The External Quality Assurance Programme for use with the FAO/IAEA Rinderpest Competitive ELISA

Interim Report (EQAP/RP/1998A)

Prepared by: Axel Colling EQAP Co-ordinator And Helder Louvandini Animal Production Unit FAO/IAEA Agriculture and Biotechnology Laboratory Agency's Laboratory A-2444 Seibersdorf, Austria Fax: + 43-1-206028222 E-mail address: <u>a.colling@iaca.org</u>

Limited Distribution



JOINT FAO/IAEA DIVISION OF NUCLEAR TECHNIQUES IN FOOD AND AGRICULTURE Animal Production and Health Section & AGENCY'S LABORATORIES FAO/IAEA AGRICULTURE AND BIOTECHNOLOGY LABORATORY Animal Production Unit



The External Quality Assurance Programme for ase with the FAO/LAEA Rinderpest Competitive ELISA

Interim Report & QAERCHINGS

Proposition And the second of the second of

tinital learning



Ammil Productine and Health Section 3 FAULAEA AGRICULTURE AND BRITECHNOLOGY 1 ANDEA AGRICULTURE AND BRITECHNOLOGY



CONTENTS

٠

Page

1.	INTRODUCTION	1
2.	MATERIALS AND METHODS	4
	2.1. Questionnaire	5
	2.2. Internal Quality Control (IQC) Data	5
	2.3. External Quality Control Test Panel	5
	2.4. Distribution	6
3.	RESULTS	6
	3.1. Questionnaire	7
	3.2. Internal Quality Control Data	8
	3.3. External Quality Control Test Panel	9
4.	DISCUSSION AND CONCLUSIONS	16
	4.1. Questionnaire	16
	4.2. IQC Data	10
	4.3. The EQC Test Panel	18
	4.4. EQAP/RP/1997a, 1998a	18
5.	RECOMMENDATIONS	21
6.	ACKNOWLEDGMENTS	23

- ATTACHMENT I Summary Questionnaire
- ATTACHMENT II Evaluation IQC data

ATTACHMENT III Accumulated Data for Determination of Status "Recognition"

SINTINO 3

.

A FEO FRITEET THE ACCOUNTED DATE for DEPEndence of File Line Recognition

THE FAO/IAEA EXTERNAL QUALITY ASSURANCE PROGRAMME FOR DISEASE DIAGNOSIS

THE EQAP FOR THE FAO/IAEA RINDERPEST COMPETITIVE ELISA; EQAP/RP/1998A.

SUMMARY

The External Quality Assurance Programme (EQAP) consists of three equally important items: the Questionnaire, the monitoring of the Internal Quality Control (IQC) data and the External Quality Control (EQC) test panel. The EQAP is conducted twice per year.

Twenty-eight laboratories participated in the fourth round of the FAO/IAEA rinderpest competitive ELISA, EQAP/RP/1998a. Of these 19 confirmed receipt of the EQA panel. The questionnaire, IQC and EQC results were returned by 16, 15 and 19 laboratories respectively and results are presented in this report.

Overall, results show that the majority of participating laboratories has an acceptable proficiency in conducting the rinderpest FAO/IAEA ELISA test. However, several laboratories still need to improve their IQC practices; i.e., they must concentrate on the monitoring and analyses of the IQC data and should regularly check the calibration of their ELISA equipment. With regard to the EQC test panel, 19 laboratories returned EQC results with an overall agreement of 100% for samples 1, 2 and 5 and of 95% for sample 3 and 4, giving an overall agreement of 98%. No sample had to be excluded. These EQC results are the best ever produced with the competitive Rinderpest ELISA.

The results of this round show that the EQAP is a valuable tool in the assessment of both the results obtained from and the proper functioning of the FAO/IAEA rinderpest ELISA. Furthermore, the EQAP can assist counterpart laboratories to establish and implement Quality Control/Quality Assurance (QC/QA) procedures for conducting the FAO/IAEA ELISA, and to advise on the implementation of similar QC/QA procedures in other laboratory activities.

Based on the results of the proficiency testing of the last 2 consecutive EQAP Rinderpest rounds 12 laboratories qualified as "provisionally recognized" and 2 laboratories qualified as "recognized".

I. INTRODUCTION

For any testing laboratory it is essential that assurance can be given that the test results produced are valid and reliable. It is also very important that results are comparable between different laboratories involved in similar assessments. Many diagnostic tests contain an element of subjectivity in their interpretation of results, and this renders both internal and external assurance difficult to operate. One of the distinct advantages of an ELISA-based system is the objectivity of reading the results and the ability to process data using a computer. Thus, it is possible to incorporate a high level of internal quality control for every ELISA test plate used. Indeed, Internal Quality Control is now a routine operation for most laboratories utilizing FAO/IAEA ELISA based testing systems [1].

Equally important is the determination whether a laboratory is giving the correct interpretation of the results even when the assay is shown to be functioning correctly. The procedures for establishing the assurance that the test results provided from a laboratory are reliable form the basis for an External Quality Assurance Programme (EQAP).

In 1990 an EQAP was carried out for the FAO/IAEA rinderpest indirect ELISA kit. These results have been published in detail [2]. In 1991, laboratories involved in the Pan African Rinderpest Campaign (PARC) switched over to a competitive ELISA rinderpest kit. In determining external quality assurance for the competitive ELISA, a test panel of 40 'unknown' sera was distributed among the

PARC laboratories. In 1992, 1993 and 1994, such test panels were sent out to a total of 20, 21 and 17 participating laboratories, respectively [3-5].

In September 1994, an FAO/IAEA consultants meeting was convened with the aim of extending and further improving the EQAP for veterinary laboratories in developing countries utilizing FAO/IAEA ELISA kits. The meeting focused on establishing procedures that would lead to "Recognition" of veterinary laboratories as competent in utilizing FAO/IAEA ELISA kits for specific diseases and tasks. The conclusions and recommendations of this meeting are contained in the report "Establishment of external quality assurance procedures for use with FAO/IAEA ELISA kits" [6].

This improved EQAP for veterinary laboratories is based on i) proof of the presence and use of Quality Assurance/Quality Control systems, ii) the continual satisfactory performance of processes and output, and iii) participation in external quality control test rounds. To obtain such proof, the EQAP consists of three critical elements as detailed below:

- Survey Questionnaire:

A questionnaire-based survey of individual laboratories is utilized to provide a regular system for monitoring the presence and use of the key quality elements. It is a mandatory requirement that all laboratories participating in the FAO/IAEA EQAP should complete and return such a questionnaire. The information gathered through the Questionnaire is updated at least once per year by the officer in charge¹ in the participating laboratory. The satisfactory presence of the relevant key elements is determined by the EQAP Coordinator in close collaboration with the appropriate Technical Officer of the Subprogramme in Animal Health and Production of the Joint FAO/IAEA Programme, and forms an essential part of the assessment of the participating laboratory.

- Internal Quality Control (IQC):

It is mandatory that laboratories fulfill the requirements for IQC as specified in the designated standard assay protocol. These include the use of appropriate reference standard control sera, the application of test acceptance criteria, the monitoring of test performance through the use of control charts, and the provision of relevant data for third party assessment. The IQC data are used to assess the repeatability and precision of the test conducted in that particular laboratory [7]. These data can be used by the test operator to detect trends and shifts in test performance, also [8].

- External Quality Control (EQC):

External Quality Control involves proficiency testing; i.e., inter-laboratory comparisons between two or more laboratories. For inter-laboratory proficiency testing, each laboratory conducts the designated test method on a defined panel of test samples, the EQC panel. Identical panels of test samples are dispatched to the participating laboratories for concurrent testing. The proficiency testing is conducted twice per year.

In February 1998 a "follow-up" consultants meeting entitled: "The FAO/IAEA External Quality Assurance Programme (EQAP) and Movement Towards a Generic Veterinary Diagnostic Testing. Laboratory Accreditation Scheme" was convened to consider the design, impact and proposals for future implementation of the current FAO/IAEA EQAP for Animal Disease Diagnosis and make recommendations with regard to its central purposes and future direction. In addition, the Consultants considered the broader question of a generic QA "accreditation" scheme for veterinary diagnostic testing laboratories that could be made available through international, regional, or national organizations as

¹ The officer responsible for the diagnosis and monitoring of rinderpest in an EQAP participating laboratory.

appropriate to the country of interest. This broader discussion was stimulated by the fact that few developed and no developing countries have nationally organized schemes to measure and recognize the QA systems and technical competence of veterinary diagnostic testing laboratories, but such a scheme is of vital importance to the quality of policy and decisions and actions taken on national animal health issues and the international trade of livestock and livestock commodities. It followed that, in the Subprogramme's role as a Collaborating Center to the Office International Epizooties (OIE, or World Animal Health Organization), it would be appropriate to consider the FAO/IAEA EQAP within the broader scope of an international scheme for veterinary diagnostic laboratory accreditation for two reasons: 1) to use information learned through the design and implementation of the FAO/IAEA EQAP to assist in the appropriate development of an international scheme and 2) to ensure that the FAO/IAEA EQAP bijectives and procedures are in harmony with international QA guidelines as they develop in this area [9].

The objectives of the EQAP effort were and remain to a) develop reference data for the assessment of new FAO/IAEA diagnostic assay performance in the field, b) determine the user's general QA status and specify assay proficiency, c) enhance the user's QA awareness and culture, d) provide an organized and transparent mechanism to enhance the national and international credibility of the user's laboratory. In addition, the data developed through the FAO/IAEA EQAP can be used from a programmatic perspective as baseline data for a) the development of appropriate intervention strategies, b) monitoring project implementation, and c) evaluation of project impact during and after the project's conclusion.

It is recognized that the FAO/IAEA EQAP is programmatic in nature and is designed to assist counterpart laboratories to bridge the gap between what they have now and formal national or international recognition of Quality Management and technical competence.

The first round of the new EQAP for the FAO/IAEA rinderpest competitive ELISA (RP95a) started in October 1995. In total, 23 laboratories participated, all from the PARC programme. From the information collected from the Questionnaire, it was concluded that the routine monitoring of the IQC data by the test operator and the calibration of equipment needed more attention in most laboratories. The IQC analysis of the different laboratories showed that most laboratories produced reliable results. However, several laboratories needed to reduce the variation in IQC data, thus avoiding that the IQC data extend beyond the Upper and Lower Control Limits (UCL and LCL, respectively). There was an overall agreement on the EQC test panel results of 97%. Only 2 laboratories wrongly identified a positive test sample as negative. A comprehensive report on this round was distributed [10].

The second round of the EQAP for the FAO/IAEA rinderpest competitive ELISA (RP96a) started in August 1996 [11]. To assure confidentiality, a code number identified the participating laboratories.

The third round of the EQAP for the FAO/IAEA rinderpest competitive ELISA (RP97a) was started in July 1997 (RP97a). An overall agreement on the EQC test panel results of 93% was observed. Each laboratory received a new code number for this round [12].

Twenty-eight laboratories participated in the fourth round of the FAO/IAEA rinderpest competitive ELISA, EQAP/RP/1998a. Of these 19 confirmed receipt of the EQA panel. The questionnaire, IQC and EQC results were returned by 16, 15 and 19 laboratories respectively and results are presented in this report. Code numbers are the same as in the RP97a.

adoption which that it is a continued the goodburnance during former 10,041 (reading to the second state of the the state of the the second state of the second state

MATERIALS AND METHODS

Many parties are involved in the different steps, of which a round of the External Quality Assurance Programme consists of and great effort from each participant is needed to assure final success. An overview of the different steps and involvement for the Rinderpest EQAP is shown below in Fig. 1.



Fig. 1. Overview of the Rinderpest ELISA EQAP round.

A total of 28 laboratories participated in the fourth EQAP round for the FAO/IAEA rinderpest competitive ELISA. Of these 28 laboratories, 21 are located in Africa and 7 in the Middle East. Participants from Africa are part of the PARC programme and participants from the Middle East are part of the of the Middle East Rinderpest Eradication Programme (MEREP).

2.1. Questionnaire

Laboratories, which had already completed the Questionnaire during former EQAP rounds, received a copy of their completed Questionnaire and were asked to review and, if applicable, update the

information. Laboratories, which had not completed or returned the Questionnaire during former EQAP round were sent a new Questionnaire and were asked to complete and return this.

The Questionnaire consisted of the following 9 categories:

- A : Administrative information
- B : General information on other diagnostic activities performed in the laboratory
- C : Laboratory facilities
- D : Maintenance and calibration of equipment
- E : Handling of test results
- F: Monitoring of IQC data
- G : Laboratory staff
- H: Other quality assurance procedures within the laboratory
- I : Availability, specifications and usage of computers

2.2. Internal Quality Control (IQC) Data

The IQC data provide valuable information on the test performance in an individual laboratory. The IQC data for the FAO/IAEA rinderpest competitive ELISA consists of the four replicates of the monoclonal antibody control (Cm), of the high positive control (C++), of the medium positive control (C+), and of two replicates of the negative control (C-) and the conjugate control (Cc). For IQC evaluation, the mean of the 4 values of the 4 wells is taken for the Cm, C++ and C+. The Cc and C- are tested in duplicate only, and the mean of the 2 values is taken.

Prior to incorporation into the competitive ELISA for rinderpest, the IQC samples were tested extensively under different circumstances by the WRL using the same ELISA. Given that the variation in optical density (OD) values and percentage inhibition (PI) values is normally distributed, ± 3 standard deviations (SD) were calculated and used to set the UCL (+ 3 SD) and the LCL (- 3 SD) of each IQC serum sample. These control limits are provided with the FACT SHEET of each new ELISA kit.

As part of the EQAP, the participating laboratories receive a diskette containing a 'batch' file ,which copies the 'instatqc' or 'eqstat.qc' file from the computer linked with the ELISA reader to the diskette. The 'instatqc' or 'eqstat.qc' file saves the IQC data of each ELISA plate read. This is applicable only for laboratories using the FAO/IAEA computer program RPEIA or EDI (ELISA Data Interchange).

If a laboratory was not using the EDI program to read and calculate the ELISA plates, the laboratory was requested to send printouts of the IQC data of the last 30-40 test plates in a table format.

For the IQC evaluation, the mean $(\pm 2 \text{ SD})$ of the 4 values of the 4 wells per ELISA plate is taken for respectively the Cm, C++, C+, and the mean of the two values for the Cc and C- respectively.

2.3. External Quality Control Test Panel

The External Quality Control (EQC) test panels consisted of 5 freezes dried serum samples; 3 positive samples and 2 negative samples. The EQC test panels for this round were prepared and dispatched by Dr. John Anderson, Animal Health Institute (AHI), F. bright, U.K. The serum samples were derived from experimentally immunized animals using rinderpest vaccine. All sera were undiluted and obtained from a single animal except for the sera used for Sample 2, which was a pooled positive sample of two immunized animals. The test samples, 1.0 ml serum per aliquot, were freeze-dried in one batch at AHI. The samples were tested prior to and after freeze-drying. Each test sample was subsequently labeled with a unique code number; hence each laboratory received uniquely coded unknown test panels.

The laboratories were requested to reconstitute the EQC samples on the day of testing using attached distilled water, and to treat and test these samples in a manner identical to that of their field

sera. The laboratories were requested to provide the EQC results in terms of positive or negative results for each sera and to submit the full computer print out of the test plate, thus including IQC data of the ELISA plate and the PI values for each EQC test sample.

2.4. Distribution

In July 1998 the EQC test panels were dispatched from AHI, U.K., principally by international courier to the UNDP offices in the different countries. All participating laboratories and their associated UNDP offices were notified by fax/telex/e:mail of the date of dispatch. The laboratories were urged to collect the EQC test panel from their respective UNDP Offices as soon as possible. It was not possible to ascertain the travel time of each test panel nor the condition it arrived in. However, since the sera were freeze-dried, it was not expected that the time or temperature experienced during shipment would add any unwanted variables.

To avoid loss or loss of track of EQA panels and enhance timely submission of results the following communication scheme was set up for future shipments²:



It is assumed that after a maximum of two weeks post-shipment the confirmation fax must be received by the TO. After receipt of the confirmation fax the laboratory is given three weeks produce and return results resulting in a maximum of 5 weeks from shipment of the EQA panel to the receipt of results.

Results from some laboratories were received very late by the EQA coordinator. These laboratories are requested to pay attention that their results are received on time during the next rounds to assure that they will be included in the report. In general, laboratories are requested to adhere more strictly to the deadline.

3. RESULTS

A total of 28 laboratories participated in this round. Nineteen laboratories (68%) confirmed receipt of the panel and of these 16 (84%), 15 (79%) and 19 (100%) laboratories sent questionnaire, IQC data and EQC results respectively. Six laboratories informed the EQAP Coordinator that they were not able to fulfill all requirements of the EQAP for various reasons. The main reasons being that a) the laboratory was still awaiting receipt of a new ELISA kit and/or b) had broken/missing equipment and/or c) the EQAP items got lost in the mail and/or d) there were customs clearance problems. An overview of the results received by the EQAP Coordinator is given in Table I.

² CP = Counterpart; TO = Technical Officer; EQAP Co = EQAP Co-ordinator

Laboratory 6 supplied EQC results from the last round (RP97a)

						(22011
Lab.Code	Quest.	IQC	EQC	Lab.Code	Quest.	IQC	EQC
1	x	х	x	17			_
2	х	х	х	18			
3				19	x		x**
4				20*	ດາວ ເວັ້ນອັນວ		XTT
5	х	x***	x	21			
6				22	х	x	
7*				23	x	x	x
8			x	24	x		х
9	x	X***	x	25*	~	х	х
10	x	х	x	26	х		
11	x	х	x	27*	~		x
12	x	х	x	28			
13			х	29	N.		
14	х	х	x	30	x	x	х
15		x	x	31	x	x	х
16			x	32	х	X	х

TABLE I. OVERVIEW EQAP RESULTS OF THE FOURTH EQAP ROUND (EQAP/RP/1998A)

IQC: Internal Quality Control data; EQC: External Quality Control data; Quest: Questionnaire

* Laboratory did not participate in this EQAP round, ** EQC data were from last round (RP97a), ***diskette did not contain sufficient or any IQC data

3.1. Questionnaire

Sixteen laboratories returned the completed and/or updated Questionnaire during this EQAP round (TABLE I.). The collected information categorized by subject per laboratory is attached in Attachment I. The information presented in *bold italic* format is new or updated since the last EQAP round. Accumulated and updated information of the questionnaire of the last four EQAP rounds of an overall number of 26 laboratories (laboratories 1, 2, 3, 4, 5, 9, 10, 11, 12, 13, 14, 15, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30 and 31) is compiled in Attachment 1. No questionnaire information at all has been received from laboratories 6, 7, 8, 16 and 17.

Power supply/Air condition

Of 26 laboratories 16 (62%) reported problems with the power supply. Six laboratories have both types of power problems, namely "power cuts" and voltage "irregularities", 4 laboratories reported only power cuts and 5 laboratories indicated only voltage irregularities. Seven laboratories do not have any power problem. Regarding the length of period of power problems 7 laboratories have power cuts of less than 12 hours and 13 reported irregular periods. Asked about the frequency of power problems 2 laboratories reported weekly and 15 irregular periods of power problems. Fourteen laboratories use a stabilizer, 3 use a stabilizer but only for selected equipment (e.g. refrigerator). Seventeen laboratories have access to an emergency power supply for their refrigerator and freezer. Sixteen laboratories have an air-condition (8 do not have air-condition). The average temperature was 25.52 °C with a variation of Min. 15°C to Max. 42°C.

Pipettes, Tips and ELISA readers

Most laboratories use pipettes from Biohit Proline® (15 labs) or Titertek (10 labs) followed by Finnpipette (8 labs), Gilsson Pipettman (6 labs) and Socorex (1 lab) and tips from Biohit Proline® (12 labs) and Micronics (9 labs). Less frequently tips from Finntips, Conetreff, Volac 200 and Costar are in use. Twenty-three and 21 laboratories use 5-50ul single and multichannel pipettes respectively. Eighteen

and 20 laboratories use 50-250ul single and multichannel pipettes respectively. Eight and 5 laboratories use 250-1000ul single and multichannel pipettes respectively.

The Multiskan Plus Mark II is the most commonly used ELISA reader (15 labs), followed by the BDSL Immunoscan Plus (9 labs) and Multiskan MCC/340 (4 labs).

Handling of test results

With regard to plate reading and calculation of ELISA results, 23 laboratories use the EDI programme: Ten laboratories are using EDI version 2.11., 9 laboratories are using RPEIA version 1.03, 4 laboratories use ED 2.2., 3 laboratories are using RPEIA version 1.01., 2 laboratories use Procomm and 2 laboratories calculate results manually. Some laboratories indicate to use RPEIA and EDI together.

Twenty laboratories have a computerized system SID (17 labs), Panacea (6 labs), EPI-info (2 labs), Access (2) or a spreadsheet programme³, to link the test results with other details of the field samples.

The majority of the laboratories (13) use the IQC data to determine whether the ELISA plate readings are 'within' limits and can be accepted. Two laboratories indicated that the IQC data are monitored using the 'instatqc' programme. Eight laboratories reported that they do not undertake any IOC monitoring.

Sample storage

All laboratories (26 labs) store serum samples at -20°C, in most cases using Cryopreservation vials (13 labs), Vacutainers (7 labs), Nalgene storage system (7 labs), Micronics (7 labs), Serum storage plates (3 labs) or others e.g. 10 ml tubes (1 lab). Twelve laboratories have access to -80°C freezers and 7 to Liquid Nitrogen facilities. Nineteen laboratories reported keeping a serum bank ranging from 500 to 45,000 samples with an average of 11.328 samples.

Computer/Data Processing

Nineteen laboratories reported that a computer is used for reading of ELISA plates and/or storage of data. For the first time more Pentiums (9 laboratories) than 486 processor-equipped-computers (6 laboratories) are in use. Three laboratories use 386 and 1 laboratory uses a 286 CPU computer. Hopefully this trend will continue.

Water quality and equipment calibration

Twenty-two laboratories use distilled water. Nineteen laboratories have access to deionized and 11 laboratories to bi-distilled water. Nineteen laboratories reported that filters and cartridges are changed in the following pattern: once per year (2 laboratories), twice per year (6 laboratories), every three months (5 laboratories), every month (1 laboratory), three times per month (1 laboratory). Six laboratories reported that cartridges and filters are changed following the manufacturers recommendations (1 lab), conductivity control (1) or "when needed" (4 labs).

Twenty-two laboratories reported that no equipment calibration (ELISA reader and pipettes) procedures are carried out. Two laboratories undertake calibration procedures following the manual. One laboratory checks the accuracy of its ELISA reader by comparing OD readings with another ELISA reader.

3.2. Internal Quality Control Data

3

Fifteen laboratories: 1, 2, 5°, 9°, 10, 11, 12, 14, 15, 22, 23, 24, 29, 30 and 31 returned IQC data but information only from 13 laboratories could be evaluated. Evaluation from some laboratories

SID (Sero-monitoring Information Database), Panacea and EPI-info are epidemiological computer programs

^{*} IQC data could no be evaluated due to lack of information e.g. diskette empty, only one printout

e.g. laboratory 14, 30 and 31 show that the assay is well within limits. Intra- and interassay variation is well under control and also the statistical parameter show a good degree of consistency. IQC results from the majority of laboratories e.g. laboratories 2, 12, 15, 22, 23, 24 and 29 show that there are still some outlayers and further adjustment and consistency is required to maintain the assay under control. Finally there is a group of laboratories e.g. laboratory 10 and 11, which apparently needs urgently substantial adjustment in the performance of the ELISA because almost all data fall outside the upper or lower control limits. In these cases it is obvious that the assay is not under control and must be adjusted as soon as possible. There is a general trend indicating low OD values for the Cm. These values are often very close or below the lower control limit (OD < 0.4). Possible reasons for this may the use of old and/or not properly stored reagents or if encountered in a freshly supplied assay wrong dilution/concentration of reagents. The producer has been informed about these findings.

3.3. External Quality Control Test Panel

EQC results of 19 laboratories have been analyzed and are presented in this report. Table II shows the qualitative results per laboratory; i.e., the determination whether a serum sample is considered to be negative or positive in the assay.

Lab. Code	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
1	Pos.	Pos.	Pos.	Neg.	Neg.
2 5	Pos.	Pos.	Pos.	Neg.	Neg.
	Pos.	Pos.	Pos.	Neg.	Neg.
8	Pos.	Pos.	Pos.	Neg.	Neg.
9	Pos.	Pos.	Pos.	Neg.	Neg.
10	Pos.	Pos.	Pos.	Neg.	Neg.
11	Pos.	Pos.	Pos.	Neg.	Neg.
12	Pos.	Pos.	Pos.	Neg.	Neg.
13	Pos.	Pos.	Pos.	Neg.	Neg.
14	Pos.	Pos.	Pos.	Neg.	Neg.
15	Pos.	Pos.	Pos.	Neg.	Neg.
16	Pos.	Pos.	Pos.	Neg.	Neg.
22	Pos.	Pos.	Pos.	Neg.	Neg.
23	Pos.	Pos.	Pos.	Neg.	Neg.
24	Pos.	Pos.	Pos.	Pos.	Neg.
26	Pos.	Pos.	Pos.	Neg.	Neg.
29	Pos.	Pos.	Neg.	Neg.	Neg.
30	Pos.	Pos.	Pos.	Neg.	Neg.
31	Pos.	Pos.	Pos.	Neg.	Neg.
Total No. of Labs	Agreement	Agreement	Agreement	Agreement	Agreement
20	100%	100%	95%	95%	100%

TABLE II. QUALITATIVE RESULTS OF THE EQC TEST PANEL PER LABORATORY (50% cut-off)

Lab. Code	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
DO PARTE COCCO DAVI	87	75	69	29	5
2	82	66	65	33	34
5	83	70	59	27	20
8	82	69	54	33	13
9	89	69	56	21	20
10	85	81	74	48	30
11 benlagout /l	89	72	69	36	14
12	87	63	56	35	13
13	75	61	53	25	31
14	81	76	68	42	25
15	93	81	72	41	26
16	90	79	66	38	12
22	75	71	56	45	10
23	88	73	65	35	38
24	96	88	84	61	29
26	90	78	68	45	27
29	74	61	47	15	5
30	88	71	55	32	35
31	88	80	71	40	37

TABLE III. QUANTITATIVE RESULTS OF THE EQC TEST PANEL PER LABORATORY (PI)

Table III shows the quantitative data; i.e., the percentage inhibition (PI) values for the EQC test samples as determined and submitted by the laboratories.

Table IV shows the summary statistics of all laboratories. The EQC test panel results as submitted by the participants show for samples 1, 2 and 5 100 % agreement for each sample. Ninety-five percent of agreement was achieved for samples 3 and 4. This is the best result achieved with EQC samples up to date.

100 TO	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Mean	85	73	64	36	22
Standard Error	1	2	2	2	2
Median	87	72	65	35	25
Standard Deviation	6	7	9	10	11
Sample Variance	37	53	84	109	116
Range	22	27	37	46	33
Minimum	74	61	47	15	5
Maximum	96	88	84	61	38
Count	19	19	19	19	19
Coef. Variation (%)	7	10	14	29	48

TABLE IV.	SUMMARY	STATISTICS OF	F THE EQC TEST SAMPLES.	

Figures 3a-e show the frequency distributions for the EQC test results. The results of the individual laboratories are presented by their respective laboratory code number in each column. These histograms provide a visual reference for each laboratory's position within the distribution of all results. The horizontal line shows the cut-off value (50%).

-20













Fig. 3d. Frequency distributions of PI values for Samples 4.



Fig. 3e. Frequency distribution of PI values for Sample 5.

Figures 4a-c show the EQC results for Samples 1, 2 and 3 as they are plotted in a simplified Youden diagram. Such a diagram consists of a rectangular plot, on which the individual laboratory's results for two samples are represented by one dot. The X-axis (horizontal component) of each dot is the laboratory's result for sample X, while the Y-axis (vertical component) contains the result for sample Y. The small rectangle inside the Youden diagram (Figure 4a-c) represents the mean ± 1 SD range for both samples.

The Youden diagram helps to identify systematic versus random differences between laboratories. Laboratories with systematic error components are either in the upper right hand quadrant (as formed by the line for the means of both samples) or in the lower left-hand quadrant. A laboratory with results positioned in the upper right hand quadrant and outside the +1 SD range, could indicate that the laboratories values for both positive samples are too high, possibly due to an increased level of diagnostic sensitivity of the assay in that laboratory. A laboratory positioned in the lower left quadrant of



Fig. 4a. Simplified Youden Plot analysis for Samples 1 and 2.



13



Fig. 4c. Simplified Youden Plot analysis for Samples 2 and 3.

the diagram and outside -1 SD range, could indicate that the laboratory obtained results too low for the both positive samples as a result of a decrease in diagnostic sensitivity of the assay. Laboratories reporting results indicating random error are located either in the upper left hand or lower right hand quadrant and outside the ± 1 SD range.

Figures 4a-c show that the results for the majority (59%) of the laboratories (lab. 1, 5, 9, 10, 11, 16, 23, 26, 31) fall within the box (including borderline values) representing the mean \pm 1 SD for each of the samples. Some laboratories (2, 8, 12, 14, 15, 22 and 30) are only for one Youden Plot within these limits. All of these laboratories still produce reliable EQC results. Results from three laboratories are completely outside the +-1 STD limits: Laboratory 24 falls permanently outside the limits in the upper right quadrant and thus has constantly very high values for all positive samples. It is only laboratory, which wrongly identified a negative sample as positive (Fig. 3d). The result of this finding is a reduced assay specificity and shows that under these conditions false positive results are possible. Laboratories 13 and even more laboratory 29 fall constantly outside the limits within the lower left quadrant and thus has constantly outside the limits within the lower left quadrant and thus has constantly outside the limits within the lower left quadrant and thus has constantly outside the limits within the lower left quadrant and thus has constantly low PI values for all positive samples. Laboratory 29 is the only laboratory which wrongly identified a positive sample as negative results are possible. The distribution pattern of laboratories indicates mainly problems of systematic errors (outside upper right or outside lower left) suggesting that laboratories 24, 29 and 13 should critically examine possible reasons for this type of

errors e.g. water quality, wrong/old/dirty filter etc.. The analysis of the Youden Plots does not indicate any sign of random error. Comparing the overall results from the Youden plot analysis with the RP96ac(55%) and RP97a (35%) this round achieved the best results (59%) where most laboratories fall within the 1 STD limit.

equival. The second control for the produced are estimated at the poly of our product of an optimizer (a the interval of the second of the control for the objective and control (control for control (in the formula interval) and the time and protein of the doman EAOTAEA. Division for controls are befored an External

i fastitulini ali 1930-let 1.090 loce (katel ve-04- van 100 actuare) Alitevyk die Unitable 1010 adder essays Tilevillift van famali aanariitadend familiaan milit vine iin

יר היין אלי האם היינא להגי איז הפאנדוטים איבורי או לא איז ביו באווידער ביו קריינג ליוידי ובעליוידים ל

- Contraction of the second

If the first part attention that count tableau use still used for upproverliger 10% produces a configuration attention of the first part attention of the second state of the second st

the management to amiliar and

which much an alternational transformer moved.

In general pendities at the second of the generation on the power angula and in proved on a factority (and press a solution - second and (a), it is cause (user in a complete book of the factority of the strangency prove before appringed with aligned the power such that the theory of almost prove (book) and the strangency prove and a basis access to a generator. But there involves the strangence of the strangency prove before a strangency of the strangency of the strangency of the strangency prove (c) it is an involves the generator. But there involves the basis of the strangency prove (c) it is a strangency of the strangency of the strangency of the strangency prove (c) it is a strangency of the strangency of the strangency of the strangency of the strangency (c) it is a strangency of the strangency of the strangency of the strangency of the strangency (c) it is a strangency of the strangency of the strangency of the strangency of the strangency (c) it is a strangency of the strangency (c) it is a strangency of the strangency (c) it is a strangency of the strangency (c) it is a strangency of the strangency (c) is a strangency of the strangency of

nonathway transport

4. DISCUSSION AND CONCLUSIONS

Quality Control/Quality Assurance procedures are essential to testing laboratories as they provide confidence in test results, as well as informing test operators of unacceptable trends in assay performance. The assurance that the test results produced are reliable is not only of importance to the test operator or owner of the animal, but for all outside interested parties. To achieve this, the Animal Production and Health Subprogramme of the Joint FAO/IAEA Division has initiated an External Ouality Assurance Programme.

The return of 19 (68%) EQC test panel results was still acceptable, although the objective is to have 100% return. The EQC test panel was distributed using a courier service.

4.1. Questionnaire

A compiled summary of the information given in the questionnaire is reported in Attachment 1.

QA/QC practices

It can be concluded that several laboratories still need to improve their IQC practices; i.e., they must pay attention to the monitoring and analyses of the IQC data and should regularly check the calibration of their ELISA equipment. Guidelines have been developed to assist counterparts in checking the calibration of pipettes and ELISA readers, and a TECDOC entitled: "Internal Quality Control (IQC) of Competitive Enzyme Linked Immunosorbent Assay for Measurement of Antibodies against Rinderpest and Peste des Petits Ruminants Viruses." has been prepared by the subprogramme of the Joint FAO/IAEA for the routine monitoring of IQC data using Control Charts [8].

Handling of test results, EDI

In general results indicate that there is an acute need to install and use the latest version of EDI. e.g. EDI 2.2. EDI 2.3.. These versions store IQC data on a separate eqstat.qc files, which ease identification, retrieve and manipulation of data considerably. From the information supplied only 4 laboratories use EDI version 2.2.. Eight laboratories (laboratory 2, 5, 12, 13, 15, 18, 27 and 30) indicate not to monitor their IQC data. The Technical Officer will assure the distribution, installation and use of the latest version of EDI.

Power supply and temperature fluctuations

In general results indicate that the situation for the power supply has improved e.g. there are less power problems encountered. At the same time there is a trend indicating that more laboratories are better equipped with alternative power supplies e.g. they are able to switch over to an emergency power net or have access to a generator. But there is still a critical situation encountered in 6 laboratories (2, 11, 12, 20, 21 and 23) because they indicate to have power cuts but have no access to an emergency power supply nor to a generator. This condition has a direct impact on the laboratories with air-condition. Temperature fluctuations from 15 °C - 42 °C may be a reason for inconsistency of results and improvement in this area is necessary.

Equipment calibration

The majority of the laboratories (22 labs) informed that they do not calibrate ELISA equipment (pipettes and reader). This is a very critical since pipetting errors may be the reason for all kind of variation in an assay. All ELISA equipment (reader, pipettes etc.) should be checked and, if necessary calibrated following the procedures as outlined by the producer or the respective protocol or manual. The ELISA reader should be checked with a Standard Absorbance plate at least during each visit of the Technical Officer. A document entitled "The Laboratory Wizard – A practical loose-leaf edition guide for all who want to share and update ordinary information reported from technical staff of diagnostic laboratories world-wide" [17] is available to assist in laboratory calibration and maintenance procedures.

Nineteen laboratories change filters and cartridges, but only 2 laboratories do so after a control (e.g. conductivity control or manufacturer's recommendation). A more uniform and controlled approach towards criteria and frequency in change of filters and cartridges is necessary. The quality of water must be checked on a more consistent basis to eliminate this possibility of assay variation.

Nineteen laboratories reported to use a computer for the reading of ELISA plates and storage of data. For the first time more Pentiums (9 laboratories) than 486 processor-equipped-computers (6 laboratories) are in use. Hopefully this trend will continue. Some laboratories did not supply any information on the availability of computers and it is not clear how they produce and store the data.

It can be concluded that several laboratories still need to improve their IQC practices; i.e., they must pay attention to the monitoring and analyses of the IQC data and should regularly check the calibration of their ELISA equipment. Guidelines have been developed to assist counterparts in checking the calibration of pipettes, ELISA readers and other laboratory equipment [13].

4.2. IQC Data

The analyses of IQC data of the individual laboratories are reported in Attachment II.

As not all laboratories were using the same ELISA kit batch and it was not known by the EQAP coordinator when a laboratory started to work with a new kit, the Upper and Lower Control Limits as shown in the Control Charts in this report are average Upper and Lower limits. Furthermore, any additional information on a specific ELISA plate was also not known by the coordinator; e.g., identification of the test operator, date of testing, and the batch number of the ELISA kit per ELISA plate. This information is very important and necessary for a correct IQC evaluation and should be written on the Control Chart. For instance, if an empty ELISA plate is run several times to test whether a system is functioning, such plates should be properly identified on the Control Charts.

As part of establishing Quality Control/Quality Assurance procedures within a laboratory, the test operators should maintain Control Charts themselves [8]. For the EQAP rounds in future, the laboratories will be asked to submit copies of such Control Charts with all relevant information of the last ± 40 plates for external assessment.

Obviously the test operator should aim to minimize both the 'within plate' and the 'between plates' variation. Furthermore it must be emphasized again that, in the possible event of the value of an Internal Quality Control sample, especially the OD value of the C++, falling outside the UCL and LCL, and the assay still giving a 'correct' positive or negative value to the test samples, the results of that assay should be considered questionable. The assay must be carefully examined in this situation and the cause for the failure to obtain controls within the limits, determined and eliminated.

The latest EDI version should be installed in the computer as soon as possible and older versions (e.g. EDI 2.1., RPEIA) should be deleted. EDI will during installation overwrite any present older EDI version and will also create a new subdirectory 'eqstat.qc' for the automatic storage of IQC data. The existing subdirectory 'Instatqc' or 'Eqstat.qc' and its file(s) will remain unchanged.

In general, the laboratories are producing reliable results as the majority of the IQC results are within the UCL and LCL with acceptable variation. However, some laboratories should take notice of the "within plate" and "between plate" variation in their IQC results and should initiate measures to reduce that variation. The most likely causes for variation of the IQC data are:

i) Water quality

Data from the Questionnaire shows that the majority of the laboratories are using distilled and deionized water. The frequency of cleaning or replacing filters and columns varies from laboratory to laboratory, depending on the type of distiller/deionizer used. The test operator should ensure that the filter/columns are changed as advised in the manufacturer's documentation. If the test operator still suspects water quality to be a problem, it is suggested that an alternative (if available) water source is utilized for the ELISA and results then compared.

ii) The test operator

Where the test is performed by more than one operator, it is almost inevitable that greater variation in results will occur. As long as test operators obtain good test results, there is no problem. However, as part of Quality Assurance, the laboratory should aim for high repeatability and precision. Therefore, it is suggested that test operators carefully compare their results with respect to IQC data and identify any differences. In this way, possible variations in the technique of performing the ELISA may be highlighted and necessary steps taken to decrease the variation.

iii) Pipetting precision

This is an important factor in variation, particularly where small volumes are being pipetted. Often it is the major cause of the differences in variation observed between test operators.

As explained in detail in the ELISA manual, the assay data expressed in OD and PI values for the Cm and the assay data expressed in PI values for the C++, C+, C- and Cc, are used to determine whether or not the test has performed within acceptable limits of variability, and therefore whether or not the test data may be accepted for any given ELISA plate.

While it is likely that, if the value of a control falls just outside the Upper and Lower Control limit, the assay will still give a correct positive or negative value to the test sera, the results as such are questionable. The assay must be examined in this situation and the cause for the failure to obtain controls within the limits determined and corrected. It is not acceptable to carry on testing sera with controls consistently falling outside the limits. Something is clearly wrong and it must be investigated and resolved.

4.3. The EQC Test Panel

With regard to the EQC test panel, 19 laboratories returned EQC results with an overall agreement of 100% for samples 1, 2 and 5 and of 95% for sample 3 and 4, giving an overall agreement of 98%. No sample had to be excluded. These EQC results are the best ever produced with the competitive Rinderpest ELISA.

Overall, the results of this EQAP for the FAO/IAEA rinderpest competitive ELISA show that each participating laboratory had a high proficiency for conducting the assay.

4.4. EQAP/RP/ 1997a and 1998a.

Provisional recognition

Since participation and submission of correct results of the proficiency testing for at least two consecutive rounds is defined as a key element for the EQA programme 12 laboratories qualified for the status "Provisionally Recognized Laboratory".

These laboratories are: 1, 5, 8, 9, 10, 12, 13, 14, 15, 24, 26 and 31.

Recognition

Two laboratories have supplied all information (questionnaire, IQC and EQC) as required during the last two rounds and have qualified for the status "recognition".

These laboratories are: 23 and 30

The recognized laboratories will receive an FAO/IAEA recognition document and this information will be forwarded to OIE and FAO.

Future changes in "recognition" status and focus of EQA programme.

During an IAEA consultants' meeting entitled "The FAO/IAEA External Quality Assurance Programme (EQAP) and Movement Towards a Generic Veterinary Diagnostic Testing Laboratory Accreditation Scheme" and subsequent discussions it was agreed that the category "Provisionally recognized" will disappear. Nevertheless in this report the category "Provisionally Recognized Laboratory" is still used for internal purpose. The category "recognition" will remain. It is emphasized that in order to achieve recognition a laboratory must fulfill and submit all components (Questionnaire, IQC and EQC data) of the EQA programme.

Quality management and documentation is an essential component of the EQA programme. Special attention will be given to calibrating procedures of laboratory equipment (ELISA reader, pipettes, pH meters, temperature measurement of freezers and refrigerators) and the self-monitoring of internal quality controls is encouraged (IQC data) [8, 13]. The recognized laboration in interaction on EAO/AEA recognition document and the mioritation with the mioritation with AO

ammentation ACU to most this status methods on ECOA measurements

Designation (EQAP) and Missement Lowards a Generic Vetermary Disproving Testing Assurence Assureduation volucing and obsorbering lowards a Generic Vetermary Disproving Testing Laboratory Assureduation volucing and obsorbering lowards in why providing the entropy Preventionally recognized with disampear Electronicies in this report the entropy Preventing Recognized Laboratory in this report of the internal purpose. The entropy for the entropy of the entropy Laboratory in this reconstruction in this report the entropy Preventing Recognized Laboratory in this reconstruction and purpose. The entropy for the entropy of the entropy that is under the activity encounters a subserve of the entropy built and polynomials (Carectering). The reconstruction of the TOA programmer

Quainty management and documentation of all estimated compound of the 102A procession. Special attention with the 102A procession of procession of the procession of the compound of the second procession of the second proc

5. RECOMMENDATIONS

1) Following the conclusions and recommendations of a recent consultants meeting entitled: "The FAO/IAEA External Quality Assurance Programme (EQAP) and Movement Towards a Generic Veterinary Diagnostic Testing Laboratory Accreditation Scheme" the three pillars of the FAO/IAEA EQA programme will remain IQC, EQC and information supplied through a questionnaire, but the focus will be on Quality Management and documentation of specific laboratory activities through Standard Operating Procedures (SOPs). It is understood that participation in the EQA Programme will assist in creating a quality management working environment, which will assist participants - especially from developing countries, who do not count with a national accreditation body - to bridge the gap between what they have now and formal national or international recognition of Quality Management and technical competence.

2) Understanding of the principles of assay validation still widely differs. The basis for any EQA participation is a correctly validated assay. The paper from R. Jacobson "Validation of Serological Assays for Diagnosis of Infectious Diseases" is recommended as a guideline to assist the continuing process of assay validation [11].

3) The Questionnaire is considered an essential component of the EQAP! It is urged that the laboratory officers complete the Questionnaire as accurately as possible. The information gathered with the Questionnaire will require regular updating by the laboratory officer in charge, in close collaboration with the test operator, and should be done at least once a year. In some cases the information provided by the laboratory might need some further clarification. This need will be determined by the EQAP Coordinator on an individual basis during future EQAP rounds. Focus will be on information about Quality Management and documentation of specific activities through Standard Operating Procedures (SOPs).

4) The maintenance and calibration of ELISA equipment needs improvement in most laboratories. Specific guidelines have been prepared and will be distributed. Additionally ELISA Standard Absorbance plates will be distributed to measure the accuracy of ELISA readers.

5) The implementation of a routine monitoring of the IQC data by the participating laboratories is a major objective of the EQAP. For that purpose a TECDOC entitled: "Internal Quality Control (IQC) of Competitive Enzyme Linked Immunosorbent Assay for Measurement of Antibodies against Rinderpest and Peste des Petits Ruminants Viruses." has been prepared and will be distributed.

6) For the continued success of the programme, it is of vital importance that participating laboratories keep to the time limits set by the EQAP Coordinator regarding confirmation of receipt of the EQC test panel and the returning of results. If a laboratory foresees problems in keeping to the time limits, it is the responsibility of the laboratory to contact and inform their FAO/IAEA Technical Officer or the EQAP Coordinator immediately.

7) The target of the EQAP is 100% participation by laboratories including the return of questionnaire, IQC, and EQC data. This involves extensive communication between the counterparts, their FAO/IAEA Technical Officer, and the EQAP Coordinator. As the EQAP becomes more of a routine for all involved, it is expected that a higher percentage of returned results could be achieved. To avoid wasting time tracing lost results/EQAP materials, it is recommended that a courier service be used where possible.

6. ACKNOWLEDGMENTS

We would like to thank all the laboratories participating in this EQAP for their contributions and continued support, as well as Dr. John Anderson from the Animal Health Institute, Pirbright, U.K., for the preparation and distribution of the EQC test panels.

Persons interested in copies of the cited references or any further information, please contact:

Head, Animal Production and Health Section Joint FAO/IAEA Division P.O. Box 100 1400 Vienna AUSTRIA Fax: +43 1 20607 Tel. 2600-26053 E-mail: M.H.JEGGO@IAEA.ORG

Or

Head, Animal Production Unit FAO/IAEA Agriculture and Biotechnology Laboratory Agency's Laboratories A-2444 Seibersdorf AUSTRIA Fax : +43 1 2600-28222 Tel. 260028355 E-mail: <u>M.ROBINSON@IAEA.ORG</u>

Further information about the activities of Joint FAO/IAEA Division can be obtained through the internet: <u>http://www.iaea.or.at/programmes/rifa/d3/index.html</u>

8) For the continued suscess of the programme if if it is proportioner that protocols in protocols in the protocols of the programme if it is not space and protocols of the protocol of the protocol of the protocols of the protocols of the protocol of the protocol of the protocols of the protocol of the protocols of the protocols of the protocols of the protocol of the protocols of the protoc

Q (As a support on the Applet's and (2000) paraticipantial, the labor to the publisher site Astrony of quart remains. If y would find data (D) is muchous an environment of the second of the second of their (Standbook) for based officer and the EQAP (Development) of the EQAP (Indemicional environment) remaining to set involved, the second method highwarp essences of the terminated constant and the achieved. (D aspect requests available, the second method highwarp essences of the terminated and a matter of the aspect requests available, the second method (D) (P) mutaneous (C) is a constant second of the achieved. (D) aspect requests available, and the NOMP mutaneous (C) is a constant required of the achieved of the acceleration of the second of the second of the formation (C) is a constant request of the second of the acceleration (S).

REFERENCES

- WRIGHT, P.F., et al., Standardization and validation of enzyme linked immunosorbent assay techniques for the detection of antibody in infectious disease diagnosis, Rev. sci. tech. Off. Int. Epiz. 12 (1993) 435-450.
- [2] JEGGO, M.H., ANDERSON, J., The FAO/IAEA ELISA kit quality assurance programme, The Sero-monitoring of Rinderpest Throughout Africa: Phase 1, IAEA-TECDOC-623, IAEA, Vienna (1991).
- [3] JEGGO, M.H., ANDERSON, J., FAO/IAEA ELISA External Quality Assurance Programme for the IAEA competitive ELISA, Results 1992, The Sero-monitoring of Rinderpest Throughout Africa: Phase II, Vienna (1992).
- [4] JEGGO, M.H., et al., FAO/IAEA External Quality Assurance Programme for the FAO/IAEA competitive ELISA, Results for 1993, The Sero-monitoring of Rinderpest Throughout Africa: Phase II, IAEA-TECDOC-772, IAEA, Vienna, (1993).
- [5] VAN DER EERDEN, B.J.M., et al., FAO/IAEA External Quality Assurance Programme for the FAO/IAEA rinderpest competitive ELISA, Results in 1994, The Sero-monitoring of Rinderpest Throughout Africa: Phase III, Vienna (1994).
- [6] INTERNATIONAL ATOMIC ENERGY AGENCY, Establishment of External Quality Assurance procedures for use with FAO/IAEA kits: Report of a Joint FAO/IAEA Consultants Meeting, IAEA, Vienna (1994).
- [7] JACOBSON, R.H., Validation of Serological Assays for Diagnosis of Infectious Diseases, OIE Review: Veterinary laboratories for infectious diseases, Volume 17 (20), August 1998.
- [8] INTERNATIONAL ATOMIC ENERGY AGENCY, Internal Quality Control (IQC) of Competitive Enzyme Linked Immunosorbent Assay (C-ELISA) for measurement of antibodies against Rinderpest and Peste des Petits Ruminants (PPR) viruses using charting methods, in preparation.
- [9] INTERNATIONAL ATOMIC ENERGY AGENCY, The FAO/IAEA External Quality Assurance Programme (EQAP) and Movement Towards a Generic Veterinary Diagnostic Testing Laboratory Accreditation Scheme, Report of a Joint FAO/IAEA Consultants Meeting, IAEA, Vienna (1998)
- [10] VAN DER EERDEN, B.J.M., The External Quality Assurance Programme for use with the FAO/IAEA Rinderpest Competitive ELISA, Interim Report (RP1995a) 1995.
- [11] INTERNATIONAL ATOMIC ENERGY AGENCY, The external Quality Assurance Programme for use with the FAO/IAEA Rinderpest Competitive ELISA, Interim Report (RP1996a) 1996.
- [12] Colling, A., The External Quality Assurance Programme for use with the FAO/IAEA Rinderpest Competitive ELISA, Interim Report (Rp1997a) 1997.
- [13] The Laboratory Wizard A practical loose-leaf edition guide for all who want to share and update ordinary information reported from technical staff of diagnostic laboratories worldwide.

23E)MIRPHR

- [9] Wetter D.D. W. and Szambedeninger and subtation of heaving hubbed minimum orders. As an Joshnapper for the h-methodist fasticody in informing disease diagnizatio. Rev. Sci. 4631, 164 Eput. J. M. 2011, 177–1891.
- [1] JP 200, M.H. ANDERSON, L. JL. FARRARN ELISA & quality assumed programme, Ph. Sciences and annual of Afrika Darkoghest. Africa: Phase 1, 1831A/TECINSC-613, 1844.
- 1.1. [LCOOC M H: ANDLESON T. LADIAEA [LLSA Extend Quality Assignment forgramming for the IAEA comparitive D ISA (Condity 1994) The Start-monitoring of Routerport Throughout O not Phase D. Compare (1997).
- [10] Distance with set al. EX-ORALL Estated Destity Vasurance Biogramme for the FNOURT is compared in SNe Results for TSPI. The Sate-monitoring of Euclidences [Discontinue et top]. Plane II: DVLA TREEDOC-FPI. RALA, Normal 1999.
- (A) Solve the Set of the Real Market Activity Associated Programme Learning LAG(IALA finitespect company on FLESA Results in 1998) The Set of antituner, M Restricted Transition (2004), Prose H. System (1994).
- Put interface of the Ministry of Ministry (California Andrew) (California of Emmun Muslin) Administry (California for mission) [A GUARA for Department (A GuART) [Constitute) Methy [A DA Marun (DAC)]
- (4) I U CHE COM IN H. VERBRUCH of Secondinizat design in Oraquides of Interstalli Chilerian OR Review. Vocestrainy Informations for infernational Images, Visionic 17 (20): August 1993.
- (a) Printerson (1994). Very up 12:140.5 Markowski, Mallakov Jakowi, Raubity Campor (0.011) or a triangentice (1995). United linewaventhis Action (C-1212A): (a) pressioned if a triangent acting the despect and Fester del Pelon (D.1990). Survey (P.P.) verses from Churchen restlictly, in projection.
- (v) INTERCONCLOMMAL ATRACT CMERCY AGENCY THE EXCENTED ELECTRY UNITS (Asymptotic Degradied (DOAP) and Frank Frankrik Degrad is Construct. Veterand Frankrik Interact Laborations. Accordination Scheme, Equal 104 June 20 (01468). Computants Meeting. (ACA). Veteract (1998).
- [10] VAN DEF EEKDEN, BLIM, The External Quality Accumute Frogrammer for less 2milliple EAOBALA Stodement Competitive HJSA, Internet Report (2019)350 (1995)
- [151] INCLERNATIONNE ATDMIC UNERGY AGDING: The estimated fracting demonstration (Programmic for use with the FAUMATEA Rendspeed formetings) [3] INA Income Investi-(NPUsering Lenger).
- [11] Contrago S., USC External Quality Accurace Programmin for each with the FAO(External Studentics Ed. S.C. Informations (Conference Ed. S.C. Informations (Conference), 1997.
- (4) The transmissy Winstd = Apprecial forection forection guide far all why way to struct and update judinary universation reported from technical coalt of diagnostic laborations would write.

- Attachment I -

Questionnaire Summary

Construction of the International Constructional Constructional Constructional Constructional Constructiona

Most Responses are providing to the first matrices for the first and the first and the first set pointered in the set of t

Rest(masker,P) w Wark Hantiel hait dimments weet black (weet) we black (weet) way my nego particular to the sec

with the Real Dr. outfleening

With degrad to relate product product and characteristic and filles. If (non-subjective) and (no transport encodering) for laboratoric lines [15] we manifeld [15] without meeting and string [11] with responsible. If this encoder and fill 2.2. Filler-matrix large string [17] 0.4 version [10]. If the interface are Proceeding and the bounder and characteristic memory system [17] 0.4 version [10]. If the interface are Proceeding for the bounder and characteristic memory system [17] 0.4 version [17] (1.4 version

heird. Noorrow (2) or a spread-laret polynamic', to just 'the fest insults' with later docubers' the right

The mapping of the laboration of 111, are the 199, this population whether in order of the second order of the second of the sec

Sumple storage

All Educations (De line) whereasture, contrast 4, 2007, at many states carrier 5 whereast states beded 10 Educt. Measureaux (C Educ), Milyrake element system (C Ottor), Nextonex (C Ottor), Sprint states where C Educ (C Educ), Contrast and States (C Educ), Feeder External Contrast Education Frances, and T in Frances, Mitragen (Educ) – Minimum (Educations) (general Executions Exciting Frances, Education (EDUC), Substates (Educ) – Minimum (Educations) (general Executions) a sympthesis (mail to the form (Education (EDUC)) (Substates (Educ) – Minimum (Educations) (general Executions) a sympthesis (mail to the form (Educa-10) (Substates (Educ) – Minimum (Educations) (general Executions) (Educations) (

Summer in the Contract of the

والمحمد والمحمد والمتحمد والمتحمة المكتم التراجي التراجي التكتيب وتجهيل والمنا والمراجع

Questionnaire summary of results

Sixteen laboratories returned the completed and/or updated Questionnaire during this EQAP round (TABLE I.). The collected information categorized by subject per laboratory is attached in Attachment I. The information presented in bold italic format is new or updated since the last EQAP round. Accumulated and updated information of the questionnaire of the last four EQAP rounds of an overall number of 26 laboratories (laboratories 1, 2, 3, 4, 5, 9, 10, 11, 12, 13, 14, 15, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30 and 31) is compiled in Attachment 1. No questionnaire information at all has been received from laboratories 6, 7, 8, 16 and 17.

Power supply/Air condition

Of 26 laboratories 16 (62%) reported problems with the power supply. Six laboratories have both types of power problems, namely "power cuts" and voltage "irregularities", 4 laboratories reported only power cuts and 5 laboratories indicated only voltage irregularities. Seven laboratories do not have any power problem. Regarding the length of period of power problems 7 laboratories have power cuts of less than 12 hours and 13 reported irregular periods. Asked about the frequency of power problems 2 laboratories reported weekly and 15 irregular periods of power problems. Fourteen laboratories use a stabilizer, 3 use a stabilizer but only for selected equipment (e.g. refrigerator). Seventeen laboratories have access to an emergency power supply for their refrigerator and freezer. Sixteen laboratories have access to a generator in case of a power failure. Eight do not have a generator. Sixteen laboratories have an air-condition (8 do not have air-condition). The average temperature was 25.52 °C with a variation of Min. 15°C to Max. 42°C.

Pipettes, Tips and ELISA readers

Most laboratories use pipettes from Biohit Proline® (15 labs) or Titertek (10 labs) followed by Finnpipette (8 labs), Gilsson Pipettman (6 labs) and Socorex (1 lab) and tips from Biohit Proline® (12 labs) and Micronics (9 labs). Less frequently tips from Finntips, Conetreff, Volac 200 and Costar are in use. Twenty-three and 21 laboratories use 5-50ul single and multichannel pipettes respectively. Eighteen and 20 laboratories use 50-250ul single and multichannel pipettes respectively. Eight and 5 laboratories use 250-1000ul single and multichannel pipettes respectively .

The Multiskan Plus Mark II is the most commonly used ELISA reader (15 labs), followed by the BDSL Immunoscan Plus (9 labs) and Multiskan MCC/340 (4 labs).

Handling of test results

With regard to plate reading and calculation of ELISA results, 23 laboratories use the EDI programme: Ten laboratories are using EDI version 2.11., 9 laboratories are using RPEIA version 1.03, 4 laboratories use ED 2.2., 3 laboratories are using RPEIA version 1.01., 2 laboratories use Procomm and 2 laboratories calculate results manually. Some laboratories indicate to use RPEIA and EDI together.

Twenty laboratories have a computerized system SID (17 labs), Panacea (6 labs), EPI-info (2 labs), Access (2) or a spreadsheet programme¹, to link the test results with other details of the field samples.

The majority of the laboratories (13) use the IQC data to determine whether the ELISA plate readings are 'within' limits and can be accepted. Two laboratories indicated that the IQC data are monitored using the 'instatqc' programme. Eight laboratories reported that they do not undertake any IQC monitoring.

Sample storage

All laboratories (26 labs) store serum samples at -20°C, in most cases using Cryopreservation vials (13 labs), Vacutainers (7 labs), Nalgene storage system (7 labs), Micronics (7 labs), Serum storage plates (3 labs) or others e.g. 10 ml tubes (1 lab). Twelve laboratories have access to -80°C freezers and 7 to Liquid Nitrogen facilities. Nineteen laboratories reported keeping a serum bank ranging from 500 to 45.000 samples with an average of 11.328 samples.

Computer/Data Processing

SID (Sero-monitoring Information Database), Panacea and EPI-info are epidemiological computer programs 1

Nineteen laboratories reported that a computer is used for reading of ELISA plates and/or storage of data. For the first time more Pentiums (9 laboratories) than 486 processor-equipped-computers (6 laboratories) are in use. Three laboratories use 386 and 1 laboratory uses a 286 CPU computer. Hopefully this trend will continue.

Water quality and equipment calibration

Twenty-two laboratories use distilled water. Nineteen laboratories have access to deionized and 11 laboratories to bi-distilled water. Nineteen laboratories reported that filters and cartridges are changed in the following pattern: once per year (2 laboratories), twice per year (6 laboratories), every three months (5 laboratories), every month (1 laboratory), three times per month (1 laboratory). Six laboratories reported that cartridges and filters are changed following the manufacturers recommendations (1 lab), conductivity control (1) or "when needed" (4 labs).

Twenty-two laboratories reported that no equipment calibration (ELISA reader and pipettes) procedures are carried out. Two laboratories undertake calibration procedures following the manual. One laboratory checks the accuracy of its ELISA reader by comparing OD readings with another ELISA reader.

Conclusions and recommendations

QA/QC practices

It can be concluded that several laboratories still need to improve their IQC practices; i.e., they must pay attention to the monitoring and analyses of the IQC data and should regularly check the calibration of their ELISA equipment. Guidelines have been developed to assist counterparts in checking the calibration of pipettes and ELISA readers, and a TECDOC entitled: "Internal Quality Control (IQC) of Competitive Enzyme Linked Immunosorbent Assay for Measurement of Antibodies against Rinderpest and Peste des Petits Ruminants Viruses." has been prepared by the subprogramme of the Joint FAO/IAEA for the routine monitoring of IQC data using Control Charts.

Handling of test results, EDI

In general results indicate that there is an acute need to install and use the latest version of EDI. e.g. EDI 2.2. EDI 2.3.. These versions store IQC data on a separate eqstat.qc files, which ease identification, retrieve and manipulation of data considerably. From the information supplied only 4 laboratories use EDI version 2.2.. Eight laboratories (laboratory 2, 5, 12, 13, 15, 18, 27 and 30) indicate not to monitor their IQC data. The Technical Officer will assure the distribution, installation and use of the latest version of EDI.

Power supply and temperature fluctuations

In general results indicate that the situation for the power supply has improved e.g. there are less power problems encountered. At the same time there is a trend indicating that more laboratories are better equipped with alternative power supplies e.g. they are able to switch over to an emergency power net or have access to a generator. But there is still a critical situation encountered in 6 laboratories (2, 11, 12, 20, 21 and 23) because they indicate to have power cuts but have no access to an emergency power supply nor to a generator. This condition has a direct impact on the laboratories with air-condition. Temperature fluctuations from 15 °C - 42 °C may be a reason for inconsistency of results and improvement in this area is necessary.

Equipment calibration

The majority of the laboratories (22 labs) informed that they do not calibrate ELISA equipment (pipettes and reader). This is a very critical since pipetting errors may be the reason for all kind of variation in an asry. All ELISA equipment (reader, pipettes etc.) should be checked and, if necessary calibrated following the procedures as outlined by the producer or the respective protocol or manual. The ELISA reader should be checked with a Standard Absorbance plate at least during each visit of the Technical Officer. A document entitled "The Laboratory Wizard – A practical loose-leaf edition guide for all who want to share and update ordinary information reported from technical staff of diagnostic laboratories world-wide" [17] is available to assist in laboratory calibration and maintenance procedures.

Nineteen laboratories change filters and cartridges, but only 2 laboratories do so after a control (e.g. conductivity control or manufacturer's recommendation). A more uniform and controlled approach towards criteria and frequency in change of filters and cartridges is necessary. The quality of water must be checked on a more consistent basis to eliminate this possibility of assay variation.

to durine 1 out level of conclume r new memory repromotions, and a

Nineteen laboratories reported to use a computer for the reading of ELISA plates and storage of data. For the first time more Pentiums (9 laboratories) than 486 processor-equipped-computers (6 laboratories) are in use. Hopefully this trend will continue. Some laboratories did not supply any information on the availability of computers and it is not clear how they produce and store the data.

11 Information to the damilied years (National Internet reported that finite and anti-stars are clanged in the following partors where per year (2) aboratories), invest (2) and (2) area internet). Such months [15] laboratories), every minitia [1] theoratorie), three takes per minitia [1] theoratories). So laboratories, reported that extualges and filters are electricit fallowing the manufactures recommendations (1) lab), conductively, control (1) in "when nearly [2] laboratories.

presentative required out [we hitten-sectors undertained allocation providences to the wine the manual [Operation] Internatives the Jack researces of excPLINA tender by compared, OD contractive and the JITTEN researce

and the second second mention of the second

In an ECONOMICAL TRUE REPORT EXECUTION AND A CONTRUCT IN THE TOT INFORMATION IN THE ADDARCE IN A DATA AND A

NGE galuma hat to southed

(a) IDE (2) IDE (a) IDE (a) IDE (basic for the interview and the optical and the latest vector and the little optical and the optical interview of the latest vector and the optical and th

section of the sectio

imperies contents

(pipeltar and reader). Effective a very section and gradient protocolored that the object (c.d. by opported variation of an axe-2. All ELESA equipment (rquber piperion are) should be deschool of all lead of calibrated following the procedures as outlined by the product writh corrected and disclosure (EESA reader blocking the disclosed with a Standard Absorbane fullier) and the disclosed of the EEESA reader blocking the disclosed with a Standard Absorbane fullier of the disclosed of protocol function of the procedures as outlined by the product writh corrected protocol or manual. The EEESA reader blocking the disclosed with a Standard Absorbane fullier of the disclosed of protocol function of the disclosed with a Standard Absorbane function of the distribution of the formation of the standard with a Standard Absorbane function of the distribution of the formation of the disclosed with a Standard Absorbane function of the distribution of the formation of the disclosed with a Standard function of the function of the distribution (or all when even for these and updates induces provide to the distribution of the distribution of absorbanes with by which [157] is available to show in function of the distribution and construction of distribution procedures.

reg continents control or insortance change others and carriages, but roly 2010 informer do to after a control of rowards presents control or insurated areas (economicalitation). A other mutation and controlled approach, forwards presents theorem in change of fitters and carriades. In necessary, the qualities of water interbe checked as a more sequenced transition (or diminute troi possibilities), assay to obtain

Startmart 1

Amerimani

SUMMARY EQAP/RP/1998A QUESTIONNAIRE Pipettes, Tips and Readers (February 1999)

				ody w			ourgic culanticity	arau		Multi channels	mels		F.I.ISA reader	PT -
ઉ	Gilson	Others	Micro-	Biohit	Others	5-50ul	50-	250 -	5-50ul	-05	250.	Multichan	Acutetalana	
Pipet	Pipetteman		nics	Proline			250u]	1000ul		250ul	1000ul	MCC/340	Plus Mart II	Imm shar Plus
			×			x	×	×	×	×		X	IT VIDEM COLL	MILLINAAD FUUS
				x		×			×			¢	~	×
				×		×	x	×	×		50-200			T
					Finntips	x	×	0.5-10ul	×	×			¢	^
						×	×		×	×			,	V
													<	
										T				
-				×		×		×				,		
-				×		×	×					<		
				×		x								x
	×				Conetreff	×	×	,	.,	,				×
			~			×	*		• •	< ,	×		Multiskan +?	×
×					Titertek/Finn tips	-			• •	<			1 itertek	
x		Socorex		×	300-1000ul	×			· / >	50.000		ourgano -	Urganon Leknika reader 280 S	280.5
									•	0000		INIW	Multiscan Flus Lab System	tem
										T				
			×			×			,	,				
-			×	×		×	×		-				x	
Jeneon Seal, Volac, BDSL, Biohit Proline					Volac 200	×	×	-	,	< ,			×	
			×	1-			.,		<	< :				×
				×			• *		~	×	×	MCC		
	×		×			×			• •	,			×	
	-			×		×	×		:	•			×	T
-			×			×	×		; ,	• •			×	
	-			×		*		,	< ,	< ;			x	
			*						ł	<	×	×		X
	 ×	T	< >	T		,	1			×	x		×	
-				,			< :	×	×	×	×		x	
F	,					~	×		×	×			x	
	×	1		×	Costar	×	×	x	×	x			×	
	and				and the second	10-100	-			×				

Note: bold indicates update

Attachment I

SUMMARY EQAP/RP/1998A QUESTIONNAIRE	Power supply	(February 1999)
SUMMARY		

۲	Length of period		S 1	Frequency of periods	iods	Lype of nower problems	of	Stabilizer	emergency	Access to	Airco	tn lab
	with supply problems:	irremilar	weeklv	with power proteins. monthly in	irregular	no power	voltage	used	power supply*	generator	in lab.	0
		minguin					×	yes	yes	yes		
×					• •	*		ves	ou	NO	no	24
		X				4 >	,		Ves	yes	ou	22
×			-		×	<	e	Vec	ou	ou	yes	26
1 5		-						26	VPC	Ves	оп	17-25
		x			×	×	×		22/			
1								94	Ves	Ves		22-30
no problems							,	Ves	ves	yes	ou	22
-		×	×		×	x	<	14 Ca		ou	ves	33
		×			x		×	UI	24	ou	Vec	22-30
		×			×	×		1	no	IIO	22	0.00
								2	6	6	yes	30-42
no problems							×	yes	yes	ои	yes	25
		×						Ves	yes	yes	yes	25
no problems												
												and the second
	3							1100	Ves	Ves	yes	25-26
no problems								yus ant all an	Vec	ves	ou	30
yes		x			×		×	Ilot all cy.	or .		e	15-25
		×			×	×	×	Shi Chi	01	04	Ves	15-42
		×			×	×	×	8	211	Sel	ves	28
no problems								yes	10	2	Ser.	23
		x			×	×		yes	OII	Ves	Ves	27
no problems								5		cometimes	Ves	25
		×			×	×		01	ATT			16-20
×	>		×				×	yes	yes	, AG	21 Jee	20
:		×						not all eq.	Ves)12	36	00.00
X		¢			×			yes	yes	yes	yes	77-07
x					.,	×		not all eq.	yes	yes	yes	25-30
-		×			< >		×	Ves	yes	yes	yes	24
×					×			,	yes	yes	по	
no problems		Solar and							*	ter and deseferance		

Note: bold indicates update

Attachment I

6

SUMMARY EQAP/RP/1998A	QUESTIONNAIRE	Handling of test results	(February 1999)
SUMMAR	QUES	Handli	(Fel

1

Lab	Using EDI		If yes, which	If yes, which EDI program	л П	Linkage test-results with other details of source sample	f source sample	Net.	IQC m	IQC monitoring
no.	yes/no	RPEIA 1.01	RPEIA 1.03	3 EDI 2.11	EDI 2.2	computerized	other	yes/no	s; Instat	Automeas. of IQC
-	yes			x	×	yes, SID		yes		
2	yes				x	yes, SID, Excel	00	ou		Tavo
~	no, manually	no, manually and spreadsheets	ets			no	T.	yes		54 20000 I
-+	yes			x		SID: at present Problems, EPI INFO		yes		8
S	yes		×			SID 3	8	ou		of tool
9										18
7							90			
∞							3			
6	yes		x	x		yes, SID	3	ves		
10	yes			x		yes, Access, EPI-info			ves	of Londo
Ξ	not yet									8
12	yes			x		ou	manual	8		14
<u>с</u>	yes		x			yes, Panacea		Q		
7	no						-			1 0V00 200
15	yes	3	2	2		Procomm, SID 3, Excel		ou		A 33800
16								1		
17	-						3			ar 1,000
18	yes		×			SID 3	ES.	ou		Red and
19	yes				x	SID3	manual	ves		4 1220 L
50	yes			×		SID 3, Access		ves		ST 14004
51	yes		×	x		OU	EPI-info*			
57	yes			×	_	yes, SID/Panacea		yes		
53	yes			×		SID 3		yes		
5	yes		x			yes, SID 3, Panacea 2		yes		- 1200B
25	yes	×	x	not yet	_	yes, SID 2		yes		0004 65
26	yes		x			yes, SID/Panacea 2	100	yes		R 12000
27	yes	×	x			no	manual	no	100	APP 200
28	yes					yes, SID	244	yes		< 30 (M)
29	yes				x	yes, Panacea		in the second	yes	1007201
õ	yes	x		×		yes, SID/Panacea/Procom		ou		Land Paulo -
31	Ves				Þ	VP6 STD3				

Attachment I

Note bold indicates update

SUMMARY EQAP/RP/1998A QUESTIONNAIRE Sample storage (February 1999)

			Comme ton	"onian eao"			natore	ALLESS IN		Output of the ou
			TOTE TITI TOC	SCIUII SIOI ago anns.			at:		Liquid	Serum bank
	Cryo-	Cryopres	Micronics	storage plates	Vacutainers	Others	-20	-80	nitrogen	yes/no
īd	preservation	OK INALIZENCE STOP	CANED TATELY			CHINA STATE	×	yes	ou	yes, 20.000
		x					×	ves limited space	ves.limited space	yes, 800
	×				,		: ×	ves	ou	yes, 45000
	x				<	C (112) 1911	* *	Ves	ves	yes, 4000
	x						•	and a	ho	ves 13000
	×		×				X	IIO	3	100 200
						CILV I				
								22	Of	ves 15000
		x					×		ou	vec 1550
	×					1010	×	yes, innited space	ou	viec 2500
			x		×	0.00	×	· OII	IIO	100, 2200
	~						×	no	0I	yes, i tuu
							×	yes	yes	2
	x			,	×	R F CH2 SHOW	×	yes	no	yes, 22800
		x		< >			×	ou	ou	yes, 7000
		×		<					2	
						Par line -	,	O#	un Du	DO
	×							AT 1	Trac	upe 15000
	×		×				×	IIO	ent i	100,000
					x		×	no	yes	yes, 200
			×				x	ou	ou	DO
	IIO		; ,				×	DI	no	
			×				×	x	no	yes
	×		;			5012	×	ou	no	yes, 4000
	x	×	×		10 1 1 10 1 10 1 10 10 10 10 10 10 10 10		,	UL UL	ou	ou
					×		< ×	ves	yes	yes, 20000
	x				×	A NUD EXC		ou	ou	yes, 13000
		x	×			110 000	; ,	6	6	2
				×	IIIC	10ml tithec	<	ves	no	yes, 7000
	I DE					TOTIN MOCS	< >	1/00	ves	ves. 17000
	1 103 44	x					< 1	303	VPS	ou
							X	yes	yca	011

Attachment I

Note : bold indicates update

⁸
SUMMARY EQAP/RP/1998A QUESTIONNAIRE Computers Data processing (February 1999)

Note : bold indicates update

Attachment I

SUMMARY EQAP/RP/1998A QUESTIONNAIRE Water quality and Equipment calibration (February 1999)

			F			
		De distilled	ves/no	Frequency	yes/no	11 Yes, now
Deionised	Distilled	parmstn-tg	AVE INC.	the second se	VES	according to manual
×	×	×	yes	otice bet mount		
		×	no		IIO	
			yes	if needed	no	(W) [
		*	Ves	every 3 months	yes	ELISA reader as per manual (1)
	×		, tree	if needed	ou	
	x	×	14			
					211 4110	
	_					
				conductivity control	ou	
	x		yes I	annual amonthe	ELISA reader	OD reading compared in other reader
	x	×	yes	STRIIGIII C (ISAS	not test	
			no		1101.751	
	*		ou	0.000	°,	
			Ves	3 x per month	оц	
			Ves	6 moths		
		; ,	ves	6 months	ou	
	×					
				3 months	ou	
	x		yes	l'enoded	ou	
	x		yes	II liecaca		
			yes	every 3 months	011	
		×	ou		ро	
			1111	3 months	no	
	×		Ves	6 months	no	
	< ;		Ves	once per year	DO	
	<			annually	оп	
	×		tiac	when necessary	ou	
	×	×	6		QL	
	×		Sell	twice a year	ou	
	×	×	746	twice a vear	DO	
	x		526	manufacturer's recom	no	new ELISA reader (Sep. 95)
	x x		ýes	International and a second	G	
			yes	SILINOIL O		-00
				* FI ISA reader and Pipettes		rpy saque. XIS

10

Attachment I

Note : bold indicate update

Let all set unwinders vie twole two set of the fill will will be the

- Attachment II -

IQC Control Charts

A second administration patternicity of a subset by the books administration of a second product in provider a based administration patternicity with a statement test the bases is not actic product and many in the second is provided. These pares is a statement test the bases is not actic provider to the base of one are difficult to the second is a spanning in the base is the base is not actic provider to the base of a second is provided. These pares is a statement to the base is the base is not actic provider to the base of one are difficult to the base is a spanning in the base is a first the base is a state of the base is a statement of the provided control of the base information of the base is a base is a space base in the base is a statement of the base is a statement of the base information of the base is a statement base.

- A Schert with the Otter Content for Instance of the Otter Science History and welfs of such for the Content for the Instance of the Association description of the Instance
- (i) a chiral with the Pit Solice South Complex Condition and Complex Condition (Star wells for parts of using Solicity) with investigation of the Solice Solice (Frontion 1977) of Solice Well
- a de seine heith airs d'harden of her the up the formula of the light of the second first from the second secon Sector for the first second from second second

A model between a biometric matter and the state a conclust the big cound process of the bayes are limited when a biometric matter licence with a conclust the big cound process (points) and there is the Council Charm, in the opposition of county there and there are boot to be been a which definitely between the base of search of the county there are been been been been been and been also and the test meretics date of metals, and the batch and the ball is the boot test of the batch of the test meretics date of metals, and the batch and been been ball by the realcounty the county of the test meretics date of metals, and the batch and be also be back with the county the county of the test meretics date of metals, and the batch and been been ball by the back of the ball by the county and the test metals of the batch and be also been been been been been been and the back of the test metals of the test of the batch and be to be and the ball by the batch and the back of the test metals and the test of the batch and be to be also and the back of the test metals and the test of the batch and be back on the ball by the back of the and the back of the test metals are the states the back of the batch on the test metals of the and the ball by the back of the and the back of the back of the back of the test of the back of the back of the test.

(a) part of establishing "partice Control/Oracity Associate processing purpose interval and a part of establishing "partice formation of the second statements of the second statement of the second statements of the sec

The viscos of the 10% data presented on the Vision of the moder of the 100 AP open separation the end exclosion of the basis per 10% supplies of Chapter (Downland, Downland, The minin terms is until stability and exclosion of the gravithe static end to a positive context on a second to a flat Standard (Standard et a) and the flat the Standard Devision of the many of the fear with a second to the flat the flat of the second response of the sample of the many of the fear with a second to the flat the flat of the second response of the sample of the many of the fear with a second to the second of the second being the basis of one 10% submine well the many of the fear with a second of the first term of the second of the second of the many of the fear with a second of the fear term is a second of the fear with a second of the term is the second of the term is a second of the second is the second of the s

1.2-A prista (the Loeth) at version particle user when the transmitter in the product of the second particle. Additionally, a finish representative which is a finish.

in the second second

EVALUATION OF THE INTERNAL QUALITY CONTROL (IQC) DATA

Fifteen laboratories: 1, 2, 5°, 9°, 10, 11, 12, 14, 15, 22, 23, 24, 29, 30 and 31 returned IQC data but information only from 13 laboratories could be evaluated. Evaluation from some laboratories e.g. laboratory 14, 30 and 31 show that the assay is well within limits. Intra- and interassay variation is well under control and also the statistical parameter show a good degree of consistency. IQC results from the majority of laboratories e.g. laboratories 2, 12, 15, 22, 23, 24 and 29 show that there are still some outlayers and further adjustment and consistency is required to maintain the assay under control. Finally there is a group of laboratories e.g. laboratory 10 and 11, which apparently needs urgently substantial adjustment in the performance of the ELISA because almost all data fall outside the upper or lower control limits. In these cases it is obvious that the assay is not under control and must be adjusted as soon as possible. There is a general trend indicating low OD values for the Cm. These values are often very close or below the lower control limit (OD < 0.4). Possible reasons for this may the use of old and/or not properly stored reagents or if encountered in a freshly supplied assay wrong dilution/concentration of reagents. The producer has been informed about these findings.

In this interim report, the following control charts of the IQC data have been plotted and are presented :

- i) a chart with the OD values for the Cm (mean of the OD values of the four wells of each Control Sample, with an error bar of ± 2 standard deviations) per ELISA plate,
- ii) a chart with the PI values for the C++ and C+ (mean of the PI values of the four wells for each Control Sample, with an error bar of ± 2 standard deviations) per ELISA plate, and
- iii) a chart with the PI values of the C- and Cc (mean of the PI values of the two wells for each Control sample)

As not all laboratories were using the same ELISA kit batch and it was not known by the EQAP coordinator when a laboratory started to work with a new kit, the Upper and Lower Control Limits as shown in the Control Charts in this report are average Upper and Lower limits. Furthermore, any additional information on a specific ELISA plate was also not known by the coordinator; e.g., identification of the test operator, date of testing, and the batch number of the ELISA kit per ELISA plate. This information is very important and necessary for a correct IQC evaluation and should be written on the Control Chart. For instance, if an empty ELISA plate is run several times to test whether a system is functioning, such plates should be properly identified on the Control Charts.

As part of establishing Quality Control/Quality Assurance procedures within a laboratory, the test operators should maintain Control Charts themselves [8]. For the EQAP rounds in future, the laboratories will be asked to submit copies of such Control Charts with all relevant information of the last ± 40 plates for external assessment.

The values of the IQC data presented on the Control charts in this EQAP report represent the mean values of the 4 wells per IQC sample ± 2 Standard Deviations. The mean value is indicated with a small circle/square/star, and the ± 2 SD range is given as a vertical error bar. The Standard Deviation or the length of the error bars is an indication of the 'within plate' variation. If the error bars are long, it means that the Standard Deviation of the mean of the four wells is high; there is a high variation between the 4 wells of one IQC sample, and therefore the 'within plate' variation is high. The 'between plates' variation represents the variation between the mean values of each IQC sample of different ELISA plates (the Coefficient of Variation (%) can be used as an indicator and is given together with other basic statistical parameters per IQC sample). Additionally a linear regression trendline, which

^{*} IQC data could no be evaluated due to lack of information e.g. diskette empty, only one printout

permits to show on one view as whereto the test is drifting is plotted for the mean values as shown in Fig. 2 below.

Obviously the test operator should aim to minimize both the 'within plate' and the 'between plates' variation. Furthermore it must be emphasized again that, in the possible event of the value of an Internal Quality Control sample, especially the OD value of the C++, falling outside the UCL and LCL, and the assay still giving a 'correct' positive or negative value to the test samples, the results of that assay should be considered questionable. The assay must be carefully examined in this situation and the cause for the failure to obtain controls within the limits, determined and eliminated.

Apart from the Control charts, the basic statistical parameters of the mean values of each IQC sample are presented in tables per laboratory.

The latest EDI version should be installed in the computer as soon as possible and older versions (e.g. EDI 2.1., RPEIA) should be deleted. EDI will during installation overwrite any present older EDI version and will also create a new subdirectory 'eqstat.qc' for the automatic storage of IQC data. The existing subdirectory 'Instatqc' and its file(s) will remain unchanged.



Fig.2. Example for IQC control chart.

The following upper and lower control limits are presented in the graphs:

		UCL	LCL
OD	Cm	1.0	0.4
PI	C++	100	80
PI	C+	81	55
PI	C-	25	-25
PI	Cc	105	95

Attachment II

3

permuts to show an one view as whereto the test is duffing is plaued for the mean values is shown all Fig. 2 tetow

Obviously the field operator should and to complete their metwork plate and the bolyages plates' variance. Furthermore terms to emplete of gam that in the possible event of the value of vainternal Qualos Control complete especially the OD value of the C = fulling nonode the ICT and 1171 and the assay still grains a correct' period period of manors value to the test samples the results of the assay should be considered questionable. The assay must be carefully "commed in the valuation and uncaracter to the bolt of the value of the test samples the results of the assay should be considered questionable. The assay must be the carefully "commed in the valuation and uncaracter to the bolt of the value to the test within the firmits determined and dominated.

Agant from the Control oburise the basic superiority pranotics of the mean solution of each of it.



really a choose (c) (c) and a ground of a second seco

(but to be a set of the second state of the second of the second second second second second second second second

	10	

Concentrative-

Evaluation of IQC data of Laboratory 1

General observation and summary:

This laboratory has submitted all EQAP components. It identified correctly all EQC samples and had performed well in the last round.

IQC data:

IQC data from 6 plates were submitted as a 3 KB Rpbrep.qc file. One plate was totally outside limits and deleted. The remaining 5 plates are displayed below. The laboratory should use the latest version of EDI to ease organization and analysis of IQC data. More data are needed (>40 plates) to get a realistic picture of the assay performance. The OD values for the Cm from 3 out of 5 plates are below the lower control limit. Intraassay variation is acceptable. The assay comes back on track but needs further observation.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate



--<u>a</u>-- PI C++ --x-- PI C+ ----- Linear (PI C+) ----- Linear (PI C++)

Control chart with mean PI values ± 2 SD and trendlines for C++ and C+ per ELISA plate



Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

TABLE BASI	OD Cm	PI C++	PIC+	PI C-	PI Cc
Mean	0.43	86.75	44.30	1.70	96.80
Standard Error	0.05	1.17	4.97	1.44	1.07
Median	0.30	86.00	56.00	1.50	96.00
Standard Deviation	0.24	5.25	22.22	4.55	3.39
Sample Variance	0.06	27.57	493.59	20.68	11.51
Range	0.59	18.00	71.00	14.00	8.00
Minimum	0.18	77.00	-3.00	-5.00	93.00
Maximum	0.77	95.00	68.00	9.00	101.00
Coef. Variation (%)	54.97	6.05	50.15	267.49	3.50
Count	5.00	5.00	5.00	5.00	5.00

Evaluation of IQC data of Laboratory 2

General observation and summary:

This laboratory had submitted only EQC data during the last round.

IQC data:

IQC data from 37 plates were submitted in two archives as Rpb2rep.qc and Rpb4rep.qc files. The laboratory should use the latest version of EDI to ease organization and analysis of IQC data. The average values for the OD of Cm are within limits, but a number of values plates is below the lower limit and there is a considerable interassay variation. The trendline shows a continuos decrease. Interassay variation must be brought under control.



. Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate





Evaluation of LQC data of Laboratory



Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

Can per ist is A phile	OD Cm	PI C++	PI C+	PI C-	PI Cc	_
Mean	0.59	81.54	59.79	-7.22	84.84	
Standard Error	0.03	1.43	1.12	15.70	2.89	
Median	0.58	86.00	63.00	15.50	94.00	
Standard Deviation	0.28	17.32	13.53	135.04	24.85	
Sample Variance	0.08	300.10	183.06	18236.17	617.59	
Range	1.30	86.00	60.00	954.00	122.00	
Minimum	0.02	10.00	26.00	-854.00	-8.00	
Maximum	1.33	96.00	86.00	100.00	114.00	
Coef. Variation (%)	47.35	21.24	22.63	-1871.36	29.29	
Count	37.00	37.00	37.00	37.00	37.00	_

TABLE BASIC STATISTICS IQC DATA LABORATORY 2

Evaluation of IQC data of Laboratory 5.

General observation and summary: This laboratory had submitted all EQA data. All EQC samples were identified correctly.

IQC data:

The file did not contain any IQC data.

Pyahuatton of IQC data of Laboratory 5

Leneral observation and second s

vys dan Die Reedal on anzenaar 190 dae

Evaluation of IQC data of Laboratory 9.

General observation and summary: This laboratory had submitted all EQA data. All EQC samples were identified correctly.

IQC data:

The file did contain IQC data from 3 plates only, which could not be used for analysis.

Evaluation of its data of Laboratory 9

viummur limit and a pasta income 6

This laboratory but submitted all EQA data. (U.U.QC statibles form depitibled converts

state 101

the true and summing the status from " planes only, which leaded not be under sumbars.

Evaluation of IQC data of Laboratory 10

General observation and summary:

This laboratory returned all EQA components. All EQC samples were identified correctly.

IQC data:

IQC data from 37 plates were submitted in two archives as Rpb2rep.qc and Rpb4rep.qc files. The laboratory should use the latest version of EDI to ease organization and analysis of IQC data. The average values for the OD of Cm are practically the same as the lower control limit. The first 20 plates are far below the lower control limit. Then values exceed the upper control limit. It seems that only the last five plates are within limits and the further performance of the assay needs to be monitored for any further judgment.



. Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate





Control chart with mean PI values ± 2 SD and trendlines for C++ and C+ per ELISA plate



Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

ITTI I DOD IT

. .

	OD Cm	PIC++	PIC+	PI C-	PI Cc
Mean	0.41	75.79	57.83	20.91	82.58
Standard Error	0.04	4.00	4.60	8.42	4.57
Median	0.26	90.00	70.00	13.00	96.00
Standard Deviation	0.39	48.46	55.81	72.47	39.34
Sample Variance	0.15	2348.77	3114.40	5251.73	1547.78
Range	1.38	333.00	607.00	656.00	242.00
Minimum	-0.01	-162.00	-400.00	-49.00	-57.00
Maximum	1.37	171.00	207.00	607.00	185.00
Coef. Variation (%)	96.45	63.95	96.50	346.65	47.64
Count	37.00	37.00	37.00	37.00	37.00

Attachment II

Afrachment 11

Evaluation of IQC data of Laboratory 11.

General observation and summary:

This laboratory returned all EQA components. All EQC samples were identified correctly.

IQC data:

IQC data from 18 plates were submitted. The operator should check whether the latest version. of EDI is in use. The average values for the OD of Cm and C++ are too low. The operator should carefully check whether the controls are placed in the right wells on the plate. It seems that they were mixed up. The values in the basic statistics do not make sense. The assay is not under control.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate



Control chart with mean PI values ± 2 SD and trendlines for C++ and C+ per ELISA plate



Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

	OD Cm	PIC++	DATA LABO PI C+	PI C-	PI Cc
Mean	0.38	22.94	299.61	-127.78	-97.06
Standard Error	0.05	22.35	83.00	46.95	38.13
Median	0.36	83.00	53.50	4.00	56.00
Standard Deviation	0.42	189.66	704.25	281.72	228.79
Sample Variance	0.17	35969.29	495965.00	79365.38	52345.77
Range	1.27	952.00	2479.00	1473.00	505.00
Minimum	-0.21	-419.00	-360.00	-940.00	-406.00
Maximum	1.06	533.00	2119.00	533.00	99.00
Coef. Variation (%)	109.40	826.59	235.05	-220.48	-235.73
Count	18.00	18.00	18.00	18.00	18.00

Evaluation of IQC data of Laboratory 12.

General observation and summary:

This laboratory returned all EQA components. All EQC samples were identified correctly.

IQC data:

IQC data from 7 plates were submitted. The operator should check whether the latest version of EDI is in use. The average values for the OD of Cm and C++ are a bit too high. More plates needed to be analyzed to come to a final conclusion. From the little data supplied it looks that the assay is on the right track.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate



Control chart with mean PI values ± 2 SD and trendlines for C++ and C+ per ELISA plate

120 UCLCc 100 LCL Cc Ť 80 PI Values 09 09 09 09 09 UCL C-Ŧ 0 LCL C--20 -40 7 6 5 3 4 2 1 **ELISA Plates** ----- PI C- ----- PI Cc ----- Linear (PI C-) ----- Linear (PI Cc)

Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

TABLE BASI	OD Cm	PI C++	PIC+	PI C-	PI Cc
Mean	0.96	91.14	54.39	12.00	98.00
Standard Error	0.02	0.27	0.79	1.10	0.48
Median	0.96	91.00	55.00	11.50	98.00
Standard Deviation	0.09	1.41	4.16	4.13	1.80
Sample Variance	0.01	1.98	17.28	17.08	3.23
Range	0.37	5.00	17.00	17.00	7.00
Minimum	0.78	88.00	45.00	4.00	92.00
Maximum	1.15	93.00	62.00	21.00	99.00
Coef. Variation (%)	9.85	1.54	7.64	34.44	1.83
Count	7.00	7.00	7.00	7.00	7.00

This Physical Contract (1994) and the second s

with their metroscore? And the local collection of the second second second second second second second second

11 위에 여러 우리하는

Evaluation of IQC data of Laboratory 14.

General observation and summary:

This laboratory returned all EQA components. All EQC samples were identified correctly.

IQC data:

IQC data from 42 plates were submitted as printouts. One outlayer was excluded. The reason why this laboratory can not store IQC data electronically must be identified and the latest version of EDI should be installed. Average values from basic statistics and all values in the graph are within limits, with very little intra- and interassay variation. The assay is under control.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate





Control chart with mean PI values ± 2 SD and trendlines for C++ and C+ per ELISA plate



Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

TABLE. BASI	OD Cm	PIC++	PIC+	PI C-	PI Cc
Mean	0.60	80.05	55.98	8.90	86.56
Standard Error	0.01	0.94	0.53	1.01	1.29
Median	0.55	83.88	56.34	8.62	90.95
Standard Deviation	0.10	11.99	6.83	9.11	11.70
Sample Variance	0.01	143.80	46.65	83.06	136.91
Range	0.35	116.15	32.16	41.83	92.19
Minimum	0.46	-21.78	38.87	-13.50	1.68
Maximum	0.80	94.37	71.02	28.34	93.87
Coef. Variation (%)	16.40	14.98	12.20	102.39	13.52
Count	41.00	41.00	41.00	41.00	41.00

Attachment II

Alkachunered II

Evaluation of IQC data of Laboratory 15.

General observation and summary:

This laboratory returned EQC and IQC data. All EQC samples were identified correctly.

IQC data:

IQC data from 34 plates were submitted as a rpbrep file. One outlayer was excluded. The laboratory should use the latest version of EDI. The average value for the OD Cm is too low. The average for the C+ is higher than for the C++ and it may be that controls were mixed up. The performance is clearly improving but the overall values indicate that the assay is not under control.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate



Control chart with mean PI values ± 2 SD and trendlines for C++ and C+ per ELISA plate



Control chart with mean PI values \pm 2 SD and trendline for Cc and C- per ELISA plate

TABLE. BAS	OD Cm	PI C++	PIC+	PI C-	PI Cc
Mean	0.33	75.42	82.62	17.23	96.45
Standard Error	0.02	17.11	26.54	26.95	21.07
Median	0.38	98.00	70.00	33.00	97.00
Standard Deviation		195.80	303.79	217.27	169.90
Standard Devlation	0.07	38337.08	92288.76	47206.93	28867.47
	1.14	1785.00	2885.00	1597.00	1103.00
Range	-0.21	-811.00	-766.00	-716.00	-406.00
Minimum	0.93	974.00	2119.00	881.00	697.00
Maximum Coef. Variation (%)		259.61	367.70	1260.95	176.16
Count	33.00	33.00	33.00	33.00	33.00

the manufacture

Evaluation of IQC data of Laboratory 22.

General observation and summary:

This laboratory returned all EQA components. All EQC samples were identified correctly.

IQC data:

IQC data from 40 plates (March-October 98) were submitted and are displayed below. The mean OD value for the Cm is a bit too low (0.39) and the linear regression trendline shows a constant downward move. The activity of the monoclonal antibody is going off. One should use a new vial and test whether the OD values return within limits.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate



--<u>-</u>△-- PI C++ ---×-- PI C+ ----- Linear (PI C+) ----- Linear (PI C++)

Control chart with mean PI values ± 2 SD and trendlines for C++ and C+ per ELISA plate



Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

	OD Cm	PI C++	PIC+	PI C-	PI Cc
Mean	0.39	85.68	56.06	16.33	98.29
Standard Error	0.01	0.94	0.96	1.84	0.69
Median	0.38	87.00	56.00	14.00	98.00
Standard Deviation	0.10	11.83	12.10	16.47	6.14
Sample Variance	0.01	140.06	146.36	271.29	37.70
Range	0.47	126.00	81.00	73.00	32.00
Minimum	0.14	-20.00	26.00	-23.00	81.00
Maximum	0.60	106.00	107.00	50.00	113.00
Coef. Variation (%)	24.74	13.81	21.58	100.89	6.25
Count	40.00	40.00	40.00	40.00	40.00

. TAD

Evaluation of IQC data of Laboratory 23.

General observation and summary:

This laboratory returned all EQA components. All EQC samples were identified correctly.

IQC data:

The diskette with the IQC data contained several folders. The most recent 40 plates were taken for IQC analysis and are displayed below. These are the same values as given in the Rp1997a. For this reason the comment in this report is the same as in the 1997a report.

The linear regression trendline and the basic statistical data indicate that there is no clear distinction between the PI values of the C++ and C+. A new batch of C++ control serum may solve this problem. The average value for the C++ is too low. The OD average value for the Cm is still within limits but is coming too close to the lower control limit. The assay is still under control but action needs to be taken.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate





Control chart with mean PI values ± 2 SD and trendlines for C++ and C+ per ELISA plate



Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

	OD Cm	PI C++	PIC+	PI C-	PI Cc
Mean	0.44	75.85	71.76	27.30	95.90
Standard Error	0.01	0.40	0.42	1.09	1.23
Median	0.43	75.00	73.00	28.00	98.00
Standard Deviation	0.06	5.03	5.27	9.79	11.03
Sample Variance	0.003	25.34	27.80	95.78	121.74
Range	0.26	32.00	29.00	53.00	65.00
Minimum	0.34	57.00	55.00	-7.00	35.00
Maximum	0.61	89.00	84.00	46.00	100.00
Coef. Variation (%)		6.64	7.35	35.85	11.51
Count	40.00	40.00	40.00	40.00	40.00

Attachment II

11 march 2000 A

Evaluation of IQC data of Laboratory 24.

General observation and summary:

This laboratory returned all EQA components. All EQC samples, but sample 4 were identified correctly. Sample 4 was identified as positive (cut-off 50%).

IQC data:

The most recent 40 plates were taken for IQC analysis and are displayed below. These are the same values as given in the Rp1997a. For this reason the comment in this report is the same as in the 1997a report.

The OD average value for the Cm is coming below the lower control limit (0.39) and there is a considerable interassay variation in the first 12 plates with an improving trend. Intrassay variation is somewhat high in all plates. PI values for C++ and C+ are converging towards 80% making a clear distinction between strong and moderate positive difficult. The assay is under control but action needs to be taken.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate







Control chart with mean PI values \pm 2 SD and trendline for Cc and C- per ELISA plate

TABLE. BASIC	OD Cm	PIC++	PIC+	PIC-	PI Cc
	0.39	86.69	72.98	11.19	96.03
Mean	0.01	1.74	1.41	2.39	1.99
Standard Error	0.39	87.50	75.00	13.00	96.00
Median	0.00	21.97	17.82	21.38	17.82
Standard Deviation	0.01	482.57	317.64	457.14	317.67
Sample Variance	0.79	211.00	149.00	143.00	154.00
Range	0.07	-48.00	-29.00	-94.00	18.00
Minimum	0.86	163.00	120.00	49.00	172.00
Maximum	28.49	25.34	24.42	191.11	18.56
Coef. Variation (%) Count	40.00	40.00	40.00	40.00	40.00

Evaluation of IQC data of Laboratory 29.

General observation and summary:

This laboratory returned all EQA components. All EQC samples, but sample 3 were identified correctly. Sample 3 was identified as negative (cut-off 50%).

IQC data:

Thirty-eight plates were taken for IQC analysis and are displayed below. The graph shows OD values slowly coming over the lower control limit but the average is slightly below this limit. Intraassay variation especially on plate 19 and 34 is excessive. Some PI C++ values drop below the cut-off. This may be a reason why one positive EQC sample was identified wrongly as a negative sample. The trend is OK but intraassay variation must be better controlled.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate







Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

00.000

TABLE BASI	OD Cm	PI C++	PIC+	PIC-	PI Cc	
Mean	0.38	71.87	42.18	5.13	85.89	
Standard Error	0.01	3.11	3.52	3.52	2.45	
Median	0.44	82.00	58.00	7.00	93,00	
Standard Deviation	0.17	38.35	43.37	30.65	21.34	
Sample Variance	0.03	1470.55	1881.01	939.18	455.51	
Range	0.81	343.00	261.00	200.00	122.00	
Minimum	-0.12	-240.00	-174.00	-108.00	-16.00	
Maximum	0.68	103.00	87.00	92.00	106.00	
Coef. Variation (%)	44.08	53.36	102.81	597.21	24.85	
Count	38.00	38.00	38.00	38.00	38.00	

Attachment II

H InstallantA

Evaluation of IQC data of Laboratory 30.

General observation and summary:

This laboratory returned all EQA components. All EQC samples were identified correctly.

IQC data:

Fourteen plates are displayed below. All values are within limits and consistent. Intra- and interassay variation is minimal. The assay is full under control.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate



Control chart with mean PI values ± 2 SD and trendlines for C++ and C+ per ELISA plate

Evaluation of IQC data of Laboratory 30



Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

	OD Cm	PI C++	PI C+	PI C-	PI Cc
Mean	0.57	87.45	67.23	13.64	99.64
Standard Error	0.01	0.47	0.54	1.64	0.09
Median	0.56	87.00	66.00	16.00	100.00
Standard Deviation	0.05	3.53	4.03	8.67	0.49
Sample Variance	0.003	12.43	16.25	75.13	0.24
Range	0.24	16.00	16.00	32.00	1.00
Minimum	0.48	78.00	60.00	-7.00	99.00
Maximum	0.72	94.00	76.00	25.00	100.00
Coef. Variation (%)	9.15	4.03	6.00	63.53	0.49
Count	14.00	14.00	14.00	14.00	14.00

TABLE BASIC STATISTICS IQC DATA LABORATORY 30

Evaluation of IQC data of Laboratory 31.

General observation and summary:

This laboratory returned all EQA components. All EQC samples were identified correctly.

IQC data:

Only seven plates are displayed below. Most values are within limits. Intra- and interassay variation is still acceptable. The assay is under control.



Control chart with mean OD values ± 2 SD and linear trendline for Cm per ELISA plate





Control chart with mean PI values ± 2 SD and trendlines for C++ and C+ per ELISA plate

Evaluation of 1QC data of Eatheratory J



Control chart with mean PI values ± 2 SD and trendline for Cc and C- per ELISA plate

	OD Cm	PI C++	PI C+	PI C-	PI Cc	
Mean	0.73	87.54	64.71	14.00	97.79	
Standard Error	0.03	1.33	1.87	2.10	0.21	
Median	0.68	88.50	65.00	13.50	98.00	
Standard Deviation	0.15	7.06	9.88	7.84	0.80	
Sample Variance	0.02	49.89	97.54	61.54	0.64	
Range	0.54	40.00	33.00	23.00	2.00	
Minimum	0.53	54.00	46.00	2.00	97.00	
Maximum	1.06	94.00	79.00	25.00	99.00	
Coef. Variation (%)	20.33	8.07	15.26	56.03	0.82	
Count	7.00	7.00	7.00	7.00	7.00	

TABLE BASIC STATISTICS IQC DATA LABORATORY 31

- Attachment III -

Accumulated Data for Determination of "Recognition"

more that informations takes investigated with the the important of the Fight end of the Fight and the second s

japanen a persis (jaituitjätuutja kiretasiista häte semerelistig elejittind gill el Tim myllioniatiistie mil'ilm Ir el persistenisen elevisen elevistichen intervine itelinen perificiente entit

eri Dorekalar iza ayamadıdır. prozestileri eritiyinde meni pedalalmır viteri eti sedi sətiyati ve eyin i eriye enerili etiriyi. V

of a disc demonstration with the wetherhouse of a supervatorie function in the demonstration of the formation of the second s

Ladings.com 2

(a) All All Although 11: Interview of a Proposition Operation (19) and their 30 at space (a) and (a) (assuming the first) minima, at 30% means for enterview of submated) and their 30 at space (assuming mean obtains (stas) succession at space (astronomic field) are by space (astronomy) and their 30 at space (astronomy) representation (state) (sector astronomy) and (sector (sector (sector (sector (sector)))).

Est fault andreaften purpose this gradient in the second and book establish an "and an and an and an and an and a second and a s

station of a material sector and a subscription provided and a subscription of the sector of the sec

ala "ali "ali badan da satancer 199 gidi. muna titil ni amin' dan ba' da lau" jad matan f bera anda mardi asimiti titina katar mal anyangar mandari ang ba' ang ba' da titi' ang bana an

an estimate the energy (19)C data tricks 10 Shek alastica english estimates and the second second second second the second s

ji epare-cedir bikensisedi. Distriction investoves britis Koffer Koffer (Dec ef Eleve 16 houd concerts for Aplanet Questionation, 14 fase parginal administration in 104, dow test (F. Decenyi might 1959) wordte for this facilit

Accumulated Data for Determination of "Recognition" or "Provisional Recognition"

The criteria for "recognition" and "provisional recognition" are outlined in the Joint FAO/IAEA document, 1994 entitled "Establishment of external quality assurance procedures fur use with the FAO/IAEA ELISA kits" [6]:

1. Recognized laboratory

Criteria: the laboratory that successfully fulfilled all of the requirements of the EQAP for the designated assay including passing the most recent proficiency tests

2. Provisionally Recognized Laboratory

Criteria: a newly participating laboratory has successfully fulfilled all of the requirements of the EQAP for the designated assay including passing its first proficiency test

or

the laboratory has successfully passed the last two or more proficiency tests but has not fulfilled other requirements of EQAP

or

a recognized laboratory has failed the most recent proficiency test

Recognition will be withdrawn if a laboratory fails to meet the necessary EQAP requirements for the designated assay. Laboratories not fulfilling the requirements of EQAP will not be granted recognition.

Criteria applied

In Attachment III the submission of information (Questionnaire, IQC and EQC) is shown first as a quantitative (registration of any result or information submitted) and then as a qualitative information (Analysis/resume of questionnaire (emphasis on quality management e.g. calibration of equipment etc.), IQC data (mean values for Cm, C++, C+, C- and Cc within limits) and EQC data (correct identification of samples and location within Youden plot analysis)

For final evaluation purpose the qualitative information has been reduced to "ok" or "not ok" for each of the three categories.

An "ok" under the category questionnaire means that the laboratory has submitted updated and relevant information about the laboratory infrastructure, facilities, staff, equipment and quality assurance procedures.

An "ok" under the category IQC data means that the mean data of the IQC fall within the lower and upper control limits. Inter- and intraassay variation and the background colour are acceptable.

An "ok" under the category EQC data means that when a common cut-off (e.g. 50%) is applied EQC samples were identified correctly as positive or negative.

Twenty-eight laboratories have participated in the Rp98a. Out of these 16 have sent new or updated questionnaires, 15 have supplied information of their IQC data and 19 have submitted EQC results for this round.

A DIREPUNCTURE

Provisional recognition

Since participation and submission of correct results of the proficiency testing for at least two consecutive rounds is defined as a key element for the EQA programme 12 laboratories qualified for the status "Provisionally Recognized Laboratory".

These laboratories are: 1, 5, 8, 9, 10, 12, 13, 14, 15, 24, 26 and 31.

Recognition

Two laboratories have supplied all information (questionnaire, IQC and EQC) as required during the last two rounds and have qualified for the status "recognition".

These laboratories are: 23 and 30

The recognized laboratories will receive an FAO/IAEA recognition document and this information will be forwarded to OIE and FAO.

Future changes in "recognition" status and focus of EQA programme

During an IAEA consultants' meeting entitled "The FAO/IAEA External Quality Assurance Programme (EQAP) and Movement Towards a Generic Veterinary Diagnostic Testing Laboratory Accreditation Scheme" and subsequent discussions it was agreed that the category "Provisionally recognized" will disappear. Nevertheless in this report the category "Provisionally Recognized Laboratory" is still used for internal purpose. The category "recognition" will remain. It is emphasised that in order to achieve recognition a laboratory must fulfil and submit all components (Questionnaire, IQC and EQC data) of the EQA programme.

Quality management and documentation is an essential component of the EQA programme. Special attention will be given to calibrating procedures of laboratory equipment (ELISA reader, pipettes, pH meters, temperature measurement of freezers and refrigerators) and the self-monitoring of internal quality controls is encouraged (IQC data) [4, 17].

nannegassa lanoistead

bases partangation and submission of correct results of the portanticy testing for at least two consecutive normalities the final as a less element for the EQA programme. If (adjunctances qualified for the status) (normality is a qualified for the first status).

[1] Data and M. S. & S. M. L., I. T. M. L. M. Marketter, M. 1998.

Surgarus

FULT laboral pice have supplied if information opproximation (OC and EQC) corresponded during the fall two forming and have pulpied the theorem in a spatiality. Hiere faller abaretry, 12 and 30.

(Diff. (color model in least firms with means and AOTATAL incompanies, forum and this information will be the color for DP and TAO.

Polyter, Obiographic Company and Company and Company (11) A critical company

Polycionum (190.07) and Messmanic mechanismi that "The ExeCl VCA Constant" (Audro-Accuration Polycionum (190.07) and Messmanic Endenie a Constant Valenary, Daemontar Testing Information Accorditation Scheme' and subsequent dedication of the approximation for tempory "Producedadi (constants) with droutpear Newmitcles) in the report the antiquity Producedadi Esboration is add(great for internal purpose. The constants of committee will report the Their report of addition of the constants for the constants of committee will report the their report of a staff great for internal purpose. The constants of committee will report the their report of a staff great for internal purpose. The constants of committee will be polycourted for their report of a staff great for internal purpose. The constants of committee will be polycourted for their report of a staff great for internal purpose. The constants of committee will be polycourted for their report of a staff great for internal purpose. The constants of committee will be polycourted for the report of the formation of the statement of the statement of the statements of the top of the formation of the statement.

Quality measurement and Seconstance on essential composition by A programmer's Special gravition will be plater in calibritude procedures of futuratory minimum (ELISA reader, population of material temptodute measurement of forecast and reingratory) and the reference of monitoring of internet quality control (consisting) (CDA) (CDA) (CDA).

pest	data
inder	lated
AP R	cumu
Щ	Ac

InH			dual lude	Ve IIII 0		3	Qualitative Into	10	Decile
	Lab code	Quest.	IQC	EQC	Quest.	lac	FOC	Validen	
95a							1 V	19000	Provisional/ Recognition
96a	5	×		×	×		to b	unner richt ordeide	
97a	+	×	×	×	8 K	too low. not ok	ł	central inside	0
98a	-	×	×	×	ok	only 5 plates, not ok	ok k	central inside	
								0500=	
	Lab code	Quest.	ВC	EQC	Quest.	lac	EQC	Youden	
95a									
96a	8			×			ok	tinner richt incide	
97a	3							מלקשי וופוויל ווופות	
98a	2	×	×	×	not ok	ok	ok	1 border, 2 slightly oustside	
	Lab code Quest	Quest		EOC	Ollect	00			
95a	-		2		1	2	2	Tougen	
96a	28	×		×	ek k		ok	central incide	
97a	З			×			5 d	central inside	
98a									
	Lab code	Quest	lac	EQC	Quest	IQC	EQC	Youden	
95a									
96a	26								
7a	4	×		×	ş		ok K	central inside	
98a									
	Lab code	Ottest	00	UU3	0.000	00	000		
95a		10011		L	Magh	140	2 L L C C	Youden	
96a	30		×			ok			
97a	5	×	×	×	ok	ok	ok	ci iteida	
Sa		×	×	×	not ok	no iqc data on diskette	ok ok	inside, lower left	PR
	Lab code	Quest	ac	EQC	Quest	00		Vaudan	
95a						05	LAC	liannoi	
96a	29								
a	9			×			and and		
98a				(central left outside	90
	Lab code	Quest.	lac	EQC	Quest.	lac	FOC	Voliden	
95a							2557	10000	
Sa									
a	7								
00~									

Q

Attachment III

pest	data
linder	lated
AP R	cumu
Ш	Ac

Adam	Info	ā	Ouantitative Info	e Info			Auditauy -		Provisional/ Recognition
Adm		Ouect	IOC	EQC	Quest.	Iac	EQC	Youden	
	Lap code	ולתבאר	0.5						
95a				;			ok	central left, slightly outside	
	25			×			ok	upper right, outside	
97a	80		_	×			ak	lower left, slightly out, 1 border	ХT
98a	8			×					
	Lab code	Quest.	ВС	EQC	Quest.	lac	EQC	Youden	
069					_		10	central inside	
059	31	×	×	×	k		20	central left bordeline inside	PR
8	50	,	~	×	ð	ok	YD.		PR
97a 98a	<u>n 0</u>	< ×	×	×	y	not ok	8		
	Lab code	Quest.	IQC	EQC	Quest.	lac	EQC	Youden	
						-	not ok	central left. slightly outside	
06a	27	×	×	×	¥0		10	central inside	æ
97a	10	×	×	×	8	OK, C++, C+ Signar iow	5 2	inside upper border	PR
98a	10	×	×	×	Ao	LIOT OK	5		
				FOC	Quest.	IQC	Eac	Youden	
	Lab code	Kuest.	2						
808	22						1		
97a	11				-	ant at	k	central inside	
98a	11	×	×	×	ð	LIGI ON			
	Lab code	Quest.	lac	Eac	Quest.	lac	Eac	Youden	
95a									
96.2	80						y c	central inside	PR
97a	12	×	×	×	OK	on John Zingtes	ok	lower left, slightly outside, 1 bord	rd PR
98a	12	×	×	×	NOT OK	UIIY / Place			
	Lab code	e Quest.	Iac	EQC	Quest.	IQC	EQC	Youden	
95a							bk	central left, slighly oustide	
63	13			×			y yo	central inside	PR
7.9	13			×	-		24	Inwer left, outside border	PR
98a	13			×	-		5		
	1 ah code	e Quest.	lac	EQC	Quest.	IQC	EQC	Youden	
659		-					sk.	lower central, outside	
96a	11			×	-				
97a	14			-	14	40	k	2 slightly outside, left, 1 inside	PR
-00		~	×	×	OK	NO.			

pest	data
inder	lated
IAP R	nmus
B	Ac

			Auditulauve 1010				Wualitative Into	0	Docula
	Lab code	Quest.	lac	EQC	Quest.	lac	FOC	Voliden	Devicional V Device
95a							284		Provisional/ Recognition
96a									
97a	15	×		×	ok K		ok k	central lower left slightly outside	
98a	15		×	×		not ok	ok	_	PR
	Lab code	Quest.	DO	EOC	Quest			Vender	
95a						22		Loudell	
96a	23		×	×		ok	Š	central unner left autoide	
7a	16						5	המומו מלאמו ובווי מתפוחם	
98a	16			×			ð	inside central	
	Lab code	Quest.	lac	EQC	Quest.	loc	EOC	Volidon	
Sa							L	lianno	
96a	17								
97a	17								
98a									
	Lab code	Quest.	200	EQC	Quest				
95a							2	Iouuei	
96a	19								
a	18	×	×	×	ok	ok	ok	Inwer left outside	
98a									
	Lab code	Quest.	ac	Eac	Quest.	loc	EOC	Volidan	
95a								Induction	-
96a		×			ok Xo				
97a	19								~
98a		×		x* from 97	7 ok				
	Lab code Quest.	Quest.	Iac	EQC	Quest.	lac	FOC	Voirden	
95a							251	liphon	
96a	20	×		×	k		ok	lower central outside	
97a		×	×	×	8 S	ok V	i ko	ido	
98a									
	Lab code (Quest.	lac	EQC	Quest.	lac	EQC	Youden	
95a									
	15	×		×	ok		ok	upper right slightly out	
97a	21								
983									

Attachment III

2

EQAP Rinderpest Accumulated data

Adm	Adm Info	a	Quantitative Info	e Info			Cualitative mile	Validen	Provisional/ Recognition
IDH	Lah code	Quest.	lac	EQC	Quest.	IQC	2 E C C	1 Outer	
060	Lab						1	central incide	PR
	14	~	×	×	Å		OK		
	50	~	×		ok		-	1 horder 2 outside lower left	
98.9	32	×	×	×	k	not ok, OD Cm too low	OK		
1				COL	Ouet		EQC	Youden	
	Lab code	Quest.	bo	LAC	MUCOL				
95a				-					
96a					10	C++ Inw C- high, not OK I	k	upper right, slightly outside	
97a	23	×	×	×	Yn -	1000	ok	central inside	Y
98a	23	×	×	×	X	1			
	I ab anda	Ouest	100	EQC	Quest.	lac	EQC	Youden	
	Lab coue	1000							aa
95a			,	×	k	ok	ð	central inside	2 A
96a	מ	×	<;		k		ok	central inside	00
97a	24	×>	××	×	k	C++ very close to C+	not ok	extremely outside lower len	
98a	24	×	<						
	Lab code	e Quest.	lac	EQC	Quest.	IQC	EQC	Youden	
95a					-		ok	central inside	PR
96.a	12	×	×	×	¥ ·	On low Chick not ok	bk k	upper right, slightly outside	2 2
97a	25	×	×	×	Xo				
98a	Dell'								
1.30			00	000	Ouest	loc.	EQC	Youden	
	Lab code	e Quest.	ac		1000				00
95a				,	-k	ok	, К	upper right, slightly out	20
96a	2	×	×	×	20	low OD but mean still ok	×	central inside	× C
97a	26	×	×	×	5		8 K	central inside	Y ₁
98a	26	×		×	5				
		-		EOC	Quest.	lac	EQC	Youden	
	Lab code	e Quest.	E C						00
95a	-			,	×	Xo	ok	lower left, outside	Ľ.
96a	9	×	×	<	5				
97a	27			+	+				
98a	8			-					
	Lab code	le Quest.	Iac	EQC	Quest.	lac	EQC	Youden	
95a			-		10		k	upper right, outside	
96a	4	×		×	5		ko	central inside	ΥT T
97a	28	×	-	×	OK				
98a									
200									

According to the second

Attachment III

est	data
lerp	ated d
Rind	ulati
AP	cum
g	Acc

	- Frank				- Chinasa	15			11007
a	Lab code	Quest.	gc	EQC	Quest.	lac	EOC	Youden	Dravininant/ December
							2	10000-	Frovisional/ Recognition
		×		×	8 8		k	central inside	
		×		×	Å		ok N	unner right slightly of feide	00
98a 29		×	×	×	6k	ok	not ok	extremely oustide lower left	
La	Lab code	Quest.	lac	EQC	Quest	lac	EOC	Youdan	
95a								lianno i	
96a 1		×	×	×	o K	6 A	k	upper right slightly out	
		×	×	×	sk	ok	ok	upper right slightly purside	- a
98a 30		×	×	×	ok	ok	6 K	1 inside, 2 slightly low outside	<u>- a</u>
La	Lab code	Quest.	DC	Eac	Quest		LOC I	Velides	
95a						0	L L L	Iondell	
96a									
97a									
98a 31		×	×	×	Å	ok, but need more data	ð	inside	PR
	Lab code	quest.	g	EQC	Quest.	IQC	Eac	Youden	
958									
97a									
98a 32									
96a EQC sample 2 was excluded from evaluation	9 2 was 6	excluded fi	rom evalua	ation					
97a EQC sample 2 was excluded from evaluation	a 2 was e	excluded fi	rom evalua	ation					

Attachment III

თ