

CHP molecular diagnostic protocols for the detection of

human swine influenza virus type A (subtype H1)

revision 2 (16 December 2009)¹

The Virology Division, Public Health Laboratory Services Branch, Centre for Health Protection (CHP), Department of Health, Hong Kong SAR, China, uses the following protocols for the detection of human swine influenza virus type A (subtype H1). This document may undergo periodic modification as information about current circulating viruses accumulates.

Contents	<i>Page</i>
1. Introduction	2
2. Specimens	4
3. Biosafety	4
4. Conventional RT-PCR	5
5. Real-time RT-PCR	12
6. Referral for confirmation and further characterization	21
7. Quality assurance	21

¹ This document replaces the document ‘CHP molecular diagnostic protocols for the detection of human swine influenza virus type A (subtype H1) *revision 1 (25 May 2009)*’.

1. Introduction

Polymerase chain reaction (PCR) is currently one of the methods of choice to confirm human swine influenza type A (subtype H1) virus infection. A sample is considered to be positive when results from tests using primers specific for universal M gene and human swine H1 haemagglutinin gene are positive while PCR for seasonal H1 and H3 genes are negative. This document provides information on conventional RT-PCR and real-time RT-PCR protocols available at CHP for the detection of human swine influenza type A (subtype H1) A/California/4/2009-like viruses and seasonal influenza subtype H1 and H3 viruses (see Table on next page). Real-time RT-PCR assay targeting swine H1 haemagglutinin gene assay (988 primer/probe set) in earlier version had been optimized by using one-step real-time RT-PCR protocol to shorten the turn-around time. In this version, a new real-time human swine H1 haemagglutinin gene assay (91M primer/probe set) has also been included for supplementary testing of screening positive samples.

As the influenza virus continues to mutate and that the prevalence of seasonal and human swine influenza viruses are changing, to ensure efficient and effective diagnosis, primer/probe used should be reviewed based on genetic characteristics and testing algorithms modified in accordance with the prevalence of circulating influenza viruses.

Table. Summary of the conventional RT-PCR and real-time RT-PCR protocols for the detection of human swine influenza type A (subtype H1).

RT-PCR detection method	Target gene	Protocol	PCR method		Materials required	Primer/probe and thermal cycling conditions	Procedure
			One-step	Two-step			
Conventional	A/MA	Conventional RT-PCR for the detection of the matrix gene - influenza virus type A	√		page 5	page 6	page 10
Conventional	A/H1v	Conventional RT-PCR for the detection of hemagglutinin gene - human swine influenza virus type A (subtype H1)	√		page 5	page 7	page 10
Conventional	A/H1	Conventional RT-PCR for the detection of hemagglutinin gene - seasonal influenza virus type A (subtype H1)	√		page 5	page 8	page 10
Conventional	A/H3	Conventional RT-PCR for the detection of hemagglutinin gene - seasonal influenza virus type A (subtype H3)	√		page 5	page 9	page 10
Real-time	A/MA	Real-time two-step RT-PCR for the detection of the matrix gene - influenza virus type A		√	page 12	page 13	page 18
Real-time	A/H1v ^a	Real-time two-step RT-PCR for the detection of the hemagglutinin gene - human swine influenza virus type A (subtype H1) by 988 primer/probe set		√	page 12	page 14	page 18
Real-time	A/H1v	Real-time one-step RT-PCR for the detection of the hemagglutinin gene - human swine influenza virus type A (subtype H1) by 988 primer/probe set	√		page 12	page 15	page 18
Real-time	A/H1v	Real-time one-step RT-PCR for the detection of the hemagglutinin gene - human swine influenza virus type A (subtype H1) by 91M primer/probe set	√		page 12	page 16	page 18
Real-time	A/H1 & A/H3	Real-time two-step RT-PCR for the detection of the hemagglutinin gene - seasonal influenza virus type A (subtypes H1 and H3)		√	page 12	page 17	page 18

^a A/H1v is referred to as human swine influenza type A (subtype H1).

2. Specimens

Respiratory specimens are most appropriate. Preferred respiratory specimens include nasopharyngeal swab, nasal aspirate or a combined nasopharyngeal swab with oropharyngeal swab. If these specimens cannot be collected, a nasal swab or oropharyngeal swab is acceptable².

3. Biosafety

All laboratory practices involving clinical specimen from patients who are suspected cases of human swine influenza virus type A (subtype H1) infection should be conducted in a laboratory with a minimum biosafety level 2 facilities. Details on biosafety guidelines for human swine influenza virus type A (subtype H1) are available at CHP web site³.

² Please see 'Interim Guidance on Specimen Collection, Processing, and Testing for Patients with Suspected Novel Influenza A (H1N1) Virus Infection', 25 May 2009, date last accessed:
<http://www.cdc.gov/h1n1flu/specimencollection.htm>

³ Please see 'Biosafety Guidelines for Human Swine Influenza A (H1N1)', 25 May 2009, date last accessed:
[http://www.chp.gov.hk/files/pdf/Biosafety_Guidelines_for_Human_Swine_Influenza_A_\(H1N1\).pdf](http://www.chp.gov.hk/files/pdf/Biosafety_Guidelines_for_Human_Swine_Influenza_A_(H1N1).pdf)

4. Conventional RT-PCR

4.A. Conventional RT-PCR - materials required

The materials required for conventional RT-PCR to detect influenza virus type A (subtype H1) in specimens from humans are given below:

Equipment and consumables

- Sterile, RNase-free pipette tips with aerosol barrier
- Adjustable pipettes (10, 20, 200, 100 μ L)
- Vortex
- Microcentrifuge tubes (0.2, 1.5 mL)
- Microcentrifuge (adjustable, up to 13 000 rpm)

RNA extraction

- QIAamp Viral RNA Mini Kit (QIAGEN)
- Ethanol (96 - 100%)

Conventional RT-PCR

- Applied Biosystems 9700 Thermocycler
- OneStep RT-PCR Kit (QIAGEN)
- Q-solution (from QIAGEN OneStep RT-PCR Kit)
- RNase Inhibitor 20U/ μ L (Applied Biosystems)
- Primers pair
- Positive control (can be obtained upon request from CHP)

4.B. Conventional RT-PCR - primers and PCR thermal cycling conditions

Conventional RT-PCR for the detection of the matrix gene - influenza virus type A

Primers

Name	Oligonucleotide sequence (5' to 3')	Direction ^a	Location ^b	Size ^c
M30F	TTCTAACCGAGGTCGAAACG	+	8-27	
M264R2	ACAAAGCGTCTACGCTGCAG	-	239-220	232 ^d

^a +, sense; -, anti-sense.

^b Location of the oligonucleotides based on the strain of human swine influenza virus type A (subtype H1) A/California/4/2009, GenBank Accession: FJ969513.

^c The size (base pairs) of the PCR product as predicted from the sequence.

^d From National Institute of Infectious Diseases (NIID), Tokyo, Japan, WHO Collaborating Centre for Reference and Research on Influenza and WHO H5 Reference Laboratory.

PCR thermal cycling conditions^a

Step	Temperature (°C)	Time
Reverse transcription	50	30 min
Initial PCR activation	95	15 min
3-step cycling (45 repeated cycles):		
- denaturation	94	30 sec
- annealing	50	30 sec
- extension	72	30 sec
Final extension	72	2 min

^a This protocol was designed without using Q-solution.

4.B. Conventional RT-PCR - primers and PCR thermal cycling conditions

Conventional RT-PCR for the detection of hemagglutinin gene - human swine influenza virus type A (subtype H1)

Primers

Name	Oligonucleotide sequence (5' to 3')	Direction ^a	Location ^b	Size ^c
H1-sw-434f	CGAACAAAGGTGTAACGGCAGCAT	+	434-457	
H1-sw-905r	GCACCCTTGGGTGTTTGACAAGTT	-	905-882	472

^a +, sense; -, anti-sense.

^b Location of the oligonucleotides based on the strain of human swine influenza virus type A (subtype H1) A/California/4/2009, GenBank Accession: FJ966082.

^c The size (base pairs) of the PCR product as predicted from the sequence.

PCR thermal cycling conditions^a

Step	Temperature	Time
Reverse transcription	50	30 min
Initial PCR activation	95	15 min
3-step cycling (45 repeated cycles):		
- denaturation	94	30 sec
- annealing	55	30 sec
- extension	72	30 sec
Final extension	72	2 min

^a This protocol was designed using Q-solution.

4.B. Conventional RT-PCR - primers and PCR thermal cycling conditions

Conventional RT-PCR for the detection of hemagglutinin gene - seasonal influenza virus type A (subtype H1)

Primers

Name	Oligonucleotide sequence (5' to 3')	Direction ^a	Location ^b	Size ^c
H1-1	GATGCAGACACAATATGTATAGG	+	78-100	
H1-2	CTACAGAGACATAAGCATTT	-	684-665	607

^a +, sense; -, anti-sense.

^b Location of the oligonucleotides based on the strain of seasonal influenza virus type A (subtype H1) A/Brisbane/59/2007, GenBank Accession: CY030230.

^c The size (base pairs) of the PCR product as predicted from the sequence.

PCR thermal cycling conditions^a

Step	Temperature	Time
Reverse transcription	50	30 min
Initial PCR activation	95	15 min
3-step cycling (45 repeated cycles):		
- denaturation	94	30 sec
- annealing	55	30 sec
- extension	72	30 sec
Final extension	72	2 min

^a This protocol was designed without using Q-solution.

4.B. Conventional RT-PCR - primers and PCR thermal cycling conditions

Conventional RT-PCR for the detection of hemagglutinin gene - seasonal influenza virus type A (subtype H3)

Primers

Name	Oligonucleotide sequence (5' to 3')	Direction ^a	Location ^b	Size ^c
H3-247	AAATGGGACCTTTTGTGTAACG	+	295-317	
H3-666	CCTGGGTCTAGATCCGATATTCG	-	713-692	419

^a +, sense; -, anti-sense.

^b Location of the oligonucleotides based on the strain of seasonal influenza virus type A (subtype H3) A/Brisbane/10/2007, GenBank Accession: CY039087.

^c The size (base pairs) of the PCR product as predicted from the sequence.

PCR thermal cycling conditions^a

Step	Temperature	Time
Reverse transcription	50	30 min
Initial PCR activation	95	15 min
3-step cycling (45 repeated cycles):		
- denaturation	94	30 sec
- annealing	55	30 sec
- extension	72	30 sec
Final extension	72	2 min

^a This protocol was designed without using Q-solution.

4.C. Conventional RT-PCR - procedure

I. RNA extraction

Extract viral RNA from clinical specimen with QIAamp Viral RNA Mini Kit or equivalent extraction kit according to manufacturer's instructions.

II. Conventional RT-PCR

1. Prepare master mixture for RT-PCR as below:

(for protocol designed without using Q-solution)

Reagent	Volume (μ L) per reaction
- 5X QIAGEN RT-PCR buffer ^a	10
- dNTP mix ^a	2
- Forward primer (5 μ M)	6
- Reverse primer (5 μ M)	6
- Enzyme mix ^a	2
- RNase inhibitor (20U/ μ L)	0.5
- Water ^a	18.5
Total volume	45

^a From QIAGEN OneStep RT-PCR Kit.

(for protocol designed using Q-solution)

Reagent	Volume (μ L) per reaction
- 5x QIAGEN RT-PCR buffer ^a	10
- dNTP mix ^a	2
- 5X Q-Solution ^a	10
- Forward primer (5 μ M)	6
- Reverse primer (5 μ M)	6
- Enzyme mix ^a	2
- RNase inhibitor (20U/ μ L)	0.5
- Water ^a	8.5
Total volume	45

^a From QIAGEN OneStep RT-PCR Kit.

2. Add 5 μ L viral RNA to the above mix.

3. Perform RT-PCR according to the following conditions:

Step	Temperature (°C)	Time
Reverse transcription	50	30 min
Initial PCR activation	95	15 min
3-step cycling (45 repeated cycles):		
- denaturation	94	30 sec
- annealing	XX ^a	30 sec
- extension	72	30 sec
Final extension	72	2 min

^a 'XX' denotes variables depending on different conventional RT-PCR protocols as described above.

4. Analyze the PCR products by agarose gel electrophoresis.

III. Interpretation of results

1. The size of PCR products obtained should be compared to the expected product size (given in each PCR protocol). If the test is run without a positive control, products must be confirmed by sequencing and compared with sequences available in databases (e.g. GenBank).
2. The absence of the correct PCR products (i.e. band of incorrect product size or no band seen in agarose gel electrophoresis) does not rule out the presence of influenza virus. Results should be interpreted together with the available clinical and epidemiological information.

5. Real-time RT-PCR

5.A Real-time RT-PCR - materials required

The materials required for real-time RT-PCR to detect influenza virus type A (subtype H1) in specimens from humans are given below:

Equipment and consumables

- Sterile, RNase-free pipette tips with aerosol barrier
- Adjustable pipettes (10, 20, 200, 100 μ L)
- Vortex
- Microcentrifuge tubes (0.2, 1.5 mL)
- Microcentrifuge (adjustable, up to 13 000 rpm)

RNA extraction

- QIAamp Viral RNA Mini Kit (QIAGEN)
- Ethanol (96 - 100%)

Real time one-step RT-PCR

- Roche LightCycler Version 2.0
- RealTime ready RNA Virus Master kit (Roche)
- Primers and probes mix

Real time two-step RT-PCR

Reverse Transcription

- 10X PCR buffer I with 15 mM MgCl₂ (Applied Biosystems)
- Random hexamer 50 μ M (Applied Biosystems)
- MuLV Reverse Transcriptase 50 U/ μ L (Applied Biosystems)
- RNase Inhibitor 20 U/ μ L (Applied Biosystems)
- RNase-free water
- Positive control (can be obtained upon request from CHP)

Real time PCR

- Roche LightCycler Version 2.0
- LightCycler - FastStart DNA Master HybProbes kit (Roche)
- Primers and probes mix

5.B Real-time RT-PCR - primers/probes and PCR thermal cycling conditions

Real-time two-step RT-PCR for the detection of the matrix gene - influenza virus type A

Primers and probes

Name	Oligonucleotide sequence (5' to 3')	Sense ^a	Location ^b
FLUAM-1F	AAGACCAATCCTGTACCTCTGA	+	144-166
FLUAM-2F	CATTGGGATCTTGCACTTGATATT	+	784-807
FLUAM-1R	CAAAGCGTCTACGCTGCAGTCC	-	238-217
FLUAM-2R	AAACCGTATTTAAGGCGACGATAA	-	864-841
FLUA-1P ^c	TTTGTGTTACGCTCACCGT	+	184-203
FLUA-2P ^c	TGGATTCTTGATCGTCTTTTCTTCAAATGCA	+	809-839

^a +, sense; -, anti-sense.

^b Location of the oligonucleotides based on the strain of human swine influenza virus type A (subtype H1) A/California/4/2009, GenBank Accession: FJ969513.

^c The probe consisted of oligonucleotides with the 5' reporter dye FAM and the 3' quencher dye TAMRA.

The concentration of the primers and probes mix:

Name	Concentration (µM)
- FLUAM-1F	10
- FLUAM-2F	10
- FLUAM-1R	10
- FLUAM-2R	10
- FLUA-1P	5
- FLUA-2P	5

Working primer and probe mix is prepared by mixing the above six reagents in equal volume.

PCR thermal cycling conditions

Step	Temperature (°C)	Time
Initial PCR activation	95	10 min
3-step cycling (50 repeated cycles):		
- denaturation	95	10 sec
- annealing	56	15 sec
- extension	72	10 sec
Cooling	40	30 sec

5.B Real-time RT-PCR - primers/probes and PCR thermal cycling conditions

Real-time two-step RT-PCR for the detection of the hemagglutinin gene - human swine influenza virus type A (subtype H1) by 988 primer/probe set

Primers and probes

Name	Oligonucleotide sequence (5' to 3')	Direction ^a	Location ^b
H1-sw-988F	AGACTGGCCACAGGATTGAGGAAT	+	988-1011
H1-sw-1171R	CGTCAATGGCATTCTGTGTGCTCT	-	1171-1148
H1-sw-1077P ^c	AGGGATGGTAGATGGATGGTACGGTT	+	1077-1102

^a +, sense; -, anti-sense.

^b Location of the oligonucleotides based on the strain of human swine influenza virus type A (subtype H1) A/California/4/2009, GenBank Accession: FJ966082.

^c The probe consisted of oligonucleotides with the 5' reporter dye FAM and the 3' quencher dye TAMRA.

The concentration of the primers and probes mix:

Name	Concentration (µM)
- H1-sw-988F	10
- H1-sw-1171R	10
- H1-sw-1077P	5

Working primer and probe mix is prepared by mixing the above three reagents in equal volume.

PCR thermal cycling conditions

Step	Temperature (°C)	Time
Initial PCR activation	95	10 min
3-step cycling (50 repeated cycles):		
- denaturation	95	10 sec
- annealing	56	15 sec
- extension	72	10 sec
Cooling	40	30 sec

5.B Real-time RT-PCR - primers/probes and PCR thermal cycling conditions

Real-time one-step RT-PCR for the detection of the hemagglutinin gene - human swine influenza virus type A (subtype H1) by 988 primer/probe set

Primers and probes

Name	Oligonucleotide sequence (5' to 3')	Direction ^a	Location ^b
H1-sw-988F	AGACTGGCCACAGGATTGAGGAAT	+	988-1011
H1-sw-1171R	CGTCAATGGCATTCTGTGTGCTCT	-	1171-1148
H1-sw-1077P ^c	AGGGATGGTAGATGGATGGTACGGTT	+	1077-1102

^a +, sense; -, anti-sense.

^b Location of the oligonucleotides based on the strain of human swine influenza virus type A (subtype H1) A/California/4/2009, GenBank Accession: FJ966082.

^c The probe consisted of oligonucleotides with the 5' reporter dye FAM and the 3' quencher dye TAMRA.

The concentration of the primers and probes mix:

Name	Concentration (µM)
- H1-sw-988F	10
- H1-sw-1171R	10
- H1-sw-1077P	10

Working primer and probe mix is prepared by mixing the above three reagents in equal volume.

PCR thermal cycling conditions

Step	Temperature (°C)	Time
Reverse transcription	50	8 min
Initial PCR activation	95	30 sec
3-step cycling (45 repeated cycles):		
- denaturation	95	1 sec
- annealing	56	20 sec
- extension	72	1 sec
Final extension	40	30 sec

5.B Real-time RT-PCR - primers/probes and PCR thermal cycling conditions

Real-time one-step RT-PCR for the detection of the hemagglutinin gene - human swine influenza virus type A (subtype H1) by 91M primer/probe set

Primers and probes

Name	Oligonucleotide sequence (5' to 3')	Direction ^a	Location ^b
H1-sw-91F	GCATAACGGGAAACTATGCAA	+	159-179
H1-sw-205R	GCTTGCTGTGGAGAGTGATTC	-	273-253
H1-sw-119P ^c	TTACCCAAATGCAATGGGGCTACCCC	-	212-187

^a +, sense; -, anti-sense.

^b Location of the oligonucleotides based on the strain of human swine influenza virus type A (subtype H1) A/California/4/2009, GenBank Accession: FJ966082.

^c The probe consisted of oligonucleotides with the 5' reporter dye FAM and the 3' quencher dye BBQ.

The concentration of the primers and probes mix:

Name	Concentration (µM)
- H1-sw-91F	10
- H1-sw-205R	10
- H1-sw-119P	10

Working primer and probe mix is prepared by mixing the above three reagents in equal volume.

PCR thermal cycling conditions

Step	Temperature (°C)	Time
Reverse transcription	50	8 min
Initial PCR activation	95	30 sec
3-step cycling (45 repeated cycles):		
- denaturation	95	1 sec
- annealing	56	20 sec
- extension	72	1 sec
Final extension	40	30 sec

5.B Real-time RT-PCR - primers/probes and PCR thermal cycling conditions

Real-time two-step RT-PCR for the detection of the hemagglutinin gene - seasonal influenza virus type A (subtypes H1 and H3)

Primers and probes

Name	Oligonucleotide sequence (5' to 3')	Direction ^a	Location ^b
H1-247F	AACATGTTACCCAGGGCATTTCGC	+	347-370
H1-361R	GTGGTTGGGCCATGAGCTTTCTTT	-	461-438
H1-278P ^c	GAGGAAGTGGGGAGCAATTGAGTTCAG	+	378-405
H3-293f F	ACCCTCAGTGTGATGGCTTCCAAA	+	266-289
H3-400R	TAAGGGAGGCATAATCCGGCACAT	-	373-350
H3-342P ^d	ACGCAGCAAAGCCTACAGCAACTGTT	+	315-340

^a +, sense; -, anti-sense.

^b Location of the oligonucleotides of subtypes H1 and H3 based on the strain of seasonal influenza virus type A (subtype H1) A/Brisbane/59/2007 and seasonal influenza virus type A (subtype H3) A/Brisbane/10/2007 respectively; GenBank Accession: CY030230 and CY039087.

^c The probe consisted of oligonucleotides with the 5' reporter dye VIC and the 3' quencher dye TAMRA.

^d The probe consisted of oligonucleotides with the 5' reporter dye FAM and the 3' quencher dye TAMRA.

The concentration of the primers and probes mix:

Name	Concentration (µM)
- H1-247F	20
- H1-361R	20
- H1-278P	10
- H3-293f F	20
- H3-400R	20
- H3-342P	10

Working primer and probe mix is prepared by mixing the above six reagents in equal volume.

PCR thermal cycling conditions

Step	Temperature (°C)	Time
Initial PCR activation	95	10 min
3-step cycling (50 repeated cycles):		
- denaturation	95	10 sec
- annealing	56	15 sec
- extension	72	10 sec
Cooling	40	30 sec

5.C Real-time RT-PCR - procedure

I. RNA extraction

Extract viral RNA from clinical specimen with QIAamp Viral RNA Mini Kit or equivalent extraction kit according to manufacturer's instructions.

II. For Real-time one-step RT-PCR

1. Prepare master mixture as below:

Reagent	Volume (µL) per reaction
- PCR grade H ₂ O ^a	7.6
- 5X Reaction buffer ^a	4
- Primers and probes mix	3
- Enzyme Blend ^a	0.4
Total volume	15

^a From Roche RealTime ready RNA Virus Master kit.

2. Add 5 µL viral RNA to the above mix.
3. Perform one-step real-time RT-PCR according to the following conditions:

Step	Temperature (°C)	Time
Reverse transcription	50	8 min
Initial PCR activation	95	30 sec
3-step cycling (45 repeated cycles):		
- denaturation	95	1 sec
- annealing	56	20 sec
- extension	72	1 sec
Final extension	40	30 sec

II. For Real-time two-step RT-PCR

Reverse transcription

1. Prepare master mixture for reverse transcription as below:

Reagent	Volume (μL) per reaction
- 10X PCR buffer I with 15 mM MgCl_2	2
- Extra 25mM MgCl_2	2.8
- 2.5 mM dNTPs	8
- Random hexamer 50 μM	1
- RNAase inhibitor 20U/ μL	1
- Reverse transcriptase 50 U/ μL	1
Total volume	15.8

2. Add 4.2 μL viral RNA to the above mix.
3. Vortex and centrifuge the tube with the mixture briefly.
4. Stand the tube at room temperature for 10 minutes and then incubate at 42 °C for at least 15 minutes.
5. Incubate the tube at 95 °C for 5 minutes and then chill in ice.

Real-time PCR

1. Performed real-time PCR on synthesized cDNA.

2. Prepare master mixture for real-time PCR as below:

Reagent	Volume (μL) per reaction
- PCR grade H_2O ^a	7.6
- MgCl_2 (25 mM) ^a	2.4
- Primers and probe mix	3
- 'Hot Start' reaction mix ^b	2
Total volume	15

^a From Roche LightCycler - FastStart DNA Master HybProbes kit.

^b Prepare 'Hot Start' reaction mix according to Roche LightCycler - FastStart DNA Master HybProbes kit's instructions.

3. Add 5 μL cDNA to the above mix.

4. Perform real-time RT-PCR according to the following conditions:

Step	Temperature (°C)	Time
Initial PCR activation	95	10 min
3-step cycling (50 repeated cycles):		
- denaturation	95	10 sec
- annealing	56	15 sec
- extension	72	10 sec
Cooling	40	30 sec

III. Interpretation of results

1. A specimen is considered positive if the C_T (cycle threshold) value is <35 . Supplementary testing using another set of primer/probe is required if the C_T falls between 35 and 40.
2. C_T value >40 does not rule out the presence of influenza virus. Results should be interpreted together with the available clinical and epidemiological information.

6. Referral for confirmation and further characterization

Specimens with PCR test results that are either (1) positive or borderline for human swine influenza virus type A (subtype H1) or (2) untypeable, that is M gene positive but negative for influenza virus type A (subtypes H1 and H3), should be forwarded to appropriate network laboratory for confirmation and characterization. Details on transport of clinical specimens are available at CHP web site⁴.

7. Quality assurance

Laboratory undertaking clinical specimen testing should have in place quality system in accordance with internationally known standards. Participation in the external quality assessment programme is highly recommended to ensure the laboratory is performing at acceptable standard.

⁴ Please see 'Safety Guidelines on Transport of Clinical Specimens and Infectious Substances for Courier Team', 25 May 2009, date last accessed:
http://www.chp.gov.hk/files/pdf/GL_Transport_of_%20specimens_EN_20041103.pdf