	器機名（規格）	数量	単価（万円）	合計（万円）	
62	フロアーポリッシャー（予備ブラシ付）	2台	8	16	共用（1F、2F）
63	電気シャ断器（機器取り付用）（100V）	6	2.3	13.8	各室1ケ
64	真空ポンプ（中型）	2	6	12	V1、V2 各1 液換え用
65	試験管ミキサー	3台×5室	4	60	各室3台
66	マグネチックスターラー（大）	7	6	42	各室1台
67	〃（中）	7	4	28	〃 〃
68	〃（小）	7	3	21	〃 〃
69	連続分注機（手動）（ステンレス先） 1 mℓ用	14本	2	28	各室2本
70	〃 5 mℓ用	6	2	12	〃 1
71	〃 10 mℓ用	2	2	4	境地室
72	〃 0.5 mℓ用	4	2	8	V1,2,B1,B2,
73	ラボクイマー（警報式、1ha用）	12	1	12	各室2
74	ストップウオッチ	4	1.2	4.8	B1、B2各2
75	急速凝集反応器	2	7	14	B1、B2
76	滅菌缶ピペット 0.5, 1, 2, 5, 10 mℓ用各10本（ステンレス）	50本	0.47	23.5	共用
77	〃 〃 20 mℓ 用	10〃	0.48	4.8	〃
78	〃 60 mm シャーレ、角ビン用共通	25	2.5	62.5	〃
79	〃 90 mm シャーレ、ルービン用共通	25	2.5	62.5	〃
80	ピペット収納箱	6	2.2	13.2	各室1
81	簡易試験管洗浄機	3	7.5	22.5	洗浄室
82	超音波洗浄機（シャープ100V、600W）（50×40×26 cm（内寸）	1	67	6.7	〃
83	肉ひき機	1	2	2	〃
84	ステンレスポット（フタ付）5 ℓ	2	0.4	0.8	〃
85	〃 培地調整用 10 ℓ	2	0.7	1.4	〃
86	〃 〃 20 ℓ	2	1.4	2.8	〃
87	〃 〃 30 ℓ	2	1.8	3.6	〃
88	ガス湯沸し器（ガス用）	1	6	6	〃
89	消毒用滅菌缶（ステン、角型フタ付）25 ℓ	3ケ×4室	1.6	19.2	4室分
90	〃（丸型密ペイ型）	5	2	10.	動物舎5棟
91	ハンドカー（耐重量150 kg）	1	2	2	共用
92	嫌気培養用ジャー（小型）（シャーレ10枚用）	2	6.5	13	B2
93	ハンドバーナー（アンプル熔封用）	1	3	3	共用
94	マイクロタイター用ドロッパー（0.025 mℓ）	25本	0.4	10	〃
95	〃 ダイリューター（0.025 mℓ）	100〃	0.3	30	〃
				（小計 648.4）	

	器機名（規格）	数量	単価（万円）	合計（万円）	
96	ペニシリンカップ	3,000ヶ	0.02	60	抗生
97	〃 収納管（50ヶ入）	20	0.6	12	〃
98	水平台（ガラス板、5㎜、1m×1m）	4	2.5	10	〃
99	水準器	2	0.5	1	〃
100	ノギス	4	0.2	0.8	〃
101	定温恒温槽	1	28	28	〃
102	恒温振盪機	1	43	43	〃
103	除湿機	1	12.8	12.8	〃
104	ガスレンジ（2連）（タイマー、火力調節付実験室用）	9	1.9	17.1	実験用兼給湯
105	〃 （単）	5	0.4	2	湯沸し用
106	ガスバーナー（ブンゼン燈）	30ヶ	0.35	10.5	
107	数取器（カウンター）	12	0.15	1.8	各室2個
108	煮沸消毒器（ステンレス製）（大）	12	1.0	12	〃 〃
109	〃 （中）	12	0.5	6	〃 〃
110	〃 （小）	12	0.3	3.6	〃 〃
111	アルミ製弁当箱（大）ゴム栓、小試験管滅菌用	50	0.04	2	共用
112	ラボピペット（㎕60チップ1～200 μℓ）2箱付	1セット	3.7	3.7	〃
113	斜位型試験管立て（ステンレス製）	50	0.9	45	V1、V2
114	卵殻カッター	2	4.8	9.6	V1、V2
115	金網カゴ（ステンレス、角型、深、大）	10	1.3	13	共用、洗浄室
116	〃 （〃 〃 〃 中）	10	1.1	11	〃 〃
117	〃 （〃 〃 浅、小）	10	0.4	4	〃 〃
118	〃 （〃 丸型、深、大）	10	1.5	15	〃 〃
119	〃 （〃 〃 〃 中）	10	1.3	13	〃 〃
120	〃 （〃 〃 〃 小）	10	1.1	11	〃 〃
121	アイスボックス中型	6	0.4	2.4	各室1ヶ
122	ドライアイス製造機	1	10	10	共用
123	オーバーヘッドプロジェクター	1	2	2	〃
124	35㎜スライドプロジェクター（自動送り、30枚入り）	2	5	10	〃
125	ビデオテレビセット	1	50	50	研修用（共用）
126	電動タイプライター（IBM） エレメント10種付	1	45	45	共用
127	〃 （オリベッティ）	2	15	30	〃
128	卓上電算機（パソコン） モニター、プリンター付	1	50	50	〃
129	16㎜プロジェクター	1	50	50	研修用
				（小計 597.3万円）	

Aランク大型備品類　　6,801万円

〃　中・小型備品類　　4,922万円（3.150+526.3+648.4+597.3＝）

Bランク大型備品類　　1,320万円

備品等器機類小計　　13,043万円

動物ケージ等（含消毒槽等）　約　900万円

計　　13,943万円

消耗品類（試薬、医用雑品、
雑品、ガラス器具等）　年約　1,500万円（日本のVet.Assay Lab.2,240万円／年）

卵、動物（兎、犬）購入費　年約　3,000万円（　〃　4,130万円／年）

（鶏、小動物等、水洗型架台（　約　6,400万円　）
台）
（鶏用、モル、兎用の一部
はケージ付）

（付属資料7. インドネシア政府要請の動物医薬品検査所設立計画）

TERM OF REFERENCE

THE ESTABLISHMENT OF ASSAY LABORATORY

I. Background and supporting information.

1. Justification of the Project.

Total agricultural production in Indonesia comprises about 27 percent contribution of its Gross Domestic Production, and livestock production shares approximately 8 percent of the total agricultural production. There is an urgent need to promote livestock industry in the country to meet the growing demand of animal protein diet, resource development, small - holder farms improvement etc. Such kinds of development schemes as poultry, dairy and beef cattle production promotion plans at national and/or local level have been launched so far. Animal health services are one of the fundamental principles to protect animal ressources, to develop animal industry and also to increase the production of animal protein.
The economic losses caused by various kinds of animal diseases as well as low productivity of livestock are the main obstacle to promote livestock industry in the country.
Haemorrhagic Septicemia, Rabies, Surra, Newcastle disease and other parasitic diseases have been reported, and animals have been vaccinated against certain infectious diseases with vaccine produced in the country itself. Moreover the amount of vaccines and vet. drugs produced is still insufficient to protect domestic animals from infectious disease.

To cope with the requirements of biological products and veterinary drugs a lot of vaccines, sera, biological diagnostics and veterinary drugs was imported from abroad. The number of such importation is getting bigger and bigger in recent years in accordance with the increasing livestock production.
Besides the production capacity of vaccine, biological products and veterinary drugs of the existing pharmaceutical industries is also increasing.
According to recent data there are 51 kind of vaccines, sera and biological diagnostics for veterinary use and 895 kind of veterinary drugs both local production and imported from aboard.

A new .....

A new method of FMD vaccine that is the tissue culture method has been developed in the Centre for Veterinary Biologics Surabaya with the aid of the Australian Government (ATA - 76).

To guarantee the safety of those commodities which cover the potency, quality, purity and harmlessness they should be tested in an assay laboratory.
Such laboratory has so far not available in Indonesia. It is therefore the donor country is expected to render technical assistances to establish an assay laboratory in Serpong. The scope of the technical assistance expected is consisting of expertise, equipment, vehicles, buildings construction and fellowship.

2. Name and project activities.

Project title : Establishment of the assay laboratory
The activities of the project will be :

(1) To test the biological products such as vaccines, sera and biological diagnostics both local production and imported from abroad.

(2) To test the veterinary drugs such as veterinary general medicaments and antibiotics both local production and imported from abroad.

The main test is covering the safety, quality, potency and the harmlessness.

3. Institutional framework.

The responsible government agency for the implementation of the project will be the Directorate General of Livestock Services Department of Agriculture. Within the Directorate General of Livestock Services the specific responsibility rest with the Directorate of Animal Health.

4. Government follow-up.

When the project has been completed the Government will carry on the operational of the assay laboratory.

II. Objectives.

a. Immediate : To establish an assay laboratory in Serpong
order to improve the zoosanitary situation in the field
and to contribute in the promotion of livestock husbandry

by .....

by taking necessary measure for testing all vaccines, sera, biological diagnostics veterinary drugs used in Indonesia.

b. Long range : The long range objective of the project are related directly to the Government's Five Year Development Plan which include increasing the productivity of livestock industry, improving the availability of animal protein, and increasing the exports through which the income of the farmers could be improved. To this end, a strategy has been adopted to improve quality of breeds, animal health, credit facilities, and to develop grassland and forage crops.

III. PLAN OF OPERATION.

Activities	duration
1. Preliminary activities :	
design of laboratory study the sites for the laboratory in Serpong. tender for contractor, building contract.	6 months (June - December'83)
2. Training of personnel	
a. Practical training (10)	6 months (June - December'83)
b. Study tour (4)	3 weeks (1984) (January-March'84)
3. Building constructions	9 months (January-September'84)
4. Procurement of equipment	5 months (March - September'84)
5. Instalation of laboratory equipment	5 months (October'84-February'85)
6. Preliminary implementation of test	1 year (February'85-January'86)
7. Full operation activities	1 year (February'86-January'87)
8. Assignment of Short term expert (1)	3 months (June - August'83).

9. .....

9. Assignment of long term expert (5) — 28 months (September'84-December'86)

10. Collaboration with the other institutes dealing with animal health services — Continous starting September '84

11. Reporting
- Semi annual review — every 6 months
- Terminal report — end of 1986.

IV. GOVERNMENT AND EXTERNAL INPUTS.

a. Expertise : - Coordinator - Pathology - Bacteriology - Virology - Parasitology - Biochemistry	180 m/m	= US $ 180,000.-
b. Training of personnel	72 m/m	= US $ 72,000.-
c. Basic equipment & construction		= US $ 5,000,000.-
d. Laboratory equipment, supplies, vehicles		= US $ 400,000.-
e. Land, road construction Electricity, and water installations		= US $ 200,000.-
f. Master plan		= US $ 40,000.-
g. Residential building construction		= US $ 300,000.-
h. Administration cost		= US $ 85,000.-
Total Cost		= US $ 6,277,000.-

Project Digest .....

PROJECT DIGEST

1. Project Title : The Establishment of assay Laboratory

2. Location : Serpong

3. Project sponsor : Department of Agriculture, Directorate General of Livestock Services.

4. Objectives :

a. Immediate : To establish an assay laboratory in Serpong in order to improve the zoosanitary situation in the field and to contribute in the promotion of livestock husbandry by taking necessary measure for testing all vaccines, sera, biological diagnostics and veterinary drugs used in Indonesia.

b. Long range : The long range objectives of the project are related directly to the Government's Five Year Development Plan which included increasing the productivity of livestock industry, improving the availability of animal protein, and increasing the exports through which the income of farmers could be improved. To this end, a strategy has been adopted to improve quality of breeds, animal health, credit facilities, and to develop grassland and forage crops.

5. Donor country : Japan

6. Duration of the project : 3 (three) years (1983/1984, 1984/1985, 1985/1986).

7. Scope of Assistance requested:

a. Expertise : (i) Short term:- One Pathologist;
- One Pharmacist.

(ii) Long terms:
- The Project Coordinator
- One Bacteriologist
- One Virologist
- One Parasitologist
- One Biochemist

180 m/m

b. Fellowship .....

b. Fellowship :

(i) Practical training : 10 counterpart for six months each.

(ii) Study tour : 4 senior officials for three weeks each.

c. Basic equipment to construction

d. Laboratory equipment, supplies, vehicles

8. Project Cost Foreign :

a). Foreign Exchange

(i)	Expertise	: US $	180,000.-
(ii)	Fellowship	: US $	72,000.-
(iii)	Basic equipments & constructions	: US $	5,000,000.-
(iv)	Laboratory equipments, Supples, vehicles	: US $	400,000.-
	Total Cost	: US $	5,652,000.-

b). Counter rupiah : Rp. 440.000.000,-

9. Related to Project Aid : ---

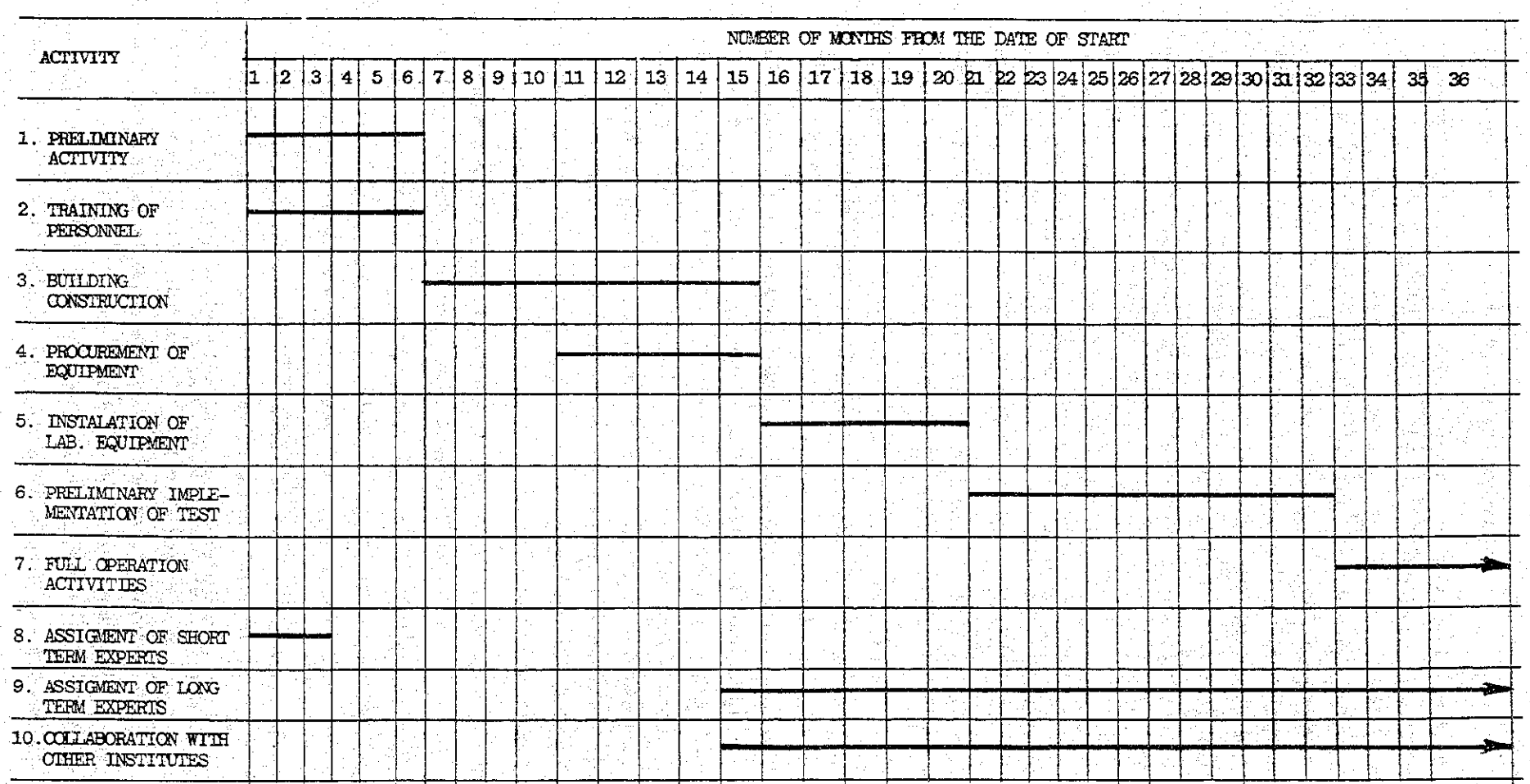
10. Brief explanation :

The function of an assay laboratory is very significant in guaranteeing the safety, potency, quality and hemlessness of vaccine, drugs or other biological products. Such a laboratory is not available in Indonesia.

An approach has been made with The Japanese Government Animal Health Survey Team organized by The Japan International Cooperation Agency headed by Dr, Muneo Ogata.

The survey Team recommended that referring to the establishment of an assay laboratory in Indonesia might be expected by Japanese cooperation.

SCHEDULE PLANNING



PUSAT VETERINARIA FARMA
(VETMA)

VETERINARY BIOLOGICS CENTRE

The present PUSAT VETERINARIA FARMA which is wellknown by its acronym " VETMA " is the center dealing with the production of veterinary biologics originated from the so-called BALAI PENYELIDIKAN PENYAKIT MULUT DAN KUKU (Foot and Mouth Disease Research Station), which was located in Jakarta. The establishment of the Institute was based on the Decree of 12 September 1952 no. 92/Um/52 issued by the Minister of Agriculture, which main program comprised the following :

1. Carrying on experiments aiming at increasing cattle products as the main source of meat, milk and energy.
2. Carrying on researches on Foot and Mouth Disease.
3. Specializing in the product of vaccine for Foot and Mouth Disease.

As Foot and Mouth Disease causes great economic losses, the idea of establishing a Research Institute was looked with favor by the Food and Agricultural Organization of the United Nations, which was then named the Economic Cooperation Administration (ECA), or the Foreign Operation Administration (FOA). Following a Decree of 29 June 1951, no. PA 97 - 47 - 806 - 6543 and PA 97 - 47 - 304 - 6543, an aid of US $ 75.000 was given to the Institute for financing the purchase of laboratory equipments, whereas the land purchase and the constructions were burdened to the Indonesian Government.
Furthermore, the agreement on technical aids stated other determinations including these following :

1. The station was destined to produce vaccine to meet the demand from Indonesia in particular, and from South East Asia in general.
2. The station would assume the function of a Regional Reference Laboratory for Foot and Mouth Disease Research in the region of South East Asia.
3. To enable the station to operate immediately, within the framework of the Expanded Technical Assistance (EPTA), FAO would provide experts for vaccine production according to Frenkel's method. They were hoped to be from Dr. Frenkel Laboratory in Amsterdam.

Later on, due to various unsurmountable difficulties, the site of the Institution was displaced to Surabaya on area of $\pm$ 13 HA in Wonocolo

(including the lot for officials'houses).

To secure favourable action in the existing conditions and situations of the time, several successive names were introduced. The former " balai " (station) became later on " lembaga " (institute), so that the former Balai Penyelidikan Penyakit Mulut dan Kuku (Foot and Mouth Disease Research Station) in its turn was renamed LEMBAGA PENYAKIT MULUT DAN KUKU (Foot and Mouth Disease Institute).

In the meantime, the construction of the Institute's building was not proceeding without difficulties. On the contrary, the difficulties were so enormous that the Institute could not be officially opened sooner than 24 June 1959.

But again, shortly after its operational commencement new problems arose : shortage of raw materials such as cow tongues (for producing vaccine according to Frenkel's method) and other necessary chemicals. The promised experts never arrived either. In the meantime the old method of Frenkel was already replaced by a new one, that made vaccine production in a larger quantity possible. The new method (the Waldman's method) did not help much, since the product was still too low, even insufficient to meet the domestic demand.

Considering the fact that a big fund had already been spent in the establishment of the Institute, yet the vaccine production was still very low, there arose an idea to make the Institute work in a more efficient way, by extending its scope of activities. In agreement with the Decree of 10 December 1966, no. Kep.30/12/66, issued by the Minister of Agriculture, the previous Foot and Mouth Disease Institute was replaced by LEMBAGA VIROLOGI KEHEWANAN (Animal Virology Institute), with extended program comprised :

1. Producing medicines and diagnostics for the prevention and eradication of the animal viral diseases.
2. Giving assistance to the Animal Health Officers on field duty in finding out the animal viral diseases and confirming their diagnosis.
3. Carrying on researches, safety tests and potency tests of the medicines.
4. Carrying on applied and development researches, aiming at progress in the prevention and eradication of the animal viral diseases.

Thenceforth, the Institute was engaged in producing various sorts of vaccine, in addition to its main duty to produce vaccine for Foot and Mouth Disease.

In order to improve and accelerate the Institute's achievements, effort were made to make use of expertise from various countries and International Organizations such as British Council, Colombo Plan (Australia), DOS (Belgium), FAO, Kobe University of Japan, OTCA and UNICEF. Cooperations were also made with several institutes and organizations such as the Agricultural Institute of Bogor, the Medical Faculty of the Airlangga University, the National Health Institute of Surabaya, the Veterinary Faculty of the Airlangga University and the like.

Since 1976, however, the vaccine production for Foot and Mouth Disease was stopped as the existing laboratory was refurbished in such a way to make it available for the vaccine production according to the method of " cell suspension tissue culture ", with a technical assistance from the Australian Government under the Colombo Plan. Using this method, not only quality is much improved, but also the production capacity is increased, namely from 200.000 doses per year to 5.000.000, which would be achieved in stages.
In addition to laboratory improvement mentioned above, assistance was also given for Foot and Mouth Disease Eradication Program in seven provinces, namely Bali, East Java, South Sulawesi, Central Java, Yogyakarta, West Java and Jakarta Raya.

Following a Decree, issued by the Minister of Agriculture, of 2 May 1975, no. 190/Kpts/Org/5/1975 that served as implementation to the Decree of the President of the Republic Indonesia nos. 44 and 45 of 1975, the Animal Virology Institute, was assumed a domain of the Agricultural Research and Development Body. When, however, research activities were seperated from those of production, following a Decree, issued by the Minister of Agriculture of 31 December 1975, no. 503/Kpts/Org/12/1975, the Animal Virologi Institute underwent a functional change, subordinated to the Directorate General of Livestock Services, as the Vaccine and Antisera Unit. This change of status was based on determinations that research function should be seperated from production one, in this regard vaccine and other biologics production.Consequently research function went into the Animal Desease Research Institute in Bogor. The Vaccine and Antisera Unit in Surabaya specialized in production. But the materialization of this change of status did not take place before a Decree of Validation as to the structure of the organization, main programs, function and status of this Unit, was issued by the Minister of Administrative Reform.

Only on 25 May 1978, a Decree was issued by the Minister of Agriculture, no. 31/Kpts/Org/5/1978, designating the status, duties and function of the Vaccine and Antisera Unit, which was henceforth named PUSAT VETERINARIA FARMA (VETERINARY BIOLOGICS CENTRE), in cooperationship with

the Minister of Administrative Reform as molded in his letter of 13 May 1978, no. B.512/Mempan/5/78. In the Decree these following points were designated :

1. The VETERINARY BIOLOGICS CENTRE (VETMA) would be the Technical Implementation Unit for vaccine, antisera, diagnostics and other biologics production within the domain of the Department of Agriculture, which was under the direction of and accounted for to the Director General of Livestock Services.
2. The VETERINARY BIOLOGICS CENTRE (VETMA) has the duty to produce and distribute vaccines, antisera, diagnostics, and other biologics, for the prevention, control and eradication of animal diseases, based on the regulations and laws in force.
3. For the implementation of the duties mentioned above, the VETERINARY BIOLOGICS CENTRE (VETMA) assumed the function of :
   - 3.1. Producing vaccines, antisera, diagnostics and other veterinary biologics.
   - 3.2. Testing the quality of the final products.
   - 3.3. Supplying and maintaining production outfit and distributing the products.
   - 3.4. Carrying on researches for the improvement of quality of the products and for the identification of the diseases.

Since its official opening on 22 September 1979, step by step the VETMA (VETERINARY BIOLOGICS CENTRE) has developed its capacity as a Vaccine Producing Unit, which main job is to meet the demand for biologics in this country.

For the improvement of production capacity and for perfecting its product quality, various efforts have been done. Increase of equipment and employees, education and training for officials abroad as well as at home, researches aiming at production of new sorts of vaccines and the support of a higher quality products have also been done. At present VETMA employs 25 expertsof different scientific disciplines. In cooperation with veterinary surgeons, who constitute the majority of the employees, there are pharmacists, biologists, economists, a medical doctor and an engineer.

VETMA, which in the beginning could only produce 8 sorts of vaccines, at the moment has made considerable progresses, so that it is able to produce 19 kinds of biologics. New sorts of vaccine have undergone production tests, suchs as Avian Encephalitis Vaccine, Marek Vaccine and the like, whereas the quality of several important biologics have been much improved.

In the installation ceremony of VETMA, the Foot and Mouth Disease Laboratory, that was just renovated by the Australian Government (under the Colombo Plan), was officially handed over by His Excellency the Australian Ambassador to Indonesia to the Indonesian Minister of Agriculture.
Despite Indonesia has only one type of Foot and Mouth Disease virus, namely O type, many difficulties had been faced during its first stages of this vaccine production such as adaptation to new equipment, limited skill, shortage of chemicals and other technical side effects, such as contamination etc.
At present the laboratory has been succesfully able to produce the Foot and Mouth Disease vaccine by applying the method of suspension tissue culture.
The first batches of the Foot and Mouth Disease vaccines on May 1981 were distributed in West Java for the rural cattle mass vaccination campaign.

In the meantime, vaccines and other sorts of biologics are undergoing improvement year after year, coping with the increasing need and demand.
The number of VETMA's products can be studied from the following table :

PRODUCT OF VACCINE & DIAGNOSTIC OF VETMA
STARTING FROM FYDP II UP TO 1981/1982
(in million doses)

No.	VACC./DIAG.	1974	1975	1976	1977	1978	1979	1980	1981
1	A n t h r a x	-	-	-	0,05	0,1	0,55	1,2	1,2
2	Bruc. Ant. (RB)	-	-	-	-	0,008	-	0,0333	0,0333
3	Bruc. Ant. (SAT)	-	-	-	-	0,003	-	0,02	0,004
4	Bruc. Str. 19	-	-	-	-	-	0,0012	0,02	0,02
5	Diphteria	0,05	0,44	0,72	1,2	1,1	1,8	0,7	-
6	Fasciola Ant.	-	-	0,001	0,01	0,01	-	0,01	0,015
7	F. M. D.	0,0739	0,014	-	-	-	-	1,2	0,6
8	Fowl Cholera	-	-	-	0,127	0,05	0,1	0,648	0,9
9	Haemoph. Gall.	-	-	-	-	0,01	0,16	0,21	0,1
10	H. S.	-	-	-	0,07	0,11996	1	2,2	3,3
11	Mycopl.Gall.(Ant)	-	-	-	-	0,005	0,02	0,03	0,01
12	N.D. (B1)	-	0,03	0,9	0,15	0,1	0,1	0,2	0,2
13	N.D. (Inactive)	0,138	0,276	0,428	0,75	0,95	0,7	1,092	0,8
14	N.D. (F)	2,73	1,225	4,095	6	9,5	6	12	16
15	N.D. (K)	23,7	28,155	27,3	31,9	39,5	37	48,2	49
16	N.D. (Lasota)	-	0,345	0,632	0,8	1,092	0,307	1,2	0,7
17	Pullorum (Ant.)	-	-	-	-	0,005	0,01	0,02	0,1
18	Rabies (Flury)	-	-	-	-	0,0005	0,0005	0,0013	0,002
19	Rabies (Semple)	0,041	0,0795	0,131	0,107	0,22	0,27	0,33	0,32

スラバヤ生物製剤センターの検定基準マニュアル

(Manual on Veterinary Biologics Test
Centre for Veterinary Biologics, Surabaya)

CENTRE FOR VETERINARY

BIOLIGICS SURABAYA

(V E T M A)

MANUAL ON VETERINARY BIOLOGICS TEST.

TABLE OF CONTENT

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1. TEST OF HS OIL ADJUVANT VACCINE.

The HS vaccine is an emulsion consisting of a mixture of paraffin liq-lanolin and the inactivated culture of HS microorganism. The type of emulsion is water in oil type, where the inactivated HS microorganism is evently divided into small particles in the oil. Each dose of HS vaccine (3 ml) containing of 1.5 ml paraffin liq-lanolin and 1.5 ml of inactivated culture of HS microorganism (equivalent to the weight of 2 mg dried microorganism).

The strain used for making culture is Pasteurella multocida Carter's Strain (type E). The culture is inactivated by adding 0.5% formalin p.a.

Test method

Prior to distribution the HS vaccine is subjected to a series of test covering the following:

1. Sterility test.

   It is conducted by using Thioglycolate, Sabouroud and agar blood/serum media.
   This test is intended to determine the existence of contamination by other microorganism of fungi.

2. Emulsion stability test.

   It is conducted by means of storing vaccine for 14 days at the temperature of 37°C. The stability is considered as good if it is not cracking namely the separation of the oil layer at the upper part and the water layer at the lowerpart. According to Bain, the water layer separation is tolerable not

exceeding .....

exceeding than 5% of the total volume. It is necessary to be noted that in the long storage the vaccine will undergo separation into two parts. The upper layer is clear (paraffin liq) and the lower layer is white (emulsion). This process is called "creaming" and if it is shaked the emulsion becomes stable again. The creaming process does not effect the quality of the vaccine.
Different from the creaming process, the emulsion will be separated into two layers instantly if it is shaked strongly. In this case the vaccine is considered as being deteriorated and no longer can be used.

3. Safety test.

It is imposed on every 15 batches by means of inoculating vaccine into two cows intramuscularly. The dose is doubled i.e. 2 x 3 ml = 6 ml for each cow. The observation is carried out for 2 weeks during which no abnormal reactions as mentioned below is tolerable :

- Swallen and inflammation on the spot of inoculation.
- Cripple or lameness.
- Lower appetite.
- Increasing body temperature, anafilaxis and death, etc.

4. Potency test.

It is imposed on every batch by means of using 3 heads of adult rabbits, each of which is inoculated with 1 ml HS vaccine intramuscularly. After 4 weeks the blood is taken from the rabbits from which sera are collected.

This .....

This serum is inoculated into 8 heads of mice subcutaneously each with 0.5 ml. The following day all mice are challenged with virulent HS microorganism culture with the dose of 0.1 cc $10^{1.5}$ mice LD_{50} subcutaneously. The same dose of challenge is also given to 8 heads of control mice.

The observation is carried out for 1 week during which all experimental mice remain alive or not more than 3 heads died, while all control mice died.

For manufacturing HS vaccine, all raw material to be used are undergone tests in accordance with requirements as indicated in the formal manual book.

Dose : 3 ml, given intramuscularly.

2. TEST OF ANTHRAX VACCINE.

Prior to distribution Anthrax vaccine is subjected to a series of test as follows :

1. Property test.

It is examined visually in which it should be colourless, if shaked the foam will develop because of saponin and there is no any other sediment except turbidity which is deriving from the microorganism spore.

2. Purity test.

From the sample of vaccine which is already put in the bottle a small quantity is taken out and planted into Thioglycollate broth media. Casein pepton agar in plate and Dextrose Sabouraud agar. On observation it should be free from any contamination of other microorganism and fungi.

3. Safety and Toxicity tests.

Two heads of healthy goats or sheep are inoculated subcutaneously with the vaccine each with double doses. Observation is carried out for 10 days during which no abnormal reactions may occur.

3. TEST OF FOWL CHOLERA VACCINE

1. Sterility test.

It is conducted by using Thioglycollate, Sabouraud and agar blood/serum media. It is intended to determine the existence of contamination of any other microorganism or fungi.

2. Safety test.

It is conducted by inoculating the vaccine into 6 birds of chicken of 6 weeks old subcutaneously each bird with double doses (2 x 0.5 ml). Observation is conducted for two weeks during which no abnormal reaction as mentioned below may occur :

- Swallen and inflammation on the spot of inoculation.
- Cripple or lameness.
- Increasing body temperature.
- Anafilaxis reaction.
- Death, etc.

For producing fowl cholera vaccine of good quality, a series of tests is conducted in every stage of the production process.

Dose : 0.5 ml, given subcutaneously at the back side of the head.

4. TEST OF MYCOPLASMA GALLISEPTICUM ANTIGEN.

1. Sterility test.

It is planted into solid media of PPLO agar and serum agar or liquid media of PPLO Broth.

2. Homogenity test.

It should be homogenized, delicate without any auto agglutination.

3. Sensitivity test.

It is tested by using the sample serum of chicken. Using pipette put one drop of blood serum on the object glass or glass plate to which add one drop of antigen, stir gently and mix until homogenized. Read the results for two minutes.

Assessment of the result.

Positive : Showing mass/agglutination which is clearly visible particularly at the margin of the mixture.

Dubious : Not clear agglutination.

Negative : No agglutination occurs.

The sensitivity test is also conducted by using reference antigen and serum positive and negative as control. The antigen to be produced may not give different reaction.

The Mycoplasma gallisepticum antigen is microorganic suspension in 0.25% phenol buffer and stained with 1% crystal violet. Expiration period is one year. The following matters should be taken into consideration.

1). .....

1). The storage should be in 2 - 8°C.

2). The suspension of Mycoplasma gallisepticum should be homogenized, delicate without any coarse mass.

3). The solution of crystal violet should be homogenized without any mass. The homogeneous solution is obtained by diluting crystal violet in 10% absolute alcohol from the total distilled water used.

4). Before used it should be shaked.

5). The serum to be examined should be fresh and uncontaminated.

6). Bottles, rubber caps, pipettes which are furnished should be in sterile condition.

7). Before its expiration period is over, it should give good result upon the sterility, homogenity and sensivity tests.

Effectiveness.

For determining of Mycoplasma gallisepticum infection in chicken.

Application.

Drop through pipette blood serum to be examined on object glass or glass plate, on which drop Mycoplasma antigen. Stir gently until homogenized.

Read the result for two minutes.

Dose : 0.05 ml for every drop of blood serum.

Packing : .....

Packing:

Mycoplasma gallisepticum antigen is packed in bottle of 10 ml covered tightly with rubber or plastic cap, furnished with pipette and leaflet wrapped and put in a box in such a way so that no leak, shock and press-proof. With this condition it is expected to maintain the quality of antigen remain good during transportation to the place of destination.

Each bottle contains 200 drops/doses.

5. TEST OF BRUCELLA ABORTUS STRAIN 19 VACCINE.

The following tests are imposed to Brucella Abortus Strain 19 vaccine before it is circulated :

1. Identification test

To ensure that the vaccine containing bacteria which its morphology and culture are identical to Brucella abortus strain 19 bacteria.

2. Dissosiation test.

Not less than 95% of the colony should be smooth colony. Examination is carried out by using Stereo microscope, Acriflavin staining substance and gentian violet solution.

Stereo microscope.

The shape of smooth colony is seen smaller than other colony.

Round colony in bluish upto greenish are visible.

Acriflavin test.

The growth of bacteria in Potato Agar Plate is taken our in small quantity put on object glass on which Acriflavin Solution is added (10 mg in 10 ml distilled water) and stirred. The solution may not be mass and should remain suspension.

Gentian violet test.

In calculating the number of bacteria in the plate agar culture gentian violet solution is put on it for 10 up to 15 minutes. And then, the solution is taken

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out with pipette.
The colony do not absorb gentian violet.

3. Calculation of Colony.

The calculation of bacteria is carried out by diluting vaccine 10 - 10^9 times dilution. 1 ml of the 10^9 dilution is planted in the agar plate (serum Dextrose agar) and incubated at 37^oC for 4 days. The growth of colony is observed. For example in 5 agar plates the growth of colony is at average 40, so the number of bacteria becomes 40 x 10^9/ml vaccine.

Requirement : 40 - 120 x 10^9 bacteria/dose of vaccine.

4. Safety test.

Not less than 10 guinea pigs are used. They are injected with 1/10 does of cattle intramuscularly. Within 10 days no abnormal reaction has occured. On the 11^{th} day blood is taken out from guinea pigs for serum collection aimed at knowing the titer of the said serum. After that the guinea pigs are killed. The bacteria infesting the spleens of guinea pigs are calculated. The titer of guinea pigs serum may not exceed than 1000 i.u/ml. The content of bacteria in the spleen must not exceed than 500,000 per gr spleen.

Packing

Brucella vaccine contains live Brucella abortus strain 19, therefore it should be stored in solid container. For this purpose vial covered with rubber cap and especially for lyophilisation it is strengthened with aluminium.
The volume of the vial is 20 ml while its content after being dissolved with physiological saline becomes 20 ml

for .....

for 10 doses. The effectiveness of brucella vaccine is to give immunization against brucellosis in female cow aging 3 - 8 months with the duration of immunization for 7 years.

6. TEST OF BRUCELLA SAT ANTIGEN.

Prior to distribution the Brucella SAT antigen is subjected to the following tests.

1. Sterility test.

It is planted in solid media (Potato agar) or liquid media (Thioglucollate).
It is considered sterile if no bacterial growth occurred.

2. Sensitivity/Standardisation test.

Both ISABS (International Standard Anti Brucella Serum = 1000 i.u) and National Standard Anti Brucella Serum Vetma (1.000 i.u) are used. These serum are diluted with 1 ml distilled water, from which dilution with 0.5% phenol saline is made becoming 1 : 150, 1 : 250, 1 : 300, 1 : 350.

The test is carried out in tubes as follows :

SERUM DILUTION

	1:150	1:200	1:250	1:300	1:350	Control
Antigen 1 x 1 ml	0.5	0.5	0.5	0.5	0.5	0.5
S e r u m ml	0.5	0.5	0.5	0.5	0.5	-
Phenol Saline ml	-	-	-	-	-	1.5
Final dilution Serum	1:300	1:400	1:500	1:600	1:700	Alguti-nation 50%

Interpretation .....

Interpretation :

Antigen 1 x normal meeting with requirement is it in the tubes number 3 in accordance with 50% agglutination (++).

Remarks : What is meant by ;

++++ is agglutination and perfect sedimentation (100%). The top liquid is almost clean.

+++ is agglutination and 75% sedimentation. The top liquid is slightly turbid.

++ is 50% agglutination and the top liquid is turbid.

\+ is 25% agglutination and the top liquid is very turbid.

Application.

Tubes of Ø 50 - 70 mm, tubes rack with 12 tubes, pipette of 1 ml are used.

With 0.5% phenol saline for dilution of serum to be examined 1 : 5, 1 : 10 and so forth up to dilution 640 (tube no. 8), to each 0.5 ml dilution add into each tube concerned 0.5 ml antigen until the last dilution is 1 : 10 and so forth up to 1 : 1280.

The tubes no. 9 and 10 are used as control :

Tubes no. 9 filled with 0.5 ml phenol saline and 0.5 ml antigen (for controlling antigen sensitivity).

Tube no. 10 filled with 0.5 ml antigen, 1.5 ml phenol saline (for controlling agglutination).

Shake and put in the water bath with the temperature of 37°C for 24 hours.

Reading of .....

Reading of the result.

The serum titre is read from the last point of agglutination 50%, expressed in the international unit, by comparing its turbidity with control tube of agglutination 50%.

The calculation is using the following formula.

$$\frac{\text{The tested serum titre against antigen}}{500} \times 1000 \text{ i.u}$$

= ..... i.u per ml serum.

Example :

Serum gives agglutination 50% at the final dilution of 1 : 160, the titre is : $\frac{160}{500} \times 1000$ i.u = 320 i.u.

Dose :

5 ml for each sample serum to be tested (for test of 10 tubes a 0.5 ml).

The Brucella SAT antigen is packed in bottle of 100 ml tightly covered with rubber cap and aluminium. The bottle containing 20 doses with the strength of 1 x normal.

7. TEST OF BRUCELLA ROSE BENGAL ANTIGEN.

The Brucella Rose Bengal Antigen consists of the suspension of inactivated Brucella abortus Strain 19 bacteria stained with Rose Bengal in the buffer solution of pH 3.65 in the concentration of 8%. It is used for rapid agglutination test for diagnosing Brucellosis. It is also known as buffered Brucella antigen (BBA). The application of this antigen is not directly injected into animal, so that it will cause no harm to the animal concerned.

The tests against Brucella Rose Bengal Antigen are as follows:

1. Sterility test	:	It is planted in the potatoes agar plate.
2. Sensivity test	:	By using at least 10 serum samples with dilution variation.
3. Solidity/Concentration test	:	Measured with spectronic or Hopkins tube, containing 8% Brucella bacteria.
4. pH test	:	the pH meeting with requirement is 3.65 ± 0.05 and the pH after added with a large amount of cattle serum becomes 3.80 ± 0.05.

The expiration period of the Brucella Rose Bengal Antigen is 1 year. Matters need to be paid attention are :

1). Storing in the temperature of 2^{o} - 8^{o}C.

2). In making suspension, the bacteria should be pure.

3). Instruments used should be sterile.

4). Bottle and rubber cap should be sterile.

5). .....

5). In testing (sterility, homogenity, sensitivity and concentration) before expiration date is over, it should be still meeting with requirement.

Application.

1. Use the following instruments:

   - WHO agglutination plate Ependorf or Oxford with the absorb volume of 0.025 ml and plastic end.
   - Micro mixer with ± 30 rpm

   The antigen to be tested is stored in the room temperature. At the hole of the WHO plate agglutination, drop 0.025 ml serum to be tested using pipette. Then drop 0.025 ml Rose Bengal Antigen, put the WHO plate in the micro mixer and shake for 4 minutes-read the reaction!

2. Using object glass or glass plate, Micro pipette or Ependorf pipette of 0.03 ml are used. The antigen to be used is kept in the room temperature. Drop serum to be tested at the object glass or squared glass plate, then add the Rose Bengal Antigen. Shake with tooth stick or match stick until homogenized. Shake with circle movement for 4 minutes and read the reaction!

Interpretation of the results.

Negative (-) : in agglutination occurs and also no circle border is visible. Antigen remains homogen.

Positive (+) : delicate agglutination occurs and sometimes the circle border is not so distinctive.

Positive 2 .....

Positive 2 (++) : Mass delicate agglutination occurs with clear circle border.

Positive 3 (+++) : clear agglutination occurs with coarse and big mass. Clearness at antigen liquid is also visible.

Serum which give positive reaction 1, 2, 3 result are continuously tested with tube agglutination test or Complement Fixation Test.

Packing

It is packed in the bottle of 10 ml tightly covered with plastic cap.

Each bottle contains 333.33 drops/doses.

8. TEST OF HAEMOPHILUS GALLINARUM BACTERIN.

Quantitative Composition :

The bacterin is made from suspension of Haemophilus gallinarum microorganism which is derived from 6-7 days embryonated chicken egg injected with Haemophilus gallinarum microorganism seed. The microorganism is inactivated with 0.25% formalin solution added with Aluminium Hydroxygel. Each does of bacterin (0.5 ml) containing at least 1 x 10^8 Haemophilus gallinarum microorganism.
Prior to distribution it is subjected to the following tests :

1. Sterility test.

It is planted in the blood agar, Thyoglycollate PPLO agar and Sabouraud agar. It should be free from live microorganism.

2. Safety test.

It is conducted by inoculated bacterin into 10 heads chicken of 8 weeks old, each bird with double doses (1 ml) subcutaneously. Observation is carried out for 2 weeks during which no sign of side effect occurs.

Method of test on raw material used.

A. Microorganism seed.

1. Purity test.

Seed is planted in the blood agar plate. After being incubated at the temperature of 37°C for 24 hours, it is stained with gram staining

substance .....

substance. In addition it is also planted in PPLO agar, thioglucolate and Saboroud agar.

2. Biochemical test.

Microorganism is planted in substratum of several kinds of chemical substance such as glucose etc.

3. Virulency test.

The seed used if inoculated into 6 - 7 days embryonated eggs, should be able to kill the embryo as much as 90 - 100% within 24 hours.

B. The embryonated chicken egg to be used should derive from poultry farm which is free from any disease and from unvaccinated chicken. Like other biological substance, the Haemophilus gallinarum Bacterin is sensitive to temperature effect. To keep bacterin remain good it is suggested to keep at the temperature of $2^{o}C$ - $8^{o}C$ (refrigerated) and not at the freezing temperature. In appropriate storage it could remain in good condition for one year.

Effectiveness.

Chicken inoculated with Haemophilus gallinarum bacterin become immune against the infection of Coryza. Vaccination is carried out only in chicken of 8 weeks old up. To obtain higher immunity vaccination is repeated in after 3 - 4 weeks. The immunity is for 6 months period. Before used it should be shaked until homogenized and inoculation is carried out by using big size capule subcutaneously at the back side of head with the dose of 0.5 ml. The inoculation with Haemophilus gallinarum bacterin causes no side effect.

Packing .....

Packing

The bottle used for packing Haemophilus gallinarum is made from Polyethylen, covered with rubber and aluminium cap.
Each bottle contains 100 ml for 200 doses.

9. TEST OF FASCIOLA HEPATICA DIAGNOSTIC ANTIGEN.

The antigen consists of suspension of fasciola hepatica fraction and SPS and containing Methylmercury thiosalicylate with the degree of 1 : 10.000 or less.

Method of test.

1. Sterility test.

   It should be free from any other microorganism. The test is conducted by planting in various media.

2. Property test.

   The antigen is in the form of satiated liquid, slightly white-yellowish, contains no strange substance, free from bad ordure and the concentration of substance in each bottle should be equal (homogen). (homogen).

3. Safety test.

   Ten heads of healthy guinea pigs weighing of 300 - 500 grams which are divided into 2 groups for test and control. In test group guinea pigs are inoculated intraperitonially with the dose of 1.0 ml of antigen, while the control group are inoculated intradermally at the lower part of abdomen with the dose of 0.2 ml of antigen. Observation is carried out for 9 days and all test animals should show no clinical symptoms at all which can be compared with the control animals.

4. Potency test.

   At least 15 heads cattle or more which are ready to

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be slaughtered in abattoir are used. Cut clearly hair around the root of the tail about 5 cm diameter. Inoculate intradermally 0.2 ml fasciola hepatica antigen in the middle part of the hair cutted spot. Keep the inoculation area from being touched by hand, alcohol rubbing or other antiseptics until the time of measuring. After 15 - 30 minutes, examine the site of injection and if there is a skin thickness measured or not. Afterwards a comparison is made between the skin reaction due to inoculation with antigen and any changes at the liver after autopsy. The skin reaction is considered good if 80% or more of tested animals show reaction in confirmity with the fact of infection which is visible after slaughtering. The skin reaction is considered as being positive if the spot of inoculation develop skin thickness measuring to 15 mm or more during the 15 - 30 minutes observation. While the negative reaction if the spot of inoculation develop no thickness or the thickness is less than 15 mm.

Packing : Each vial of 5 ml for 25 doses.

10. QUANTITATIVE COMPOSITION OF ANTHRAX VACCINE.

Anthrax vaccine is suspension form which in each mili-litre containing not less than 10 millions avirulent spore of bacillus anthracis strain 34 F-2 Weybridge, in the solution of physiological saline and glycerin and containing also 0.05% saponin.

Sterile casein digest media is used for the growth of bacillus anthracis.

The anthrax vaccine is live vaccine which could give active immunization against anthrax disease.

The usage of glycerine is meant to slow the vaccine entrance or the vaccine is absorbed by blood slowly so that it is effective in developing antibody.

Meanwhile the usage of saponin is meant to keep the smoothness of blood circulation so that no necrosis in the spot of inoculation.

11. QUANTITATIVE COMPOSITION OF FOWL CHOLERA VACCINE.

The vaccine is suspension form consisting of culture of Pasteurella multocida microorganism the causal agent of fowl cholera which is inactivated with 1% alum adjuvant. The microorganism used for making vaccine is local strain of Surabaya. Each dose (0.5 ml) containing of not less than 10 x 10 inactivated microorganism.

As chemical used for inactivating the microorganism culture is formalin p.a as much as 0.5% to prevent the occurence of foam when it is shaked added with 0.004% silicon solution.

For making 50 litres inactivated culture of fowl cholera microorganism (1 batch = 100.000 doses) the following substance are needed.

1). Casein hydrolysate by acid	1000 gr.
2). Yeast extract	200 gr.
3). Casein hydrolysate by trypsin	240 gr.
4). Pancreas extract	4 litres
5). Equivalent to 9 kg fresh pancreas	-
6). Nutrient Broth	240 gr.
7). Di sodium hydrogen phosphate 12 hydr.	344 gr.
8). Potassium hydrogen phosphate 7 hydr.	88 gr.
9). Sucrose	96 gr.
10). Formalin	250 ml.

For making 1% alum adjuvant it is needed:

- Pot aluminium sulphate.	500 gr.

For purifying microorganism and inactivation test it is needed blood agar base. 50 gr.

12. QUANTITATIVE COMPOSITION OF BRUCELLA ABORTUS STRAIN 19 VACCINE.

It is in freezed dried form. Each dose containing $40 - 120 \times 10^9$ Brucella abortus strain 19 microorganism enzymatic digest of casein, sucrose and sodium glutamate are used.

For the growth of microorganism it is used special media that is sterile potato agar media. No substance which may kill or weaken microorganism is added because this vaccine is live one.

As stabilizer it is used mixture of enzymatic digest of casein, sucrose and sodium glutamate. These substance are not toxic to animals and the inoculated vaccine is easily absorbed by body.

Before use the vaccine is diluted in physiological saline.

Application is subcutaneous in cattle of 3 - 8 months old.

13. QUANTITATIVE COMPOSITION OF MYCOPLASMA GALLISEPTICUM ANTIGEN.

It is suspension form of inactivated Mycoplasma gallisepticum strain S-6.

The suspension is made with 0.25% phenol buffer and stained with 1% solution of crystal violet.

The antigen is used for serum agglutination test (serum plate agglutination) for determining the Mycoplasma gallisepticum infection in chicken.

14. QUANTITATIVE COMPOSITION OF BRUCELLA SAT ANTIGEN.

The Brucella SAT 1 x normal antigen is used for tube agglutination test in diagnosing brucellosis.

It is suspension of inactivated Brucella abortus strain 99 microorganism which manufactured in conformity with the recommendation of FAO/WHO Expert Committee on Brucellosis 1964, containing 0.5% phenol so that it will not give negative effect in either animals or human being who conducts the test.