amersham pharmacia biotech

To whom it may concern

ON ANIMAL DERIVED MATERIALS IN THE MANUFACTURE OF
17-0448-00*) Cytodex™ 1 CELL CULTURE MEDIA

The only use of animal derived components in the manufacture of Cytodex 1 chromatography media is in the manufacture of the base material raw dextran. The raw dextran is further processed to Dextran substances used also for medicinal products intended for injections. These medicinal products are and have been during many years been extensively used throughout the world.

Animal-derived components in raw material in the production of Dextran substances

The manufacturing of Dextran products, used for both clinical and technical applications, at Amersham Pharmacia Biotech AB involves the two production sites at Staffanstorp and Uppsala, Sweden.

The raw dextran is manufactured in a fermentation plant in Staffanstorp. A change in raw materials, comprising exclusion of the animal-derived components Trypton and porcine Liver Broth, in the inoculum substrate for production of inoculum culture for *Leuconostoc mesenteroides* has been introduced.

Another change, introduced concurrently, comprises the exclusion of all sources of animal origin when growing the bacterias for new working cell banks.

These changes have been validated to assure the consistency in process performance and product quality.

The only animal-derived component in the production of dextran that now remains is the skim-milk powder acting as cryo-protective agent at the lyophilisation of new cell banks. This skim-milk powder corresponds to level of less than 0.01 ppm in the final Dextran bulk substances.

The country of origin for this skim milk powder is USA. Milk is not a specified risk material as defined in Commission Decision 97/534/EC and is also unlikely to present any risk of contamination according to the European Pharmacopoeia monograph Products with risk of transmitting agents of animal spongiform Encephalopathies. The supplier intends to submit an application for a Certificate of Suitability no later than mid-September, 2000.

The changes in Staffanstorp were finally introduced in regular production of dextran in 1999 and are fully implemented for all batches of Dextran substances manufactured after 1 January 2000 (lot numbers > 281800).

*) The last two digits (-00) of the code number includes all the different packaging sizes.

Amersham Pharmacia Biotech AB tel +46 18 612 0000
Björkgatan 30 fax +46 18 612 12 00
SE-751 84 Uppsala
Sweden http://www.apbiotech.com

CAIF SA
Dra. Bernarda Belay
Co-Directora Tecnica
M.N. 15.148

CAIF
Compañía Argentina de
Investigaciones Farmacéuticas S.A.
Dra. María Bernarda Belay
Apoderada
DNI 29378925

Dextran substances manufactures before January 1, 2000

For batches manufactured before January 1, 2000 was the above mentioned Tryptone and porcine Liver Broth used.

Tryptone is derived from milk. This sources is not defined as specified risk material as defined in Commission Decision 97/534/EC

The tryptone used is, as of December 1996, manufactured from animals originating from countries free from bovine spongiform encephalopathy (BSE) as declared by the supplier. The countries of origin for the tryptone are Australia, New Zealand and USA. The supplier intends to submit an application for a Certifiate of Suitability no later than mid-September , 2000.

Uppsala, Sweden, August 25, 2000

Lena Hellquist
Regulatory Support

CAIF SA
Dra. Bernarda Belay
Co-Directora Tecnica
M.N. 15.146

CAIF
Compañía Argentina de Investigaciones Farmacéuticas S.A.
Dra. María Bernarda Belay
Apoderada
DNI 93.120.675

GE Healthcare

Animal Origin Statement

Regarding animal derived materials in Cytodex 1 surface microcarriers

Product:	Code Number:
Cytodex™ 1	17-0448-xx*
	*)-xx refers to all pack sizes

Based on the information below it is hereby stated that Cytodex 1 surface microcarriers do not contain, and is not derived from, specified risk material as defined in Regulation (EC) No 999/2001 (as amended).

One of the raw materials used for manufacture of Cytodex 1 microcarriers is dextran. This is the only raw material in the manufacture of the mentioned products containing animal derived materials.

After January 1, 2000
A change in raw materials, comprising exclusion of the animal-derived components tryptone and liver infusion broth, in the inoculum substrate for production of inoculum culture for *Leuconostoc mesenteroides* has been introduced. The only animal derived component in the production of dextran, after January 1, 2000, is the skim milk powder acting as a cryo-protective agent at the lyophilization of new cell banks. The country of origin for this skim milk powder is USA. More information on the skim milk is found below.

Before January 1, 2000
During the manufacture of one of the raw materials, native dextran, minute amounts of broth containing extracts from porcine liver (liver infusion broth) and bovine tryptone (Bacto Tryptone), are added to the stock culture of *Leuconostoc mesenteroides*.

Milk is the bovine component of the liver infusion broth. The countries of origin for the milk are Australia, New Zealand and USA. The porcine material is sourced from USA, Canada and Italy.
The countries of origin for the tryptone (Bacto Tryptone) are Australia, New Zealand and USA.

GE Healthcare Bio-Sciences AB
Björkgatan 30
SE-751 84 Uppsala
Sweden
T +46 (0)18 612 00 00
F + 46 (0)18 612 12 00
Reg.No SE 55 61 08 1919

GE Healthcare

Skim milk powder is used as cryo-protective agent at the lyophilization of new cell banks. This skim-milk powder corresponds to level of less than 0.01 ppm in the final Dextran bulk substances. The country of origin for this skim milk powder is USA.

The milk for liver infusion broth, tryptone and skim milk are sourced from animals "either deemed fit for human consumption or deemed healthy" as stated by the supplier, BD Diagnostics, USA. This information is in agreement with the European guideline "Minimising the risk of transmitting animal spongiform encephalopathy agents via human and veterinary medicinal products (EMEA/410/01- Rev 2)". Certificates of Suitability are not relevant for these raw materials.

It should be added that, during manufacture, dextran is subjected to several denaturing processes:

- fractionation at an ethanol concentration of 40% for a minimum of 32 hours
- treatment at pH 1.3 at 85°C for a minimum of 10 hours
- spray drying- inlet air temperature 230°C

Uppsala, Sweden, August 29, 2006

GE Healthcare Bio-Sciences AB
Regulatory Affairs

Lena Hellquist
Senior Regulatory Affairs Manager

CAIF SA
Dra. Bernarda Belay
Co-Directora Tecnica
M.N. 15.148

GE Healthcare Bio-Sciences AB
Björkgatan 30
SE-751 84 Uppsala
Sweden
T +46 (0)18 612 00 00
F + 46 (0)18 612 12 00
Reg.No SE 55 61 08 1919

CAIF
Compañía Argentina de Investigaciones Farmacéuticas S.A.
Dra. María Bernarda Belay
Apoderada
DNI 23378925

Celite 577

Specification	Requirement
Identity	
-	
Chemical tests	
Total ash	≤ 0.50 %
Particle size	≤ 40.0 µm
pH	< 7.0
Microbiological properties	
-	

CAIF SA
Dra. Bernarda Belay
Co-Directora Tecnica
M.N. 15.148

CAIF
Compañía Argentina de Investigaciones Farmacéuticas S.A.
Dra. María Bernarda Belay
Apoderado
DNI 29078925

Guanine hydrochloride

Specification	Requirement
Identity	
Melting point	≥ 300 °C
Chemical tests	
Total ash	≤ 0.05 %
Total impurities	≤ 2 %
Content	≥ 99.0 %
Microbiological properties	
-	

NVI
nederlands vaccin instituut

The animals (Cynomolgus monkeys (Macaca fascicularis)) used for monkey kidney cell preparation were tested and complied according to the following tests:

Specification	Requirement
Antibodies to	
- Herpesvirus simiae (B virus)	negative
- SV40	negative
- Foamyvirus type 1, 2 and 3	negative
Tuberculosis (Mantoux reaction)	negative
Physical condition	no signs of illness

In a frequency of once per year, the monkeys were tested for the absence of antibodies against the agents mentioned in the table. Just prior to the kidney perfusion, these tests were repeated, with the exception of SIV.

Specification	Requirement	Validity result
Antibodies to:		
- SIV	negative	12 months
- Foamy	negative	14 days
- Herpes simplex virus*	negative	14 days
- SV 40	negative	14 days
- Polyoma	negative	14 days
M. tuberculosis (Mantoux test)	negative	14 days
Physical condition	no signs of illness	
Pathology	no abnormalities	

* as indicator for HV simian B

CAIF SA
Dra. Bernarda Belay
Co-Directora Tecnica
M.N. 15.148

CAIF
Compañía Argentina de Investigaciones Farmacéuticas S.A.
Dra. María Bernarda Belay
Apoderada
DNI 29[illegible]925

NVI
nederlands
vaccin
instituut

The monkey kidney cells were tested and complied according to the following requirements:

Control cell cultures:

Test	Method	Requirement
Test for CPE and haemadsorbing viruses	According to Ph. Eur. 2.6.16. CPE is assessed by microscopical examination. Addition of guinea-pig red blood cells.	No evidence of CPE or haemadsorbing agents is found.
Extraneous agents	According to Ph. Eur. M-0214 Test in rabbit kidney cell cultures.	No evidence of Herpes-B virus or other adventitious agents is found
Extraneous agents	According to Ph. Eur. M-0214 Test in cercopithecus kidney cell cultures	No evidence of SV40 or other adventitious agents is found
Mycobacteria	According to Ph. Eur. 2.6.2 Inoculation in Loewenstein, 7H10 and Tween-albumin media.	No growth of mycobacteria

CAIF SA
Dra. Bernarda Belay
Co-Directora Tecnica
M.N. 15.148

CAIF
Compañía Argentina de Investigaciones Farmacéuticas S.A.
Dra. María Bernarda Belay
Apoderada
DNI 29378925

Invitrogen
life technologies

Invitrogen Limited
3 Fountain Drive
Inchinnan Business Park
Paisley PA4 9RF
Tel: 0141 814 6100
Fax: 0141 814 6258
www.invitrogen.com

CERTIFICATE OF ORIGIN STATEMENT

Product Description: Trypsin 1:250
Catalogue Number: 27250

This is to certify that the above mentioned product has the following source and origin:

Source	Origin
Porcine Pancreas	USA/Canada
Lactose	USA

We can confirm that USA and Canada are considered to be free of BSE/TSE.

Debbie Hamilton
Quality Systems

CAIF SA
Dra. Bernarda Belay
Co-Directora Tecnica
M.N. 15.148

CAIF
Compañía Argentina de Investigaciones Farmacéuticas S.A.
Dra. María Bernarda Belay
Apoderada
DNI 29378925

GIBCO™
Invitrogen Corporation

Raw Material Technical Information

Product: Trypsin 1:250

Raw Material Part #: 840-7072 (Used in all GIBCO brand Trypsin solutions)

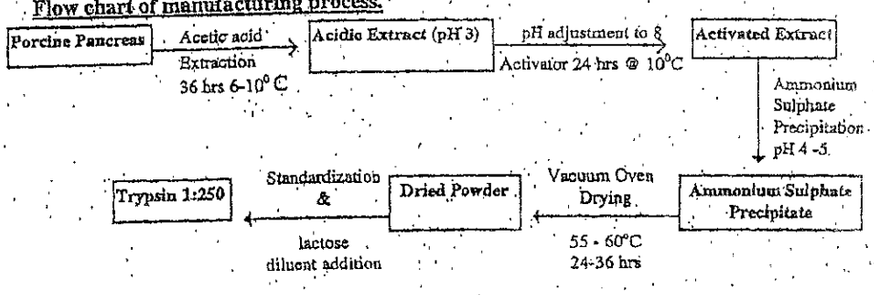
Source: Porcine Pancreas

Country of Origin: US or Canada

Country of Manufacture: Canada

Manufacturing Process:
Flow chart of manufacturing process

Flow chart of manufacturing process.



Manufacturing notes:

- The bovine lactose used to adjust the activity of trypsin powder is of US origin. This lactose is refined from bovine milk and derived from healthy animals in the same conditions as milk collected for human consumption. No other ruminant materials are used during the processing.
- The raw powder Trypsin 1:250 is then irradiated at a minimum of 25 kGY. A certificate of irradiation accompanies each lot of Trypsin Invitrogen receives.

Regulatory Information:
The bovine lactose and trypsin itself are excluded from the scope of the Note for Guidance on "Minimizing the risk of transmitting animal Spongiform encephalopathy agents via human and veterinary medicinal products" (Revision February 2001, EMEA/410/01-Final).

Additional Information:
Pancreas Collection
Pork Pancreas glands are collected in USDA licensed pork slaughter plants under the inspection of USDA veterinarians. Pigs are subjected to ante- and post-mortem inspection, and glands are saved only from animals judged acceptable for human consumption.

HyClone®

HyClone New Zealand
Omokoroa Farm
441 Old Highway
P.O. Box 656
Tauranga, New Zealand

Product: **DONOR BOVINE SERUM**
Collected and Processed in New Zealand
Custom Filtered for NVI
Catalog#: **SH3A1248.01** Manufacture Date: **OCT/2006**
Lot#: **DRK0265** Expiration Date: **OCT/2011**
Filtration: **0.2 µm Sterile Filtered** Total Batch Volume: **1034.7 L**

CERTIFICATE OF ANALYSIS

Test/(Method)	Specification	Units	Results
Endotoxin (Limulus Amebocyte Lysate-Gel Clot, Current USP and EP)	≤ 5	EU/mL	<0.06
Haemoglobin (Spectrophotometric)	≤ 4	mg/dL	2
Sterility Testing (Ph. EUR. 2.6.1) (Current USP and EP)			
Bacteria and Fungi	No Growth		No Growth
Virus Testing (Full EMEA)			
Fluorescent Antibody			
Bluetongue	Not Detected		Not Detected
Bovine Adenovirus	Not Detected		Not Detected
Bovine Parvovirus	Not Detected		Not Detected
Bovine Respiratory Syncytial Virus	Not Detected		Not Detected
Bovine Viral Diarrhoea Virus	Not Detected		Not Detected
Bovine Leukemia Virus	Not Detected		Not Detected
Rabies	Not Detected		Not Detected
Reovirus	Not Detected		Not Detected
Cytopathogenic Agents – e.g. IBR	Not Detected		Not Detected
Haemadsorbing Agents – e.g. PI3	Not Detected		Not Detected
Bovine Virus Diarrhoea Virus (Pre-Irradiation)			
Interference/Comparative Titration	No Interference		No Interference
Bovine Virus Diarrhoea Virus (Post-Irradiation)			
Interference/Comparative Titration	No Interference		No Interference
Irradiation Dose	25-35 kGy		27,53 – 32,06
pH	For Information Only		8.02
Osmolality (Ph. Eur 2.2.35)	240-340	mOsm/kg	269

Phone: + 64 7 548 0225
Fax: + 64 7 548 1296
Toll Free: 800-522-5355
Internet: www.hyclone.com
Email: info@hyclone.com

HyClone

HyClone New Zealand
Omokoroa Farm
441 Old Highway
P.O. Box 568
Tauranga, New Zealand

Product: **DONOR BOVINE SERUM**
Collected and Processed in New Zealand
Custom Filtered for NVI

Catalog#: **SH3A1248.01** Manufacture Date: **OCT/2006**
Lot#: **DRK0265** Expiration Date: **OCT/2011**
Filtration: **0.2 µm Sterile Filtered** Total Batch Volume: **1034.7 L**

CERTIFICATE OF ANALYSIS

Test/(Method)	Specification	Units	Results
Total Protein	≥ 35	mg/mL	69
Albumin	For Information Only	g/dL	3.55
Alpha – 1 Globulin	For Information Only	g/dL	0.27
Alpha – 2 Globulin	For Information Only	g/dL	0.99
Beta Globulin	For Information Only	g/dL	1.55
Gamma Globulin	For Information Only	g/dL	0.74

The raw material used to produce this batch of serum has been sourced exclusively from clinically healthy cattle which were born and maintained in a country which is officially recognised as being free from Bovine Spongiform Encephalopathy (BSE), namely New Zealand

The raw material used was not derived from animals:

- Which have died on the farm, including newborn animals or foetuses.
- Which have been killed to eradicate epizootic diseases.
- Showing any clinical signs and/or lesions of contagious diseases for humans, or for reasons of residues, were declared unfit for human consumption.

The raw materials used were sourced from animals free from Tuberculosis, Leptospirosis, Brucellosis, Bluetongue and Bovine Leukemia Virus. The donor animals are screened regularly for these infectious agents unless the country of origin is declared to be free from these agents by the current OIE. The source herd is not vaccinated against these agents.

South Pacific Sera has been granted a Certificate of Suitability to the European Pharmacopoeia Monograph for New Zealand Sourced Donor Bovine Serum. The current certificate R1-CEP 2000-341 is included.

21 JUN 07
Caroline Newcombe
QA/QC Manager

Page 2 of 2

Phone: + 64 7 548 0226
Fax: + 64 7 548 1295
Toll Free: 800-522-5355
Internet: www.hyclone.com
Email: info@hyclone.com

HyClone®

HyClone New Zealand
Omokoroa Farm
441 Old Highway
P.O Box 668
Tauranga, New Zealand

Product: **DONOR BOVINE SERUM**
Collected and Processed in New Zealand
Custom Filtered for NVI
Catalog#: **SH3A1248.01**
Lot#: **DRK0265**
Filtration: **0.2 µm Sterile Filtered**

Manufacture Date: **OCT/2006**
Expiration Date: **OCT/2011**
Total Batch Volume: **1034.7 L**

BIOCHEMICAL ASSAY

Test	Lot	Value
Alkaline Phosphatase	133	U/L
Blood Urea Nitrogen	10	mg/dL
Calcium	9.7	mg/dL
Chloride	98	mmol/L
Cholesterol	108	mg/dL
Creatinine	1.2	mg/dL
Glucose	38	mg/dL
Glutamic Oxaloacetic Transaminase (SGOT)	59	U/L
Glutamic Pyruvic Transaminase (SGPT)	61	U/L
Inorganic Phosphorus	6.5	mg/dL
Lactate Dehydrogenase (LDH)	2743	U/L
Potassium	4.5	mmol/L
Sodium	136	mmol/L
Specific Gravity	1.00372	g/mL
Total Bilirubin	<0.1	mg/dL

Phone: + 64 7 548 0225
Fax: + 64 7 548 1295
Toll Free: 800-322-6355
Internet: www.hyclone.com
Email: info@hyclone.com