in the diagnosis of rabies were continued under the "Donation Research Endowment". Out of 32 tests performed 25 were on brains and 7 were on salivary glands. Out of these 25 brains, 1 was of a cat and another of a human, while all the rest were dog brains. The results

obtained so far indicate that the C. F. test has a definite place in the diagnosis of rabies in animals, specifically for Negri negative specimens and salivary glands.

Studies on antigenic components of Semple's Vaccine and antibody components of human and animal sera obtained after the vaccination are being made with a view to study the responses and the components involved. Studies are being made with techniques such as electrophoresis, gel-diffusion, etc.

The Expert Committee of the I. C. M. R. on Virus Diseases had desired this Institute to carry out field trials on Hyperimmune Rabies Serum with the help of Government grants under the Second Five Year Plan. It has been possible to produce fourteen litres of horse hyperimmune serum and the same is being administered by field trials all over the State. So far only twenty-six patients have completed the therapy. The trials are being extended to more centres.

Studies on sensitivity of the isolated Asian strains to inhibitors are in progress. Refinements in vaccine production have been taken in hand. Trials of vaccines and sera are being conducted in man and animals with a view to evolve a method for biological standardization for these products.

ROUTINE

Antirabic Outpatient Clinic.—During the year 11,288 persons applied for antirabic treatment, while 7,288 of these completed the full course. Amongst these were 1,210 "advice cases". Nearly 2,790 cases discontinued treatment after starting it. There has been no report on hydrophobia amongst those treated at this Institute during the year.

Rabies Section.—Semple's Vaccine for human and canine treatment continued to be made available for supplies. The total number of centres receiving the supplies have increased from 191 to 215 and 133 of these are in Bombay State. Canine vaccine was issued to the extent of 19,968 ml. for prophylactic use. The total production of the said vaccine was 21,582 ml. in the year. A total of 28,30,714 ml. of antirabic vaccine was issued for human prophylaxis while 32,92,924 ml. were produced during the year. The demand for the vaccine has increased partly due to increased area now under the jurisdiction of the Bombay State. The Department has felt lot of strain in accommodating increased demand, as this was not anticipated.

The total number of human treatments for which this Institute was responsible during the year were 31,545. There were 8 cases of hydrophobia reported from different antirabic treatment centres during the year. The number of veterinary treatments supplied during the year amounted to 1,351. Following the new classification, the human treatments are classified according to the class of treatment as follows: Class I: 2,236; Class II (a) Child: 2,574; Class II (a) Adult and Class II (b) Child: 15,999 and Class

II (b) Adult: 10,736. In addition to the 133 centres in Bombay State which have been regularly taking supplies, 18 railway centres, 18 military centres and 46 centres outside the State have been given the supplies during the year.

The Institute continued to offer diagnostic facilities for rabies in animals. As many as 67 brain specimens were received at the Institute and of these 41 "positive". Besides microscopic and histological examinations, 33 biological tests were also carried out in mice for this purpose. It is regrettable that no results of examinations could be supplied for as many as 6 specimens as they did not arrive in a condition suitable for examination. Many of the specimens those have been examined were suitable only for histological studies.

Amongst the animals that have been reported to be rabid, there were 3 dogs, 1 buffalo, 2 cows, 1 goat and 6 human cases from post-mortem material. One of the latter cases had a clinical history of dog-bite 7 years ago. As hydrophobia is not likely to occur after such a long incubation period, it is possible that the history was misleading in some way and the deceased might have had a contact with a rabid animal without leaving any impression on his memory.

Influenza Section.—This is one of the laboratories in the World Health Organization network of influenza laboratories linked with the World Influenza Centre in London. In May 1957 a widespread epidemic of influenza broke out in this country and the Institute gave the help and technical advice to the Research Committee appointed by Government of Bombay. During the epidemic, 184 throat washings and 36 paired sera were examined, majority of these being from City Fever Hospital, Bombay. Few were also received from districts in Bombay State. The Government of India also decided to undertake manufacture of Influenza Vaccine and made special grants for staff and supply of eggs. The Institute was required to manufacture 21,000 doses of Influenza Vaccine from Asian A/57 strain and the target has been exceeded by another 9,000 doses. The staff was temporarily employed purely for this purpose. Additional space was also made available.

All the 14 isolated strains were sent to the World Influenza Centre, London, which confirmed them to be belonging to the Asian type-A. Two of these strains were in Q phase. One of the strain A/India/47/57 was later adopted for the preparation of Influenza Vaccine at the Institute.

The vaccine prepared was monovalent killed virus suspension on obtained from the allantoic fluid of influenza infected chick embryos, processed purified, concentrated and preserved with a titre of 1000 HA Units/cc. Strain A/Singapore/1/57 was also used in the initial stages for the preparation of the vaccine.

All the 35 paired sera collected from cases occurring in the members of the staff of the Institute were tested along with 37 samples received from acute phase cases and 201 convalescent samples from other sources. The H A and C.F. tests were used. There was four-fold increase of titre in the

convalescent sera over those of samples obtained from acute phase. Many of the convalescent sera also gave a positive C F Test. All strains isolated were tested against known strains and were confirmed to be all of Asian strain variety.

Yellow Fever Section.—Under this programme Yellow Fever Vaccine was stored and tested for its potency from time to time. A total of 9,181 inoculations were made to the outgoing passengers from this country; no case of post-vaccinal reactions was reported. Supplies of the potent vaccines were also made to 12 different centres in this country. Total number of potency tests performed were 13. Some of the vaccines have retained full potency even after 11 years of storage at 0-4° C.

Section of Virus Diseases other than Rabies, Influenza and Yellow Fever.—Rabbit and monkey kidney monolayer cultures were used for growing influenza and APC group of viruses. Attempts to isolate a new virus on rabbit kidney cells were successful. Antisera are being raised in the rabbits from the tissue culture fluids, strains are being locally isolated from removed tonsils and strain differences are being studied by comparing them with known strains obtained from abroad.

IX. BIOCHEMISTRY SECTION

GENERAL

Academic Activities.—Shri E. D. Bharucha, Technician in the department, has completed his research problem and has submitted the synopsis to the University of Bombay for the Master's Degree in Biochemistry.

Training of Workers.—Eight persons were given training for various periods in the department. Amongst these were Dr. M. K. Tambwekar, D. G. Hospital, Sholapur, Shri B. Chowdhary of Central Railway, Shri K. Subramanian of Railway Hospital, Perambur, Shri H. V. Davey of Western Railway, Shri Narayanswamy Naidu of Southern Railway and Kumar Piroja Gai of Bombay.

RESEARCH ACTIVITIES

Serum amylase activity in the blood of normal donors and laboratory workers using a standardised method has been completed. Wide ranges are usually reported for serum amylase values in normal cases. We have examined sixty five cases donors and laboratory workers for serum amylase activity. Soluble starch, washed free from traces of reducing materials, was used as substrate in phosphate buffer. After incubation, the reducing materials were estimated in terms of glucose, using Somogyi's copper reagent (J. Biol. Chem. 195, 19, 1952) and Nelson's arsenomolybdate reagent (J. Biol. Chem. 153, 517, 1944). The control values correspond to serum glucose levels. Eighty-five of the figures were within the range of 40-80 units per 100 ml. of serum. Only 5 per cent. were above 100 units, the average value being 62.

It is well known that blood sugar undergoes changes due to glycolytic processes and this glycolysis is supposed to be complete *in vitro* in about 4-6 hours at 37°C, all the glucose being converted into lactic acid. The rate of glycolysis, however, is variable in different samples and depends on the temperature and anticoagulant used. Although, a mixture of potassium oxalate and sodium fluoride is commonly used, sufficient emphasis is not made for the proper collection and preservation of blood samples for sugar estimations, particularly when the samples have to be sent to a distant laboratory for analysis. The problem was reinvestigated, to make sure that the concentration of sodium fluoride used was enough for satisfactory preservation over a certain specified period. The studies made so far indicate that even in the presence of fluoride, the samples should be kept in the cold and the estimation be carried out within four hours.

The study of the various serum protein fractions, including the alpha and beta lipoproteins using paper electrophoretic technique was continued during the year. The results indicate clearly that while there was a definite decrease in the albumin value, the gamma globulin fraction showed a corresponding increase. The ratio of the alpha to beta lipoproteins seemed to be reversed in all the cases of liver diseases studied.

ROUTINE

During the year under review, the department continued its activities of examination of clinical materials from hospitals and private clinics. The total number of analysis carried out increased to 6,957 as against 6,353 in the previous year. It is observed that the emphasis was more and more on quantitative blood chemistry, such as serum protein fractions, cholesterol, glucose, urea, calcium, phosphorus, and enzymes like amylase and acid and alkaline phosphatases. There were 4,717 samples of blood, 659 of urine and stools, 998 of cerebrospinal fluid and other body fluids and 339 milk samples. Two hundred and thirty-seven special tests were carried out.

It is hoped that the department will in future be in a position to undertake more specialised type of work. This would include assays of hormones like 17-ketosteroids, estrogens, adrenocortical steroids, enzymes like lipase, esterase, histaminase, oxidase, dehydrogenase and transaminases; vitamins, like Vitamin A, C and E and B and other estimations such as blood gases, protein-bound iodine, plasma iron and copper and paper electrophoretic studies for plasma proteins and various types of hemoglobins.

X. BLOOD BANK SECTION

GENERAL

The demands for dried human blood plasma increased and so also the demands for blood grouping sera. The Government has raised the payment to prisoners for donating blood from Rs. 3 to Rs. 5 and this might help in getting more blood donors from this source.

Training of Workers.—One Medical Officer and two Laboratory Technicians were trained in Blood Bank methodology during the year.

RESEARCH ACTIVITIES

Under the auspices of the Blood Group Reference Centre of the Indian Council of Medical Research, the preparation of standard Blood Grouping Sera was undertaken. Standard Anti-A and Anti-B were prepared. Anti-D (Rho) serum was prepared by immunising Rho negative donors by giving intravenous injections of Rh positive blood. Eight donors were immunised and four of them showed good titre of Anti-Nh antibodies. Anti-Rh serum is being prepared by absorbing redundant antibodies.

Experiments on the preparation of plasma fractions using Ion exchange Resin has been in progress.

ROUTINE.

Lyophilization Unit.—During the year under report, 4,500 samples of blood were collected for processing into dried plasma. Six hundred eighty-four litres of dried plasma were prepared. One hundred fifteen litres of plasma were distributed free and 454 litres were sold making a total of 570 litres. In addition, 31,781 ampoules of Antivenene (10 ml. each) were also lyophilised.

Two thousand six hundred eighty Kahn tests were performed. Out of these 26·23 per cent were positive, 446 samples were referred for Rh investigations. Thirty-six (8·07 per cent) were Rh negative. One thousand six hundred eighty-three voluntary blood donors from St. Georges' Hospital were tested for Rh state, of which forty-seven (2·79 per cent) were found to be negative.

XI. DEPARTMENT OF NUTRITION

GENERAL

The department forms an independent unit under Assistant Director Dr. M. V. Radhakrishna Rao, M.B., Ph.D., F.N.I. working for research in collaboration with Haffkine Institute.

RESEARCH ACTIVITIES

The beneficial effect of 'idli' feeding at 50 and 100 per cent replacement of dietary protein (casein) on experimentally produced fatty infiltration of the liver of rats fed high-fat low protein diet (30 per cent fat and 10 per cent casein) was reported before. In these tests, the composition of 'idli' consisted of four parts of black gram flour and one part of rice. This work was extended further in which the composition of 'idli' was changed to one part of black gram flour to two parts of rice, to be in conformity with the usual dietary practice.

Thirty-six weanling rats (male) of the Haffkine inbred strain weighing about 30 g. each were distributed into three groups of twelve animals each. The control group received a high-fat low protein diet. The second and the

third groups were given the 'idli' and the corresponding unfermented preparation, at 50 per cent replacement of the dietary protein (casein) on dry weight basis, from the start of the experiment. All the other experimental details were as reported earlier.

The experimental results showed that when the composition of 'idli' was one part of black gram to two parts of rice, there was no significant difference in the fat content of livers of animals of all the three groups.

XII. CRITICS CLUB

Under the auspices of the Critics Club, twenty-three Scientific Meetings were held during the year. The scientific staff of the different departments of this Institute presented original research work and reviews. There were four lectures by Guest speakers. The following is the list of subjects and speakers :-

Subject.	Speaker.
1. Newer Methods of Bacteriological Diagnosis.	Lt. Col. Oscar Felsenfeld, U. S. Army Medical Corps in Far East, Japan.
2. Major Medical Problems of the Philippines.	Dr. (Mrs.) Isabel Ng, Medical College, University of Philippines.
3. Phosphorylated Guanidines ...	Dr. A. H. Ennor, Dean, John Curtin School of Medical Research and professor of Biochemistry, University of Canberra (Australia).
4. Opportunities for Scientists in India.	Major General Sir S. S. Sokhey.
5. Diet and Life-span ...	Shri N. C. Shah.
6. Estimation of Serum Potassium.	Shri R. N. Joshi.
7. Synthesis of Chloramphenicol ...	Dr. K. D. Kulkarni.
8. Crime Investigation and Blood Examination.	Shri C. J. Karkhanis.
9. Recent Trends in Pregnancy Tests.	Dr. B. G. Mody.
10. Further Observations on the Complement Fixation Test as Applied to the Diagnosis of Rabies.	Shri Y. I. Sethna.
11. Heat Stability of Vi-antigens of Typhoid.	Shri R. S. Karbhari.
12. Pharmacognosy in the Study of Drugs.	Shri B. C. Mehta.

Subject.	Speaker.
13. Purification of Antitoxic Sera.	Dr. S. S. Rao.
14. Diet and Nutritional Survey Among the Dubla Community.	Dr. V. N. Jai and Shrimati Udas.
15. Observation on Feeding Mechanism of Fleas:	Shri A. K. Joshi.
16. A Brief Account of the Present Influenza Epidemic (Laboratory Aspects).	Shri Y. S. Nimbkar.
17. Chemistry of Viruses ...	Dr. (Shrimati) A. Venkataraman.
18. Psychosomatic Drugs as Anti-metabolites of Serotonin.	Shri R. A. Bellare.
19. The Wound Shock ...	Dr. K. V. Kulkarni.
20. Salmonellosis: The Problem of Salmonella and Food Poisoning.	Shri A. S. Narasinhani.
21. Antigenic Complexity of Salmonella.	Shri R. S. Karbhari.
22. Newer Antibiotics ...	Shri M. L. Chainani.
23. Proteases of Snake Venoms ...	Kumari L. S. Kadaba.

XIII. LIBRARY

GENERAL

Staff.—Shri K. N. Pai, B.Sc., B.T., S.T.C., Dip. Lib. took over the charge of the post of the Librarian from 10th December 1957. Kumari S. M. Charegaonkar, B.Sc., Dip. Lib. was appointed as an Assistant Librarian from the same date.

ROUTINE

The Library was kept open from 10 a.m. to 5-15 p.m. to all the readers interested in medical research. The members of the staff could read in the Library from 8-45 a.m. to 8 p.m. on all working days and 8-45 a.m. to 5 p.m. on Saturdays.

The stock of the Library comprised of 8,312 books and 16,000 Periodicals and reports. About 180 periodicals were on our list this year.

The Library worked in co-operation with 7 other Libraries of the scientific and medical institutions in the City under inter-library loan system. About 248 books were issued to other institutions from this library and about 58 books were procured for the members of our staff. About 1,264 readers from the different institutions in the City made use of the library facilities and 1,083 books were issued to the members of our staff.

Efforts are being made to increase the Library grant to enable us to get more new books and periodicals. The Library will be in a position to issue a Weekly Bulletin for the staff. This will include the classified list of articles published in the periodicals received every week and it will also introduce translation service in the near future.

XIV. THE ANIMAL LABORATORY

GENERAL

Training of Workers.—Shri B. C. Deole, Stockman from the Veterinary Biological Products, Bombay, received training in the management of laboratory animals.

ROUTINE

The following table shows the number of laboratory animals bred and issued during the year :—

Laboratory Animal.	No. Bred.	No. issued.
Rabbits	1,200	1,039
Guinea-pigs	1,007	989
White mice (H. I. strain)	18,191	16,036
White mice (Swiss strain)	1,827	1,563
Albino rats	2,457	2,319
Hamsters	69	36
Cotton rate	21	

Besides the animals mentioned in the above table following animals were maintained in the animal house and issued to various departments as and when required : Sheep 2,284, Dogs 4, Cats 198, Fowls 283, Pigeons 4, Monkeys 169 (includes monkeys maintained for Polio Research Unit of the I.C.M.R.). On an average 152 horses were maintained.

XV. BUSINESS MANAGEMENT

The productive activities of the Institute continued to expand and the sale of Institute's products reached a level of about Rs. 47,00,000 during the year under review.

Stores Section was separated from Purchase Section. The absence of the Stores Officer for a long time was keenly felt.

Throughout the year there was heavy pressure both on Purchase and Stores Sections due to increase in Institute's activities.

The Profit and Loss Accounts for the years 1955-56 and 1956-57, were worked out. During the former period the Institute had shown a net profit of Rupees 2,18,806 and during the later period it made a net profit of Rupees 17,71,351. The profit has been arrived at after accounting for all expenses like depreciation, interest, leave and pension reserve, reserve for doubtful debts, etc.

XVI. WORKSHOP SECTION

During the year the Section has established a workshop consisting of machineries like a lathe, drilling machines, shaping machines, grinding machines etc., in addition to a carpentry section which was already existing.

It helped the different departments of the Institute in the installation of new equipments, maintenance of all the Mechanical equipments apparatus etc., and designing and fabricating new machineries, cages, laboratory tables, cupboards, etc.

XVII. LIST OF VISITORS DURING 1957

Name of Visitor.	Date of Visit.
1. Academician Englehardt, <i>U. S. S. R., Moscow</i> ...	19th January 1957.
2. Prof. N. U. Sissanian, Institute of Biochemistry, Academic of Sciences of <i>U. S. S. R., Moscow</i> ...	24th January 1957.
3. Lt.-Col. Oscar Felsenfeld, <i>U. S. Army Medical Corps in Far East, Japan</i> ...	25th January 1957.
4. Prof. d'Antona, Technical Director in charge "Sclavo" Institute, <i>Siena, Italy</i> ...	19th February 1957.
5. Dr. Goldman, Naval Medical Research Institute, <i>Bethesda-14, U. S. A.</i> ...	19th March 1957.
6. Smt. Padmawati Devi, Minister of Public Health, <i>Madhya Pradesh, Bhopal</i> ...	21st August 1957.
7. Shri G. L. Sharma, Director, Health Services, <i>Madhya Pradesh, Indore</i> ...	21st August 1957.
8. Shri Y. B. Chavan, Chief Minister, <i>Bombay</i> ...	23rd August 1957.
9. Group of Doctors from <i>U. S. A.</i> ...	8th October 1957.
10. Dr. G. W. Rake, Scientific Director of Olin <i>Mathieson Chemical Corporation, U. S. A.</i> ...	17th October 1957.
11. Prof. E. H. Ennor, Dean, John Curtin School of Medical Research and Professor of Biochemistry at the Australian National University, <i>Canberra</i> ...	13th November 1957.
12. Dr. J. N. Quastel, Professor of Biochemistry, McGill University and Director, McGill Montreal General Hospital Research Institute, <i>Canada</i> ...	19th November 1957.
13. Prof. Dr. Ton-nhat-Tung, Vice Minister of Public Health and Secretary-General, Red Cross of the Democratic Republic of <i>Viet Nam Viet Nam</i> ...	19th November 1957.

Name of Visitor.	Date of Visit.
14. Dr. Nguyen-Van-Thu, Doctor of Dentistry and Vice-President of the Medical Association of the Democratic Republic of Viet Nam, <i>Viet Nam</i>	19th November 1957.
15. Dr. Khin Maung Win, Medical College, <i>Rangoon</i> .	27th November 1957.
16. Prof. Baubaug Soetarso, Faculty of Medicine, Gadjra Mada University, <i>Jogjakarta</i> ...	27th November 1957.
17. Dr. M. H. Soetrinso, Faculty of Medicine, University of Indonesia, <i>Djarkarta</i> ...	27th November 1957.
18. Dr. Oei Hong Hoei, Faculty of Medicine, University of Airlangga, <i>Surabaya</i> ...	27th November 1957.
19. Prof. Pirom Suvanatemiya, Chulalongkorn Medical School, <i>Bangkok</i> ...	27th November 1957.
20. Prof. Pradit Tansurat, Siriraj Medical School, <i>Bangkok</i>	27th November 1957.
21. Dr. Geoffrey Edsall, Walter Reed Army Institute of Research, <i>U. S. A.</i> ...	28th November 1957.
22. Mr. E. H. Bluescher, Walter Reed Army Institute of Research, <i>U. S. A.</i> ...	28th November 1957.

XVIII. LIST OF PAPERS PUBLISHED FOR THE YEAR 1957

1. Bhagwan, N. V., Nimbkar, Y. S. and Rao, S. S. Fractionation of Soluble Antigens of *Pasteurella pestis*. *Ind. J. Med. Res.* 45 : 1-8 ; 1957.
2. Dutta, N. K. and Thadani, K. C., Fibrin (horse) Hydrolysate for Parenteral Administration. *Ind. J. Med. Res.* 45 : 493-498 ; 1957.
3. Joshi, D. V., Tamhane, R. G. and Dutta, N. K. Chemical and Pharmacological Investigation of Roots of *Wagates Spicota Dalzell*. *Current Science*, 26 : 147-148 ; 1957.
4. Kulkarni, M. E. and Rao, S. S. Antigenic Composition of the Venoms of Poisonous Snakes in India. *Venoms* : Pub. No. 44. American Association for the Advancement of Science. Washington D.C. : 175-177 ; 1956.
5. Palande, B. N. A General Review on Tablet Manufacture, Part V. *Pharmaceutist*, 2 : 15-22 ; 1957.
6. Shanta S. Rao and Rao, S. S. Proteases of Cobra (*Naja naja*) Venom Venoms. Publication No. 44. American Association for the Advancement of Science, Washington, D. C., 16 BC : 148-151 ; 1956.

7. Shanta S. Rao and Rao S. S. Proteolytic Activity of Russell's viper Venom and Its Inhibition by Heparin. J.Sc. Ind. Res. 16C : 148-151 ; 1957.
8. Sant, M. V. Plasma Cell Mastitis (In Lactating Breast). Journal of Post Graduate Medicine, 3 : 93-98 ; 1957.
9. Soman, D. W. and Nimbkar, Y. S. Studies on Influenza. J. of J. J. Group of Hospitals and G. M. C. 2 : 277-287 ; 1957.
10. Soman, D. W. and Nimbkar, Y. S. Inactivation of Normal Serum Inhibitors. J. Ind. Med. Assn. 29 : 432-435 ; 1957.

XIX. GRANTS AND BENEFICIARIES.

1. *Indian Council of Medical Research, New Delhi*—
 - (i) Enquiry on "Adaptation of Rabies Street Virus Collected Locally, to Chick Embryo".
 - (ii) Investigation into field trials with Protein rich Foodstuffs.
 - (iii) Enquiry on "Nutritive Value of Foodstuffs."
 - (iv) Enquiry on the "Study of Indian Poisonous Snakes."
 - (v) Drug Research Unit.
 - (vi) A Fellowship on Bacteriological Training.
2. *Council of Scientific and Industrial Research, New Delhi*—
A Fellowship for "Isolation Study of Biologically Important Constituent of Venoms of Cobra and Russell's Viper".
3. *Government of India*—
A Research Scholarship for the Study of Tissue Culture Technique.
4. *Government of Bombay*—
 - (i) A Scholarship for the Study of "Influenza Virus".
 - (ii) A Scholarship for "Salmonella typing".
5. *Unichem Laboratory, Bombay*—
 - (i) A Scholarship for Research on "Indigenous Drugs".
 - (ii) A Scholarship for Investigation of Cholera.
6. Thanks are due to—
 - (i) Messrs. Alembic Chemical Works Co. Ltd., Baroda.
 - (ii) Dumex Private Ltd., Bombay.
 - (iii) Boots Pure Drug Co. (India) Private Ltd., Bombay.
 - (iv) Unichem Laboratories, Bombay.
 - (v) The Himalayan Drug Co. of Bombay and
 - (vi) Indian Pharmaceutical Association, Bombay State Branch.

for their kind co-operation during the Second Analysts' Conference held at this Institute in April 1957.

