Reference A
Paper 2 of 2

INVESTIGATION CONCERNING EVENTS SURROUNDING THE INTRODUCTION OF HEAT TREATMENT FOR BLOOD PRODUCTS IN THE MID 1980's

ADDITIONAL INFORMATION REQUESTED BY THE SCOTTISH EXECUTIVE

Scottish National Blood Transfusion Service
Edinburgh

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1. INTRODUCTION

In December 1999, the Scottish National Blood Transfusion Service (SNBTS) submitted a report to the Scottish Executive describing the development of hepatitis-safe Factor VIII concentrate by SNBTS. Subsequently, additional information was requested by the Scottish Executive (letter from C Dora to F Gibb, 14 February 2000). Our response to this request is provided below, with each item being dealt with in the order listed by the Scottish Executive. Reference numbers for cited literature are those given in the original report. Copies of references not cited previously are appended.

2. QUESTIONS CONCERNING THE SNBTS SUBMISSION OF DECEMBER 1999

2.1 Question

"Paragraphs 2.6 and 5.1 *Can more information be provided on the "purification" of FVIII (ie. in lay terms, what is being removed in order to "purify" the product?) When did SNBTS make the findings about the behaviour of NY under heat treatment?"*

Response

FVIII is a trace component of human blood plasma, accounting for less than 0.001% by weight of the protein present. The objectives of the fractionation process are to separate this material from plasma and provide it to the patient in a concentrated dose form which is stable, convenient and as safe as possible.

In the preparation of FVIII concentrate, FVIII is separated from most of the plasma proteins by being concentrated into insoluble (solid) material which remains when frozen plasma is thawed at low temperatures. This solid material or fraction, known as cryoprecipitate, can be redissolved to form an impure solution of FVIII which is composed mainly of Fibrinogen and Fibronectin, proteins which tend to co-purify with FVIII.

Fibrinogen and Fibronectin are poorly soluble, adherent proteins which prevent cryoprecipitate from being filtered to remove bacterial contaminants and solutions of FVIII from being concentrated into a convenient dose size.

In addition, some liquid plasma is inevitably carried over with the cryoprecipitate and certain other proteins (enzymes) present in this material can degrade FVIII leading to product instability.

The first generation of FVIII concentrates resolved these problems by further purifying the redissolved cryoprecipitate to specifically remove both the least soluble protein and the damaging enzymes.

In the 1970's, the purification methods available for this purpose also resulted in significant loss of FVIII. For those manufacturers aiming to

achieve self-sufficiency, yield was considered most important; whilst some commercial companies chose to increase purity at the expense of yield in order to provide a greater product solubility, the convenience of which was attractive for marketing purposes.

The purity of different products is characterised by their "specific activity"; that is, the FVIII activity (in International Units) divided by the total protein content (in milligrams of protein). Some examples are given below for first generation concentrates, together with the degree of dry heat treatment that each product was able to withstand before becoming insoluble and unsuitable for use.

PRODUCT	FVIII SPECIFIC ACTIVITY (IU/mg)	DRY HEAT TOLERATED (°C, hours)
Plasma	0.02	
Cryoprecipitate	0.2	
FVIII Concentrates:		
SNBTS, NY	0.4	68, 2h 60, 24h
Alpha, Profilate	0.5	60, 24h
Armour, Factorate	0.5 - 1.0	60, 36h
Cutter, Koate	1.0	68, 72h
Baxter, Hemofil	1.5	60, 72h

During 1983 we learned that Baxter and Armour were both investigating dry heat treatment of FVIII at 60°C. Preliminary studies on dry heat treatment of NY were carried out by SNBTS in November 1983, indicating that NY could tolerate heating to a similar degree before becoming insoluble, but the degree of virus inactivation measured at the same time was lower than we had obtained in our studies of pasteurisation (heating in a liquid state).

By the Autumn of 1984 we were aware that the causative agent of AIDS had been identified and that its sensitivity to heat was being investigated in the USA. In order to better define the options available, should HIV be found to be sensitive to dry heat treatment, we decided to make further measurements on the behaviour of NY under heat treatment. These measurements were completed in October 1984.

The first indication that HIV might be inactivated by dry heat treatment of FVIII at 68°C was published in the USA on 26 October (Morbidity Mortality Weekly Report, 33, 589-591,1984) with more detailed information being presented on 2 November 1984 at a conference in the Netherlands at which SNBTS staff were present. A programme to implement dry heat treatment of NY at 68°C for 2 hours was initiated immediately by SNBTS.

As a result of research undertaken during November/December 1984 we discovered that heating of NY at 68°C could be extended from 2 hours to 24 hours by the addition of carbohydrate to the final product formulation. This change to the manufacture of NY was implemented in January 1985. Research continued on dry heat treatment of NY during 1985 in an attempt to further extend these heating conditions, but no further changes were identified.

2.2 **Question**

"Paragraph 2.12 *When was the new equipment designed and constructed"*

Response

New equipment was designed and constructed in order to thaw plasma and recover cryoprecipitate in a more rapid and controlled manner than the established technology in the belief that this would reduce loss and degradation of FVIII at this step. This concept and how it might be achieved were explained by SNBTS in September 1978 (Lancet 2, 574, 1978).

Two items of equipment were designed, the first being a prototype (pilot) unit that was constructed for evaluation in production and the second being the finalised unit, the design of which was based on information gained from the operation of the prototype.

Design and construction of the prototype unit were undertaken in the period August - December 1978. Following commissioning trials in early 1979, the unit was introduced into production in March 1979 and operated in parallel with the established equipment in order to compare the performance of the different systems. By June 1979 sufficient data were available to demonstrate a marked increase in FVIII yield with the new equipment and the older procedure was discontinued.

The prototype was used to evaluate a number of aspects of the process and to provide the information required to specify and construct a definitive and larger unit in anticipation of increased volumes of plasma becoming available. The design and construction of this second unit were completed in the latter half of 1980 with the larger unit replacing the prototype in January 1981.

Each of these units performed in a similar manner, both providing a 40% increase in FVIII yield, as well as increased specific activity and product

solubility. However the greater capacity of the definitive unit was more suited to processing the increased quantities of plasma supplied subsequently. This equipment was used for thawing all plasma at PFC for the next 17 years. In mid-1998, new equipment was introduced following the ban on the processing of plasma from UK donors. Construction of the new unit was based on the original design.

2.3 **Question**

"Paragraph 2.12 *When (month and year) did Scotland become self-sufficient in Factor VIII concentrate derived from unpaid blood donors?"*

Response

In paragraph 2.12, self-sufficiency is defined as having available "sufficient Factor VIII for the treatment of all people in Scotland with haemophilia A according to UK clinical practice". To estimate when this was achieved it is necessary to examine year by year the quantity of FVIII concentrate used in the UK per head of population and to compare this with the quantity of Factor VIII concentrate produced for use in Scotland by SNBTS per head of population. Information available to SNBTS on the purchase of commercial imports is also listed. This information is shown below for the period 1978-1988.

YEAR TO 31 DEC	QUANTITY OF FACTOR VIII CONCENTRATE (IU/head of population)		
	UK USAGE NHS+COMMERCIAL	ISSUED BY SNBTS FOR USE IN SCOTLAND	COMMERCIAL IMPORTS SCOTLAND
1978	0.60	0.36	N/A
1979	0.72	0.42	N/A
1980	0.86	0.63	0.20
1981	1.01	0.87	0.27
1982	1.20	0.95	0.27
1983	1.17	1.20	0.20
1984	1.32	1.36	0.02
1985	1.29	1.17	0.01
1986	1.48	1.07	0.02
1987	1.47	1.78	0.04
1988	1.63	1.83	0.03

These data indicate that it was in 1983 that SNBTS produced, for the first time, a quantity of Factor VIII concentrate sufficient to treat all patients in Scotland at the level of treatment being practiced in the UK. The subsequent fall in the purchase of commercial products is consistent with this.

It should be appreciated that these estimates are not precise measurements and that calculations on a monthly basis would not be meaningful, as data on the UK usage of Factor VIII and on the purchase of commercial concentrates are only available on an annual basis.

2.4 **Question**

"Paragraph 3.1 *When was a screening test for hepatitis B introduced?"*

Response

A screening test for hepatitis B was introduced by SNBTS in 1970 using the method of counterimmunoelectrophoresis (CIE), improved methods were introduced subsequently.

2.5 **Question**

"Paragraph 4.1 *When did SNBTS introduce heat treatment of albumin products?"*

Response

Human Albumin was first produced by SNBTS in 1965. The product was heat treated by pasteurisation for 10 hours at 60°C.

2.6 **Question**

"Paragraph 4.10 *Was the ICTH protocol followed in the UK? Did SNBTS have any evidence that patients in the studies actually knew they were being studied; were they told about test results and about their condition?"*

Response

The ICTH recommendations were based on a protocol designed for a study undertaken in haemophilia centres in London, Paris, Milan and Heidelberg (see reference 32 in the original SNBTS report).

The protocol for the study of viral safety of the SNBTS products Z8 and HTDEFIX (ref 64) was based on the ICTH protocol as revised in 1988 (ref 39). Some patients did not comply fully with the protocol either because insufficient samples were collected or because patients had previously been treated with a small number of single donations. The ICTH protocol was

devised to enable the risk of transmission of NANB hepatitis to be determined using non-specific liver function tests and the advent of specific, sensitive tests for HCV infection that were used in the Scottish study made some of these requirements unnecessary. The number of patients included in the Scottish study was less than recommended in the ICTH protocol, a consequence of the study being restricted to a country with a relatively small population.

SNBTS was not involved in the evaluation of products from BPL nor in UK studies in which products from other manufacturers were used. However, according to the published report (ref 60) the first hepatitis safety study of BPL's 8Y and 9A products did not comply fully with ICTH criteria. It was for this reason that a further study of 8Y was carried out (ref 62), adhering strictly to the ICTH guidelines as revised in 1988 (ref 39).

The protocol for residual infectivity studies on SNBTS heat treated Factor VIII concentrate, drafted by SNBTS in February 1985, stated that informed consent was required from patients or their parents. However, the SNBTS medical staff concerned with these studies are no longer employed by us and as SNBTS neither treats haemophilia patients nor holds their medical records we are unable to provide the evidence requested.

2.7 **Question**

"Paragraphs 5.4, 5.5, 5.6 — I think it would be useful to our investigation to have dates as precisely as possible."

Response

These paragraphs describe our work on a new method of purification of FVIII that was aimed at resolving the difficulties that we had encountered in attempting to develop a pasteurised FVIII concentrate, the similar approach taken by Behringwerke in Germany and the length of time taken by Bayer in the USA to develop a pasteurised FVIII concentrate.

SNBTS

We first learned that Professor Johnson was working on a new method of FVIII purification on 27th June 1983 at the Stockholm Congress of the World Federation of Hemophilia when, in a private discussion following an SNBTS presentation, he enquired if we might be interested in working on this project with him as he believed that we were thinking along the same lines as himself.

His procedure was claimed to capable of producing FVIII with a specific activity of over 100 IU/mg, to be high-yielding and relatively simple to adopt. The potential value of this in resolving the technical difficulties that we were experiencing in the development of pasteurisation was immediately appreciated; we agreed to collaborate with Professor Johnson and formal agreements were drawn up for this purpose.

In January 1984 we were advised by Dr Ludlam that our pilot batch of pasteurised FVIII which had been infused in September, October and November 1983 had produced "significant and unacceptably adverse reactions in the recipient". Although the cause of these reactions was not known, this response provided another reason for seeking the very substantial increase in purity offered by Professor Johnson's procedure.

Professor Johnson was planning to exploit his discovery to fund his research group at New York University Medical Centre and, due to commercial concerns over secrecy, he was unable to provide SNBTS with details of the procedure immediately. Further information was eventually supplied to SNBTS at a meeting in his laboratory on 14th June 1984.

The information disclosed indicated that the procedure utilised high concentrations of carbohydrate and calcium stabilisation, similar to our ZHT process. However, although the method seemed very promising, the specific procedures and reagents proposed by Professor Johnson possessed insufficient capacity for large-scale production. To address this problem, we arranged a meeting between Professor Johnson and Pharmacia AG, Europe's leading supplier of the type of purification reagent and equipment used in Johnson's process. This meeting was held in Munich on 14th July, during the Congress of the International Society of Blood Transfusion.

Pharmacia identified one of their products under development as a potential candidate for this purpose and agreed to supply samples for evaluation; these were received by SNBTS on 22nd August and we began work immediately. By October 1984, we had initial results from small-scale experiments, which suggested that this new material was effective and that the early stages of our ZHT process could be integrated into Johnson's process. Some of these data were included in Johnson's patent application which was filed on 1st February 1985 (ref 40).

In moving to a new purification reagent, it was necessary to redefine all of the processing conditions, and to investigate scale-up of all of these operations. In April 1985 three members of SNBTS staff were sent to the laboratories of Pharmacia in Sweden to receive training in scale-up and in the large-scale operation of the new purification technology as well as to discuss the performance of the purification reagent and further potential developments. Equipment for operating the purification technology in production was specified by SNBTS in October 1985 with delivery being completed by Pharmacia by mid-1986.

It was also necessary to design a stable dose form for the highly purified final product. It was whilst working on this latter aspect that on 21st October 1985 we discovered a set of freeze drying conditions which allowed a control preparation of lower purity FVIII to tolerate dry heat treatment at 80°C.

Previously, the nature of the freeze drying cycle had not been considered to be particularly important for achieving 80°C dry heat treatment and the relevant conditions had not been included by BPL in their patent application for 8Y. After completing further experiments to confirm our results, details of the freeze drying process used for 8Y were requested from BPL. This information was received on 17th December and indicated that the freeze drying conditions used for 8Y were comparable to those discovered by ourselves, confirming to us that freeze drying was indeed the key process stage for achieving 80°C dry heat treatment, rather than FVIII purification per se. On 23rd December 1985 we reviewed all of this information and decided to begin development of an 80°C dry heat treated FVIII concentrate without the use of extensive purification.

Behringwerke

On 31st August 1984 Behringwerke filed a patent application (ref 41) concerning the use of calcium stabilisation of FVIII during pasteurisation and the use of a purification method for the recovery of FVIII following pasteurisation that was similar to that developed by Johnson's group. This was published (in German) on 6th March 1986. According to this application, Behringwerke were following a similar route to ourselves, presumably because they were having similar technical difficulties with their own process of pasteurisation. The experiments described in the patent application were carried out at a relatively small-scale (ie. 2.5 litres of plasma); when large-scale application was achieved is not known.

Bayer

A patent application concerning the pasteurisation of FVIII was originally filed by Bayer (Cutter Laboratories) on 5th March 1980 based on discoveries made in their research laboratory during 1978/79. This process was very similar to the original pasteurisation process of Behringwerke. The final version of the patent (ref 42) was published on 3rd April 1984 and the product (Koate HS) was approved for clinical use by the FDA in April 1986 (ref 29). This was superceded by a chemically (solvent /detergent) treated FVIII concentrate (Koate HP) which was licensed by the FDA in March 1989 (ref 29).

2.8 **Question**

"Paragraph 6.12 *"Some years later" - when?"*

Response

A total of six batches of NY-HT prepared prior to the introduction of HIV screening were subsequently found to have been prepared from plasma containing an HIV-positive donation. These batches were heat treated in November 1984 (two batches), January 1985, February 1985, May 1985 and June 1985 and were issued for use in December 1984 (two batches), March 1985 (two batches), August 1985 and September 1985.

Following the introduction of HIV testing, SNBTS tested archive samples of earlier donations for any donor found to be HIV-positive and the fate of these previous donations was traced. The date at which this information became available depended on the date at which these individuals returned to donate blood following the introduction of HIV screening. The dates on which it was learned that HIV-positive donations had been processed ranged from 29th January 1986 to 7th December 1988.

Products found to have been prepared using an HIV-positive donation were recalled immediately this information was available. No haemophilia patient who was treated with any of these batches was infected with HIV as a consequence.

2.9 **Question**

"Paragraph 8.7 When were ovens obtained and put to use?"

Response

The process of specifying a high accuracy heat treatment cabinet for use by SNBTS was begun in January 1985. The first cabinet that we purchased was received and commissioned in July 1985 and was used immediately thereafter for the dry heat treatment of Factor VIII concentrate at 68°C and Factor IX concentrate at 80°C.

3. GENERAL QUESTIONS

3.1 **Question**

"The document sets out a very useful explanation of events. To supplement this, would it also be possible for SNBTS to provide in table form a basic chronology (a kind of "at a glance guide" to what happened with dates)?"

Response

A basic chronology in tabular form is given in table 1.

3.2 **Question**

"What was SNBTS expenditure on R&D each year 1980-1990?"

Response

During the period 1980-1990, SNBTS did not record its expenditure on R&D as a specific category of expenditure.

In 1992 an internal audit of R&D was undertaken in which expenditure for the financial year 1990/91 was documented. This exercise only covered non-medical staff employed wholly in an R&D capacity, excluding medical staff and those employed in a "service" capacity who might also contribute to

R&D activities. According to this audit, the total SNBTS revenue expenditure on R&D for 1990/91 was £1.22M, representing 5.9% of the SNBTS revenue budget for that year. The addition of relevant clinical staff, plus "service" staff contributing to R&D would probably have increased this figure to about 7.5% of the total SNBTS revenue budget. These sums do not include funding for the implementation of service developments arising from R&D programmes (eg. to implement the manufacture of hepatitis-safe FVIII).

Despite being a relatively small organisation, SNBTS was able to develop hepatitis-safe coagulation factor concentrates well before most other manufacturers in the world, including most of the large commercial companies operating in this field. This achievement demonstrates that it is the quality of the R&D which is most important, rather than the size of the R&D budget.

3.3 **Question**

"What are the details of the history of donor selection policy in the 1970's and 1980's?"

3.4 **Question**

"What screening methods were used between what dates from 1970 to 1990? How did this compare with international measures?"

3.5 **Question**

"We understand that SNBTS did not export any blood products at the time, but was any whole blood exported, and if so what testing was undertaken before export?"

Response

The information requested in questions 3.3, 3.4 and 3.5 will be provided separately in a specific document.