DIVISION OF VIRAL PRODUCTS
NIBSC

To: Dr. D.P. THOMAS, DIVISION OF BLOOD PRODUCTS

TESTS FOR HTLV-III/LAV ANTIBODIES

TEST USED: Wellcozyme (competitive ELISA)

Manufacturer	Material	Batch No.	Date Rec'd.	Date Tested	Result
SNBTS, Edinburgh	Factor II IX + X DEFIX				
	IX	3511-60170	26/5/87	28-5-87	—
	IX	3502-7020[illegible]	26/5/87		—

Comments:

Signed..... GRO-C .....

Date.....28th May 1987.....

DT3AHW

MEDICINES ACT 1968

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EXAMINATION OF SAMPLES AND PROTOCOLS

Product.... FACTOR VIII.... Licence Holder's Name..... BPL.....

Licence No..... Address..... ELSTREE.....

Date protocol received ..... 3-6-86.....

Batch No. ..... 8Y-2264..... Date samples received ..... 3-6-86.....

Tests carried out	Results (Mark 'not applicable' if the test is inappropriate to the product)
Identity	
Abnormal toxicity	
Potency	244 vs LABEL 225 (108%)
Pyrogens	
Sterility	
Special tests (please detail below)	
ANTI-HIV	-ve
Protocol	

REMARKS

ASSAYED WITH 8Y-2239

Date released.....

DIVISION OF VIRAL PRODUCTS

NATIONAL INSTITUTE FOR BIOLOGICAL STANDARDS

AND CONTROL

To ..... Dr. D. P. THOMAS .....

Division/Section ..BLOOD PRODUCTS...

TESTS FOR HTLV-III/LAV ANTIBODIES

Test Technique(s) used..WELLCOZYME (Competitive EIA)

Manufacturer	Material	Code No	Date Received	Date Tested	Result
ARMOUR	Factor VIII	B52702 B52902 B53002	30.5.86	5.6.86	— — —
BLOOD PRODUCTS LABORATORY (CRISTAL)	Factor VIII Factor IX	8Y3386 8Y3387 9A3340 9A3357	3.6.86	5.6.86	— — — —
PFL (Oxford)	Factor VIII Factor IX	8Y2264 9A2217	3.6.86	5.6.86	— —

Signed ... GRO-C .....

Date ..... 6th June 1986 .....

COAGULATION RESULTS SHEET

Test material: factor VIII

Test sample: Elstree BPL

Assay method(s): 2 stage

Batch No.: Various

Standard: 85/650

Recon.Vol.(ml): 10

Standard potency (iu/ml): 3.8

Nominal potency (u/ml): various

Recon.Time (min): ~10min

Comments: most clear

Vial No.	Operator	Assay No.	Potency iu/ml	95% limits	Weight	Parallel
8Y 2239	GKC	17.9.1	28.6	26.1-31.5	6122	1.52
8Y 2264	GKC	17.9.1	24.4	22.3-26.8	6164	"
8Y 3386	SJE	17.9.2	20.6	17.4-24.4	1925	1.47
8Y 3400	GKC	17.9.3	22.4	18.7-26.9	1672	0.53
8Y 3411	GKC	17.9.3	22.0	18.4-26.4	1672	"
8Y 2279	SJE	17.9.4	29.2	25.0-34.3	2192	1.07
8Y 3419	SJE	17.9.4	21.7	18.6-25.3	2251	"

Combined potency/ml =

Date of completion 17.9.86.

COMBINED RESULTS

	Potency iu/vial	95% limits	Weight	Homogeneous
NIBSC				Yes/No
Manufacturer				
NIBSC Man. %				
GENABD				

NIBSC ARRIVED 3-6-86

QUALITY CONTROL SUMMARY-PFL 8Y QC-02(a)

File reference : QC8Y0202.PFL
Date effective : 10.12.85
Authorized by : GRO-C

Product : DRIED HIGH PURITY FACTOR VIII
Batch No. : 8Y 2264

1. Results for these tests must be within limits for labelling authorization.

Test	Limit Ref.	Limit	This Batch	Expect*
Sterility	Ph.Eur.'80	Pass	Pass	Pass
Pyrogenicity, °C/n	Ph.Eur.'80	Pass	Pass 0/3	Pass
Abnormal Toxicity	Ph.Eur.'80	Pass	Pass	Pass
Hepatitis Bs Antigen	BPL (RIA)	Pass	Pass	Pass
HTLV III Antibody	ELISA	Pass	Organon-Pass.	Pass
Solubility 20°C, min	BP '80	≯20	7	1-10
Appearance of solution	BP '80	Complies	"Usual hits" complies.	Complies
Stability 20°C, h	BPL	≮3	Pass.	>3
pH at 20°C	BP '80	6.8-7.4	6.87	6.31-6.97
Factor VIII potency, iu/ml	BP '80/82	≮6.0	22.6	13.1-40.6
Specific activity, iu/mg	BP '80/82	≮0.2	3.21	2.05-7.97
Total protein, g/l	BPL	≯25	7.04	2.66-7.94
Clottable protein, g/l	BPL	≯20	5.25 (75%)	1.45-6.26
Sodium ion, mmol/l	BP '80	≯200	124	118.1-129.3
Potassium ion, mmol/l	BPL	≯5	<0.05	<0.05
Chloride ion, mmol/l	BPL	≯200	109	95.9-111.9
Citrate ion, mmol/l	BP '80	≯55	9.79	8.76-10.64
No.of donations used	BPL	≯10000	623	<10000
Heparin, u/ml	BPL	≯2	<0.2	<0.2(n=10)
Sucrose, g/l	BPL	≯20	—	-
Tris, mmol/l	BPL	≯60	10.6	7.96-11.92
Glycine, mmol/l uncorrected	BPL	≯50	9.78	6.19-12.11

This summary sheet completed by : Initials/date GRO-C 6/5/86

* Based on 14 batches (8Y2212-8Y2231) assuming log distribution for F.VIII and specific activity

Moisture Content: (untreated) 1.04 % w/w HS.
" " (HTS) 0.947 " HT

QUALITY CONTROL SUMMARY-PFL 8Y QC-02(b)

File reference : QC8Y0202.PFL
Date effective : 10.12.85
Authorized by : GRO-C

Product : DRIED HIGH PURITY FACTOR VIII
Batch No. : 8Y 2264 (part 2)

2. Results of these tests will be available.

PKA, units/ml	-	-	<5	<5
Conductivity, $mScm^{-1}$ @ $20^{o}C$	-	-	11.1	8.2-12.2
Factor VIII, iu/vial	-	Not assigned	225	112.2-367
Calcium, ions µM	-	-	37	51.8-93.4
F.VIII R:Ag, u/ml	-	-		27.1-79.4
Anti A,Oxford units/ml	-	-	41	41.7-114.1

This summary sheet completed by : Initials/date no F.VIII R:Ag. GRO-C 6/5/86.

* Based on 14 batches (8Y2212-8Y2231) assuming log distribution for F.VIII and specific activity

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EXAMINATION OF SAMPLES AND PROTOCOLS

Product... FACTOR VIII ..... Licence Holder's Name... BPL .....

Licence No. ..... Address... ELSTREE .....

Date protocol received ... 4-2-86 .....

Batch No. ... 87-2244 ... Date samples received ... 4-2-86 .....

Tests carried out	Results (Mark 'not applicable' if the test is inappropriate to the product)
Identity	
Abnormal toxicity	
Potency	252 vs LABEL 285 (88%)
Pyrogens	
Sterility	
Special tests (please detail below)	
ANTI-HIV	-ve
Protocol	

REMARKS

RESULTS WITH 87-2233

Date released.....

DIVISION OF VIRAL PRODUCTS

NATIONAL INSTITUTE FOR BIOLOGICAL STANDARDS

AND CONTROL

To .. Dr. D. P. THOMAS .....

Division/Section .BLOOD ... PRODUCTS ..

TESTS FOR HTLV-III/LAV ANTIBODIES

Test Technique(s) used.. WELLCOZYME (Competitive ELISA)

Manufacturer	Material *	Code No	Date Received	Date Tested	Result
Alpha	Profilate	A6.1210	6.2.86	11.2.86	—
Armour	Factorate	A.44411	3.2.86	11.2.86	—
		A.45211			—
		A.46311			—
Blood Products Lab. (Elstree)	Factor VIII	84.2239	4.2.86	11.2.86	—
		2244			—
		2250			—
		2257			—
		3342			—
		3356			—
	Factor IX	84 3331	4.2.86	11.2.86	—

* NB - all reconstituted in ½ volume of sterile water the recommended for reconstitution

Signed .. GRO-C .....

cc Dr. J. Bannerjee Date ..... 13th February 1986.

ARRIVED 4-2-86

COAGULATION RESULTS SHEET

Test material: Factor VIII

Test sample: ELSTREE

Assay method(s): 2-stage

Batch No.: 84 2244 (T_2)

Standard: 85/650

Recon.Vol.(ml): 10.0

Standard potency (iu/ml): 3.8

Nominal potency (u/ml): 28.5

Recon.Time (min):

Comments:

Vial No.	Operator	Assay No.	Potency iu/ml	95% limits	Weight	Parallel
1	SJE	29.8.4	25.2	19.9 - 31.7	1020.78	0.90

Combined potency/ml = 25.2

Date of completion 29.8.86

COMBINED RESULTS

	Potency iu/vial	95% limits	Weight	Homogeneous
NIBSC	252	199 - 317	1020.78	Yes/No
Manufacturer	285			
NIBSC / Man. %	88	70 — 111		
GENABD				

File reference : QC8Y0101.REP Product : DRIED HIGH PURITY FACTOR VIII

Date effective : 12th March 1985

Authorized by : GRO-C Batch No. : 8Y 2244

Label : (completed sample label, exactly as attached to vials)

The reconstituted solution contains, per litre, not more than:- 25g protein (including not more than 20g fibrinogen), 60 mmol Tris, 200 mmol sodium, 200 mmol chloride, 55 mmol citrate, 50 mmol glycine, 20 g sucrose and 2000 units heparin.

STORE IN THE DARK, BELOW +6°C

BLOOD PRODUCTS LABORATORY
Dagger Lane, Elstree, Herts WD6 3BX

DRIED FACTOR VIII FRACTION

High Purity: Heat Treated

Directions for use: before reconstituting, warm the vial and Water for Injections to 20°C to 30°C. Add the indicated volume of water and mix gently, avoiding frothing, before releasing vacuum. Use immediately; discard if not used within 3 hours of reconstitution, or if a gel forms.

Warning: the preparation is of human origin (less than 10,000 plasma donations used per batch). It has been subjected to heat treatment, in the vial, to reduce the risk of infection by viral agents (including hepatitis and AIDS viruses) but cannot be assumed to be free from risk of infection.

Batch information:

Factor VIII content (iu) 285

Dissolve in (ml) 10

Batch 8Y 2244

Expiry OCT 86

8Y1

Date of expiry taken as one year from date of assigning potency (16.10.85)

Formulation : See BP1980 "Dried Factor VIII Fraction" and 1982 Addendum. See also label side-panels and report QC02.

Product Licence No.:

Process Summary :

Related batches from same pool	8Y 1921	8Y 1922
Plasma source (RTC)	G	T
Number of donations	445	520
Fractionation date	22.8.85	28.8.85

Fill (pre-dry) volume 10 ml. Recommended reconstitution volume 10 ml.

Date of terminal filtration 11.9.85. Defined batch 307 vials

Date of dry heat treatment 7/10/85 and conditions HT3

Number of vials considered for release 275. Initials/date 5/12/85

Water for Injections Manufacturer and Batch No Fisons 520 008

Release authorization : I have examined the Batch Manufacturing Record and I authorize the issue of 271 vials for clinical use ~~without restriction~~/ subject to the following restrictions : in patients to be named.

Signed : GRO-C Head of Quality Control

Date : 29.1.86 BPL/PFL.

Inventory : 271 vials placed at issue Initials/date:

2 reference samples removed Initials/date: GRO-C 26.11.85

2 vials removed for N.I.B.S.C. Initials/date: GRO-C 26.11.85

File reference : QC8Y0201.PFL
Date effective : GRO-C
Authorized by : 28.10.85

Product : DRIED HIGH PURITY FACTOR VIII
Batch No. : 8Y 2244

1. Results for these tests must be within limits for labelling authorization.

Test	Limit Ref.	Limit	This Batch	Expect*
Sterility	Ph.Eur.'80	Pass	Pass	Pass
Pyrogenicity, °C/n	Ph.Eur.'80	Pass	Pass	Pass
Abnormal Toxicity	Ph.Eur.'80	Pass	Pass	Pass
Hepatitis Bs Antigen	BPL (RIA)	Pass	Pass	Pass
Solubility 20°C, min	BP '80	⩾20	10	1-10
Appearance of solution	BP '80	Complies	See QC 15.2.16	Complies
Stability 20°C, h	BPL	⩽3	Pass	>3
pH at 20°C	BP '80	6.8-7.4	6.87	6.81-6.97
Fctor VIII potency, iu/ml	BP '80/82	⩽6.0	28.4	13.1-40.6
Specific activity, iu/mg	BP '80/82	⩽0.2	4.83	2.05-7.97
Total protein, g/l	BPL	⩾25	5.88	2.66-7.94
Clottable protein, g/l	BPL	⩾20	77%	1.45-6.26
Sodium ion, mmol/l	BP '80	⩾200	125	118.1-129.3
Potassium ion, mmol/l	BPL	⩾5	<0.05	<0.05
Chloride ion, mmol/l	BPL	⩾200	105	95.9-111.9
Citrate ion, mmol/l	BP '80	⩾55	10.0	8.76-10.64
No.of donations used	BPL	⩾10000	465	<10000
Heparin, u/ml	BPL	⩾2	<0.2	<0.2(n=10)
Sucrose, g/l	BPL	⩾20	—	-
Tris, mmol/l	BPL	⩾60	106	7.96-11.92
Glycine, mmol/l uncorrected	BPL	⩾50	10.0	6.19-12.11

2. Results of these tests will be available.

PKA, units/ml	-	-	<5	<5
Conductivity, $mScm^{-1}$ @ 20°C	-	-	11.0	8.2-12.2
Factor VIII, iu/vial	-	Not assigned	285	112.2 - 367
Calcium, ionsμM	-	-	32	51.8-93.4
F.VIII R:Ag, u/ml	-	-	50.2	27.1-79.4
Anti A,Oxford units/ml	-	-	100	41.7-114.1

This summary sheet completed by : Initials/date GRO-C 11/11/85.

* Based on 14 batches (8Y2212-8Y2231) assuming log distribution for F.VIII and specific activity

PCT HIV -ve

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EXAMINATION OF SAMPLES AND PROTOCOLS

Product FACTOR VIII

Licence Holder's Name BPL

Licence No. ———

Address ELSTREE

Date protocol received ~~25-9-86~~ 22.10. GRO-C [PG]

Batch No. 8Y-3428

Date samples received 25-9-86

Tests carried out	Results (Mark 'not applicable' if the test is inappropriate to the product)
Identity	
Abnormal toxicity	
Potency	173 vs LABGL 181 (95.6%)
Pyrogens	
Sterility	
Special tests (please detail below)	
ANTI-HIV	-ve
Protocol	

REMARKS

~~ACC PROTOCOL~~

NO LABELLED POT.

Date released.....

QUALITY CONTROL SUMMARY — NIBSC — 8Y QC-02(a)

File reference : QC8Y0209.REP — Product : DRIED HIGH PURITY FACTOR VIII

Date effective : 7.8.86
Authorized by : GRO-C — Batch No. : 8Y 3428

1. Results for these tests must be within limits for labelling authorization.

Test	Limit Ref.	Limit	This Batch	Expect*
Sterility	Ph.Eur.'80	Pass	PASS	Pass
Pyrogenicity, °C/n	Ph.Eur.'80	Pass	0.0 (3)	Pass
Abnormal Toxicity	Ph.Eur.'80	Pass	PASS	Pass
Hepatitis Bs Antigen	BPL (RIA)	Pass	PASS	Pass
Anti-HIV	ELISA	Pass	PASS	Pass
Solubility 20°C, min	BP '80	⩽20	2½	1-16
Appearance of solution	BP '80	Complies	Complies	Complies
Stability 20°C, h	BPL	⩾1	PASS	>1
pH at 20°C	BP '80	6.8-7.4	6.95	6.82-7.02
Factor VIII potency, iu/ml	BP '80/82	⩾6.0	18.1	18.9-27.2
Specific activity, iu/mg	BP '80/82	⩾0.2	4.56	2.01-4.73
Total protein, g/l	BPL	⩽25	3.97	4.5-9.6
Clottable protein, g/l	BPL	⩽20	2.65 (67%)	3.4-8.2
Sodium ion, mmol/l	BP '80	⩽200	123	108-138
Potassium ion, mmol/l	BPL	⩽5	<0.05	<0.05
Chloride ion, mmol/l	BPL	⩽200	98.3	93-123
Citrate ion, mmol/l	BP '80	⩽55	9.16	7.3-11.8
No.of donations used	BPL	⩽25000	5704	<25000
Heparin, u/ml	BPL	⩽2	<0.2	<0.2
Sucrose, g/l	BPL	⩽20	-	-
Tris, mmol/l	BPL	⩽60	9.6	8.1-13.8
Glycine, mmol/l(Uncorrected)	BPL	⩽50	11.7	7.1-13.14

This summary sheet completed by : Initials/date GRO-C [DS] 21/10/86

* Based on 56 batches (8Y 3336 - 8Y 3394) Mean ± 2 S.D

DIVISION OF VIRAL PRODUCTS

NATIONAL INSTITUTE FOR BIOLOGICAL STANDARDS

AND CONTROL

To ..... Dr. D. P. Thomas .....

Division/~~Section~~ . Blood ... Products ..

TESTS FOR HTLV-III/LAV ANTIBODIES

Test Technique(s) used.... Wellcozyme (competitive ELISA)

Manufacturer	Material	Code No	Date Received	Date Tested	Result
BPL (ESTREE)	Factor VIII	84 3427	22.9.86	30.9.86	– *
		84 3428			– *
		84 3429			– *
	Factor IX	9A 3366	22.9.86	30.9.86	–
	Fibrinogen	FC 805	22.9.86	30.9.86	–

* abnormally high OD - (absorption of conjugate ?)

Signed ... GRO-C .....

Date ..... 1st October 1986 .....

cc Dr J.W. Barnes Cliffe

t

COAGULATION RESULTS SHEET

Test material: Factor VIII

Test sample: Elstree FVIII

Assay method(s): 2-stage

Batch No. 8Y 3426 / 3427 / 3428 / 3429

Standard: 85/650

Recon.Vol.(ml): 10 / ? / ? / ?

Standard potency (iu/ml): 3.8

Nominal potency (u/ml): 24.6 / ? / ? / ?

Recon.Time (min):

Comments:

Batch No.	Operator	Assay No.	Potency iu/ml	95% limits	Weight	Parallel
8Y 3426	CAJ	23.10.7	23.2	18.4 - 29.2	1029.15	0.09
8Y 3427	CAJ	23.10.7	22.3	17.7 - 28.1	1030.5	0.09
8Y 3428	SJE	23.10.8	17.3	8.6 - 31.4	155.4	0.43
8Y 3429	SJE	23.10.8	19.5	10.0 - 35.9	159.1	0.43

Combined potency/ml =

Date of completion

COMBINED RESULTS

	Potency iu/vial	95% limits	Weight	Homogeneous
NIBSC				Yes/No
Manufacturer				
NIBSC Man. & GENARD				

COAGULATION RESULTS

Assay: FVIII 2 st

Assay No.: 23.10.8

Operator: STE

Date: 23.10.86

Standard: 85/650

Test: T_1 Elstree 84 3428
T_2 " 84 3429

Potency (iu/ml): 3.8

Potency (iu/ml): ? ?

Initial dilutions: 1/12

Initial dilutions: 1/70 1/70

Preparation	Dilution	Clotting times 1	2	Ave.	Comment
S	x30	15.6	16.6	16.1	
	x60	19.4	19.8	19.6	
	x120	24.4	25.0	24.7	
T_1		19.0	17.6	18.3	
		~~25.4~~	21.4	21.4	
		34.4	32.0	33.2	
T_2		16.8	16.0	16.4	
		21.4	20.2	20.8	
		31.8	33.8	32.8	
S		18.0	16.0	17.0	
		22.6	20.0	21.6	
		~~36.0~~	31.4	31.4	

Substrate plasma:

Reagent: Home made/combined

Subsampling times (min.):

ASSAY TITLE 23.19.3
PREPARATIONS 1 STANDARD + 2 UNKNOWN(S)
FIDUCIAL INTERVAL 95%
TRANSFS & CONSTANTS LOG 0.000000 0.000000

STANDARD S50

Dose	Responses	
22.20000	24.70000	31.40000
44.40000	19.60000	21.20000
88.80000	16.10000	17.00000

UNKNOWN T1

Dose	Responses
1.00000	33.20000
2.00000	21.40000
4.00000	16.30000

UNKNOWN T2

Dose	Responses
1.00000	32.60000
2.00000	20.00000
4.00000	16.40000

TREATMENT GROUPS

Prep Name	Dose	Mean Response	No of Resp	Variance
s50	22.20000	1.44481	2	0.005432
s50	44.40000	1.31335	2	0.000890
s50	88.80000	1.21864	2	0.000279
Mean Log Dose =	1.64738			
Mean Response =	1.32560			
T1	1.00000	1.52114	1	0.000000
T1	2.00000	1.33041	1	0.000000
T1	4.00000	1.26245	1	0.000000
mean Log Dose =	0.30103			
mean Response =	1.37133			
T2	1.00000	1.51587	1	0.000000
T2	2.00000	1.31006	1	0.000000
T2	4.00000	1.21484	1	0.000000
mean Log Dose =	0.30103			
mean Response =	1.34959			

ANALYSIS OF VARIANCE

ANALYSIS OF VARIANCE

Source of Variation	d.f.	Sum Squares	Mean Square	F-Ratio	
Between Treatments	8	0.13873	0.01734	7.88	
Preparations	2	0.00436	0.00218	0.99	
Regression	1	0.12804	0.12804	58.18	**
Parallelism	2	0.00189	0.00094	0.43	
Linearity (S50)	1	0.00045	0.00045	0.20	
Linearity (T1)	1	0.00251	0.00251	1.14	
Linearity (T2)	1	0.00149	0.00149	0.68	
Residual	3	0.00660	0.00220		
Total	11	0.14533			

* indicates significance at 5% level
** indicates significance at 1% level
*** indicates significance at 0.1% level

SLOPES, INTERCEPTS etc

Prep	Slope	Y-intercept	$\bar{X}S - \bar{X}U$	$\bar{Y}U - \bar{Y}S$	V($\bar{Y}U$-$\bar{Y}S$)
S50	-0.37567	1.94447			
T1	-0.42767	1.90068	1.34633	0.04573	0.00110
T2	-0.50000	1.80011	1.34633	0.02399	0.00110

Common Slope (b) =	-0.42025	Standard Dev (s) =	0.04671
s/o =	-0.11162	Variance of b =	0.00304
Finney's g =	0.17407	Standard Error of b =	0.05509

POTENCY TABLE

Prep		Potency	Fiducial Limits		Weight (1/V(M))
T1	Log	1.23753	0.93278	1.49641	155.44
		17.27952	8.56612	31.30233	
T2	Log	1.28726	0.99933	1.55513	159.09
		19.46543	9.98465	35.98301	