22/2 Vol 10 (2466)

INTRODUCTION OF ANTI HTLV3/LAV ("AIDS VIRUS") TESTING IN UK LABORATORIES, 1985

SUMMARY

1. A programme is presented for the introduction during July - October 1985 of anti HTLV3/LAV testing in all Blood Transfusion Service (BTS) Laboratories and in Pubic Health Laboratories where demand requires.

2. A dependable supply of reliable, commercially produced 'solid phase' anti HTLV3/LAV assay kits, preferably from at least two manufacturers and based on different methodologies, is needed to cope with screening and confirmatory tests. Ideally this would be from one UK (Wellcome) and one of the several US manufacturers.

3. During July first evaluations by the PHLS Central Public Health Laboratory (CPHL) Colindale of available commercial kits will be completed. Further evaluation of these and other kits will be carried out at CPHL and in BTS laboratories.

4. During the Autumn, training courses will be run at CPHL to familiarise laboratory workers with the commercial assays available and with the arrangements for confirmation of test results where necessary and for quality control.

5. The PHLS Division of Microbiological Reagents and Quality Control (DMRQC) is preparing sets of anti HTLV3/LAV positive standard sera to establish uniformity in testing, and will regularly issue panels of coded sera to laboratories in order to assess performance. This is essential if tests done in different laboratories are to be comparable.

6. Positive and unexpected negative results will be confirmed by testing at CPHL or in one of the six designated PHLS laboratories. This will also act as a check on the continued quality of commercial assays used in screening tests.

7. The spread of anti HTLV3/LAV infection must be monitored by an effective, confidential reporting system to the Communicable Disease Surveillance Centre, which will provide monthly 'updates' in the Communicable Disease Report. Further discussions between the Director CDSC and BTS directors and others involved, are recommended, so that this reporting system can be comprehensive and duplication avoided.

BOX 3 (5) RAC 1 131

[redacted]

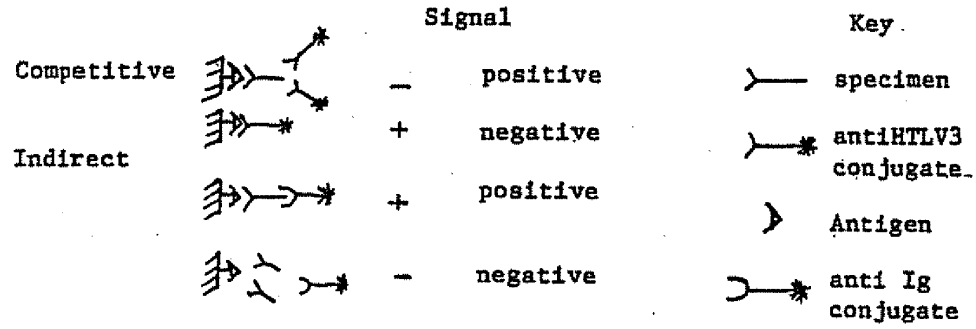
EXPLANATORY NOTES

Kit production/evaluation

At present (June 1985) two UK laboratories, Middlesex Hospital Medical School and PHLS Virus Reference Laboratory, Colindale, have their own solid phase immunoassays. These and four other laboratories, Newcastle, Leeds, Oxford and Birmingham Public Health Laboratories, use immunofluorescence to test for anti HTLV3/LAV.

Commercial assays are being developed by Wellcome (a competitive enzyme assay based on the test described by Cheinsong Popov and Colleagues) and by Abbott, Electronucleonics (ENI), Organon, Litton/Ortho, Travenol and Production Pasteur. (The six last are enzyme assays using the indirect principle).

Figure:



According to manufacturers' representatives, Abbott and ENI are able to supply the British Blood Transfusion and clinical market immediately, though slight delays in delivery of kits for evaluation and the request by both manufacturers to withdraw the initial delivery and send a replacemnt possibly call these claims into question. At the end of May representatives of Wellcome suggested they could supply the British market by September. In our view these verbal undertakings need to be tested further.

The first stage evaluation of commercial assay kits, at CPHL Colindale, has begun. Three companies, Abbott, ENI and Wellcome, have demonstrated their assays. Aliquots of 380 sera have been made into 15 sets. A set will be used to evaluate each commercial product (protocols available). Sets will also be tested by PHLS Virus Reference Laboratory by their radio - immunofluorescence assays, as well as by [redacted], Middlesex Hospital. The DHSS Sub-Group will be convened as required to review the results of these assays.

The second stage evaluation at BTS laboratories at Edgware and Manchester, will be based on 5000 donor specimens aliquoted by [redacted] and colleagues.

TRAINING PROGRAMME

Introductory courses will be held in the Autumn of this year at CPHL, at which assays evaluated, and equipment leased by DHSS for the CPHL evaluation will be demonstrated and assessed. Arrangements for standardisation, kit quality assessment, performance assessment and confirmation of results will be discussed. Manufacturers will be asked to provide explanatory literature etc., but the courses will be private and attendance will be by invitation. Initially representatives of the designated Confirmation Laboratories and some BTS directors and representatives will be invited. Further courses will be arranged for PHLS laboratory and BTS staff.

STANDARDISATION

DMRQC Colindale will prepare two standard sera in amounts required for general UK use:

i) a high titred anti-HTLV3/LAV serum supplied heat activated in 0.25ml amounts

ii) a 'cut-off' serum, probably a 1 in 200 dilution of (i) in anti-HTLV3/LAV negative serum, supplied in 2ml amounts. It is intended that testing laboratories will store small aliquots of this cut-off serum at -30° and use an aliquot in each assay run.

QUALITY CONTROL

Quality control involves :

i) Investigation of variations in reagent supply, presentation and performance. Initially this will be investigated by the evaluation process, but continuing control of this depends on an effective system of test confirmation. A system of bulk buying by PHLS and BTS with preliminary quality control of each new batch may be a further means of safeguarding quality, as well as having cost advantages. It may not be easy to do, however, because of the quite short intervals to expiry date.

ii) Investigation of laboratory performance. This is best achieved by regular distribution of panels of sera with mixed anti HTLV3/LAV reactivity. At first these panels will be 'open' to allow self-assessment by laboratories. Later the sera will be coded and laboratories' performance analysed by DMRQC.

CONFIRMATION

The serious implications of a positive finding for individuals, who may be blood donors, healthy members of "at risk" groups or patients, demand that tests are accurately performed and that all positive and all suspect negative findings are repeated by the same test and confirmed by at least one, preferably two, methodologically different assays. It is estimated that 10,000 - 20,000 sera will require confirmatory tests during 1986. While most results will be readily confirmed, a few sera will need further analysis and arrangements may have to be made to test these by Western blotting and/or radio immunoprecipitation, or both.

REPORTING ANTI HTLV3/LAV RESULTS

It is hoped that all positive results will be reported to PHLS Communicable Disease Surveillance Centre, but arrangements need to be made to identify and distinguish provisional from confirmed positive findings and to avoid duplication. The Blood Transfusion Service has yet to consider its attitude to the reporting of positive findings. Physicians caring for the "at risk" groups, homosexuals, haemophiliacs and drug abusers, have already expressed misgivings about monthly publication in the Communicable Disease Report (CDR) of analyses of positive results and about confidentiality, and Blood Transfusion Directors may also have reservations. Nevertheless, these regular analyses will be a very useful measure of the spread of infection. It is suggested that further discussions of the mechanics and acceptability of this reporting system are needed between CDSC and the main groups involved.

[redacted]
June 1985

PHLS Colindale

DMRQC - PANEL I. ANTI-HTLVIII/LAV TESTS

tests performed at PHLS Virus Reference Laboratory, May/June '85

RIA

[redacted] - COLINDALE

BLOCKING TEST

PANEL NO.	PERCENTAGE INHIBITION	RESULT
1	−16	−
2	90	+
3	92	+
[redacted]	91	+
5	4	−
6	92	+
7	78	+
8	2	−
9	86	+
10	2	−
11	86	+
12	58	?

[redacted] Inhibition − positive

50–70% Inhibition − equivocal

<50% Inhibition − negative

ELISA

[redacted]/MIDDX

BLOCKING TEST

SUBSTRATE - OPD

PANEL No.	Absorbance 492nm	RESULT
1	1·30	−
2	0·06	+
3	0·05	+
4	0·07	+
5	1·13	−
6	0·08	+
7	0·07	+
8	1·16	−
9	0·06	+
10	1·30	−
11	0·10	+
12	0·30	+

high +ve Abs = 0·07

negative Abs = 1·50

cut off Abs = 0·84

values <0·84 are positive

[redacted] HTLV III ELISA

INDIRECT TEST

SUBSTRATE - OPD

PANEL No.	Absorbance 492nm	RESULT
1	0·01	−
2	1·36	+
3	0·30	+
4	1·00	+
5	0·01	−
6	0·70	+
7	0·68	+
8	0·02	−
9	1·11	+
10	0·02	−
11	0·85	+
12	0·11	?

high +ve Abs = 0·951

low +ve Abs = 0·128

negative Abs = 0·019

[redacted] HTLVIII Bio-EnzaBead

INDIRECT TEST

SUBSTRATE - ABTS

PANEL No.	Absorbance 690nm	RESULT
1	0·070	−
2	0·455	+
3	0·279	+
4	0·534	+
5	0·097	−
6	0·355	+
7	0·499	+
8	0·110	−
9	0·403	+
10	0·102	−
11	0·561	+
12	0·225	+

positive Abs = 0·360

negative Abs = 0·077

cut off Abs = 0·187

(neg mean + 0·11)

[redacted] HTLVIII GIA

INDIRECT TEST

SUBSTRATE - OPD

No	Absorbance 492	RESULT
1	0·038	−
2	1·593	+
3	0·830	+
4	1·645	+
5	0·040	−
6	1·601	+
7	1·535	+
8	0·050	−
9	>2·0	+
10	0·052	−
11	1·789	+
12	0·28	+

positive mean 0·520

negative mean 0·018

cut off 0·070

(neg mean + 1/10 pos mean)

JUNE 1985

[redacted]

BOX 3 (S) R+C/135

[redacted]

DMRQC - PANEL I. ANTI-HTLVIII/LAV TESTS

tests performed at PHLS Virus Reference Laboratory, May June '85

RIA

[redacted] - COLINDALE

BLOCKING TEST

PANEL No.	PERCENTAGE INHIBITION	RESULT
1	−16	−
2	90	+
3	82	+
4	91	+
5	4	−
6	92	+
7	78	+
8	2	−
9	86	+
10	2	−
11	86	+
12	58	?

>70% Inhibition − positive

50-70% Inhibition − equivocal

<50% Inhibition − negative

ELISA

[redacted] E/MIDDX

BLOCKING TEST

SUBSTRATE - OPD

PANEL No.	Absorbance 492nm	RESULT
1	1·30	−
2	0·06	+
3	0·05	+
4	0·07	+
5	1·13	−
6	0·08	+
7	0·07	+
8	1·16	−
9	0·06	+
10	1·30	−
11	0·10	+
12	0·30	+

high $+^{ve}$ Abs = 0·07

negative Abs = 1·50

cut off Abs = 0·84

values <0·84 are positive

[redacted] HTLVIII ELISA

INDIRECT TEST

SUBSTRATE - OPD

PANEL No.	Absorbance 492nm	RESULT
1	0·01	−
2	1·36	+
3	0·30	+
4	1·00	+
5	0·01	−
6	0·70	+
7	0·68	+
8	0·02	−
9	1·11	+
10	0·02	−
11	0·85	+
12	0·11	?

high $+^{ve}$ Abs = 0·851

low $+^{ve}$ Abs = 0·128

negative Abs = 0·019

[redacted] O/LITTON

HTLVIII Bio-EnzaBead

INDIRECT TEST

SUBSTRATE - ABTS

PANEL No.	Absorbance 690nm	RESULT
1	0·070	−
2	0·455	+
3	0·279	+
4	0·534	+
5	0·087	−
6	0·355	+
7	0·499	+
8	0·110	−
9	0·400	+
10	0·102	−
11	0·561	+
12	0·225	+

positive Abs = 0·360

negative Abs = 0·077

cut off Abs = 0·187

(neg mean + 0·11)

[redacted]

HTLVIII EIA

INDIRECT TEST

SUBSTRATE - OP

No	Absorbance 492nm	RESUL
1	0·038	−
2	1·593	+
3	0·830	+
4	1·645	+
5	0·040	−
6	1·601	+
7	1·535	+
8	0·050	−
9	>2·0	+
10	0·052	−
11	1·789	+
12	0·28	+

positive mean = C

negative mean = 0

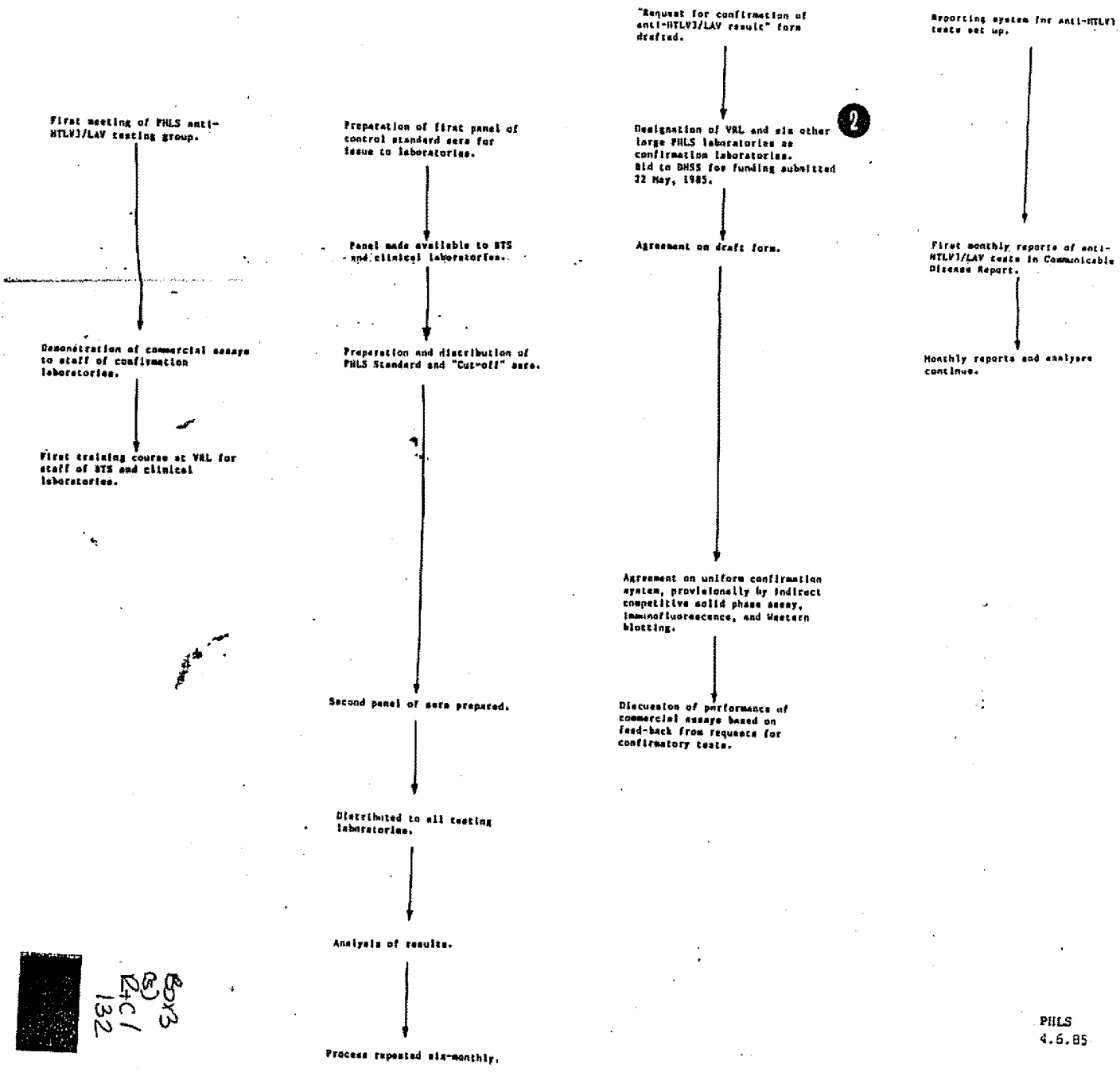
cut off 0·070

(neg mean + ½ pos me

JUNE 1985

[redacted]

(RUS") TESTING IN ENGLAND & WALES (SITUATION AT JUNE 1985)



[redacted]

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PHLS
4.6.85

FLOW CHART FOR THE INTRODUCTION OF ANTI-HTLV3/LAV

PRODUCTION, EVALUATION & INTRODUCTION OF SCREENING TESTS

SUMMER '84

"In house" anti-HTLV3/LAV assays developed in a few foreign and UK research laboratories. PHLS Virus Reference Laboratory (VRL) and the Middlesex Hospital available as primary testing centres.

WINTER '84/85

Antigen producer cell lines made available by US National Cancer Institute to five US companies.

CEM-J producer cell line developed at Chester-Beatty Institute, London.

APRIL '85

Licensing of first US commercial assays by FDA (Abbott, Electronucleonics, ENI).

CEM-J line made available to Wellcome UK.

MAY '85

Licensing of other commercial assays by US FDA (Litton, Travenol).

Demonstration of Abbott, ENI and Wellcome assays to PHLS staff and DHSS officers.

JUNE '85

Demonstration of further assays to PHLS and DHSS.

PHLS evaluation of first three assays available (Abbott, ENI and Wellcome). 1

BTS

Visit to PHLS by BTS officer who is organising BTS evaluation, 12 June 1985.

JULY '85

Presentation of PHLS findings to DHSS Subgroup.

AUGUST '85

PHLS evaluation of Litton and Travenol assays, if available.

BTS evaluation of suitable assays begins. 3

SEPTEMBER '85

Presentation of PHLS findings to DHSS Subgroup.

Introduction of anti-HTLV3/LAV assays in clinical laboratories designated as confirmation centres. 2

BTS evaluation continues. 3

OCTOBER '85

Introduction of anti-HTLV3/LAV screening tests in PHLS clinical laboratories. 2

Introduction of anti-HTLV3/LAV screening in BTS.

NOVEMBER '85

DECEMBER '85

1986

1 First batches delivered by manufacturers were withdrawn (on 23 May by ENI and 29 May by Abbott). Replacements expected 5 June (Abbott) and at a date to be notified by ENI, who have no agent in the UK. Wellcome kits expected on 21 June.

2 The "Confirmation Laboratories" other than VRL all require upgrading to meet current ACDP safety guidelines, and will need some new equipment. A bid for the funding needed was submitted to DHSS on 22 May 1985.

3 The two intermediate steps introduce a degree of