INFORMATION FOR US CONSULTATION ON FORMULATION OF INFLUENZA VACCINES

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1. EPIDEMIOLOGY

Influenza virus isolates for antigenic and genetic analysis are sourced from a number of countries as indicated in Table 1.1.

TABLE 1.1
Influenza Isolates Received for Analysis

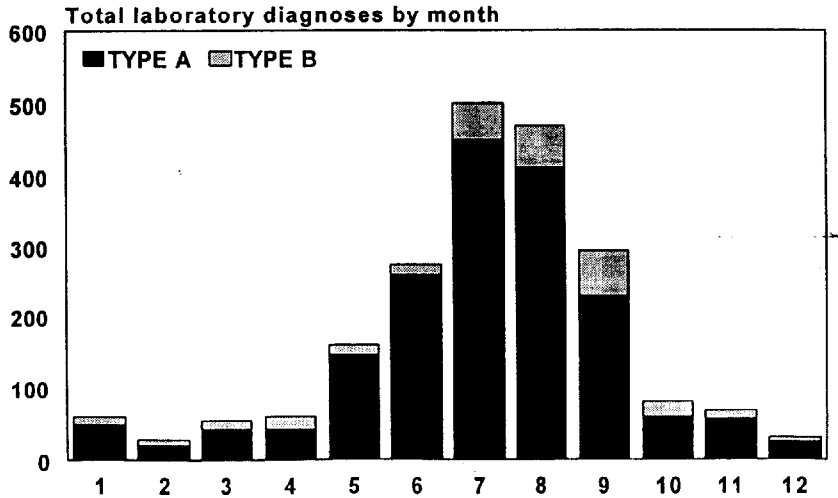
Country	January - August 1999		September 1999 - January 2000	
	Influenza A	Influenza B	Influenza A	Influenza B
Australia	631	67	195	84
Malaysia	8	2	0	0
New Caledonia	20	0	0	6
New Zealand	271	17	173	65
Philippines	9	3	13	2
Singapore	17	0	12	4
South Africa	14	18	0	0
Thailand				
TOTAL	**1026**	**143**	**401**	**193**

Epidemiological summaries for 1999 from the major sources of virus isolates are as follows.

Australia

Moderate influenza activity was experienced and consisted principally of influenza A(H3N2). Some influenza B was experienced later in the season and there were late sporadic isolates of influenza A(H1N1).

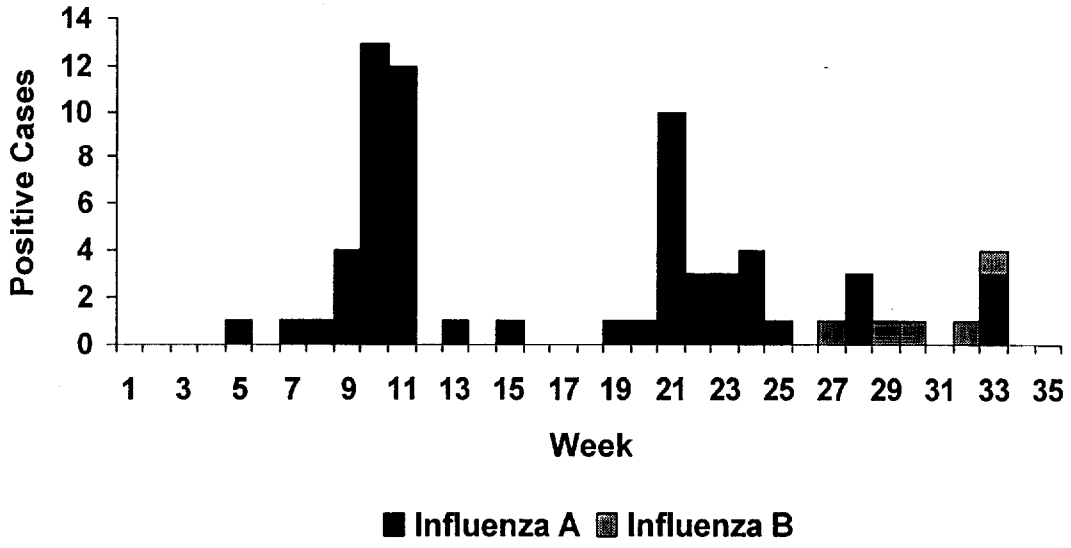
Figure 1.1
Laboratory Diagnosed Influenza – Australia 1999



- **New Caledonia**

New Caledonia experienced two influenza outbreaks, an A(H3N2) outbreak in February-March and an A(H1N1) outbreak in May-June. Later sporadic activity included cases of influenza B.

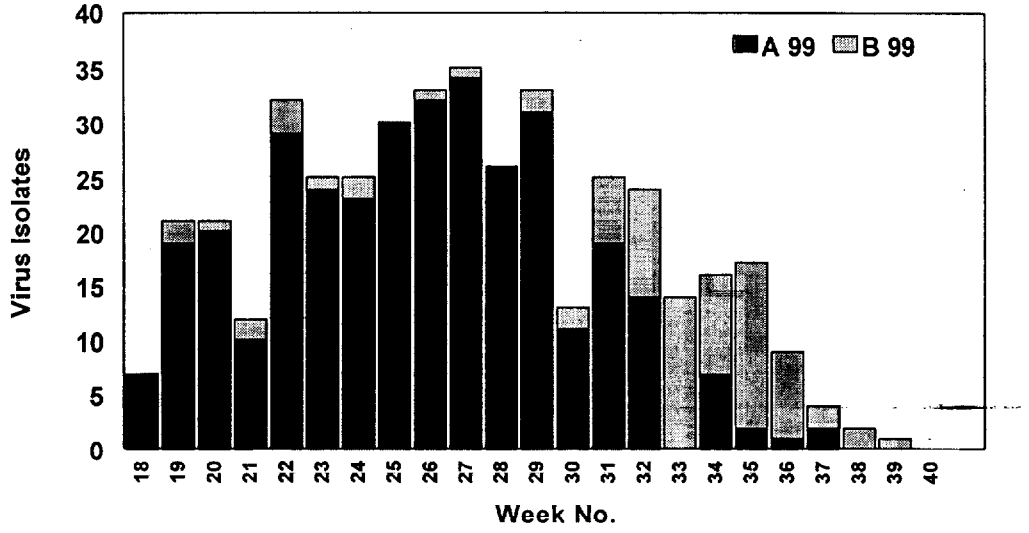
Figure 1.2
Influenza in New Caledonia 1999



New Zealand

There was moderate to widespread influenza in New Zealand consisting mainly of influenza A(H3N2) but with increasing levels of influenza B later in the season.

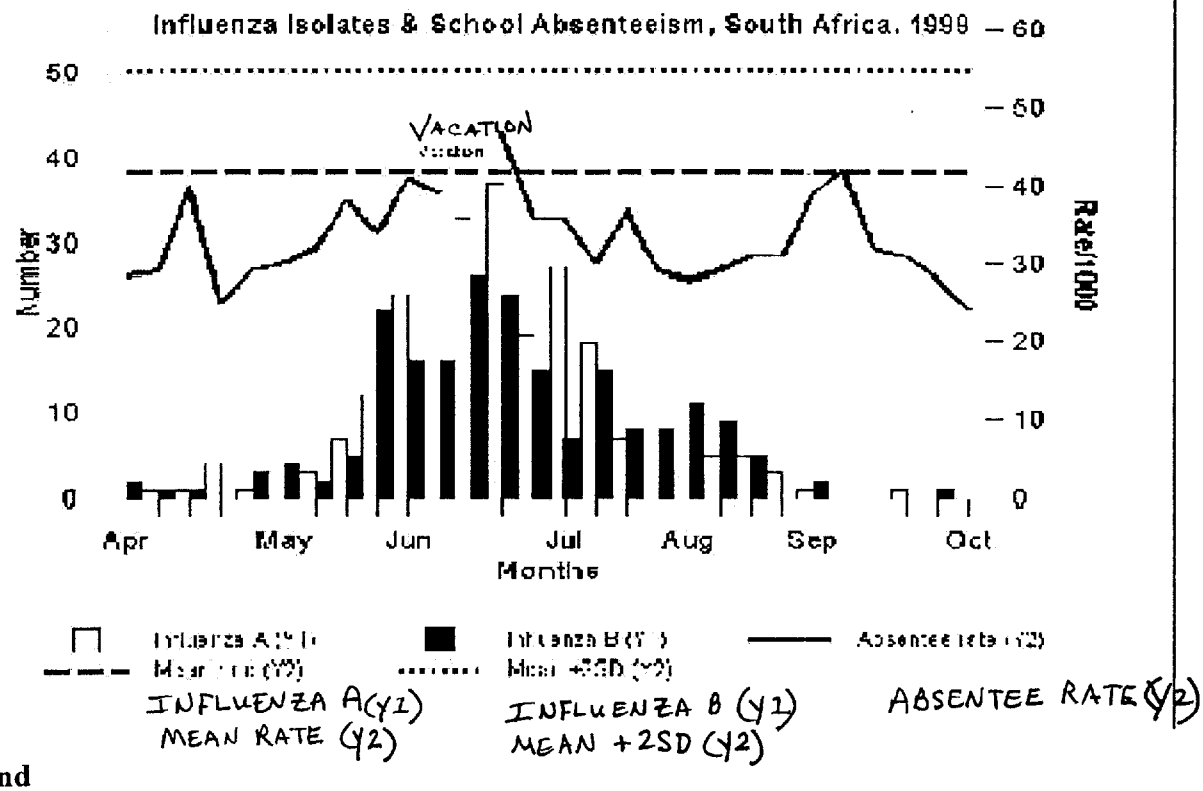
Figure 1.3
Influenza in New Zealand 1999



South Africa

Moderate influenza activity was experienced and consisted of a mixed influenza A(H3N2) and influenza B outbreak throughout the season.

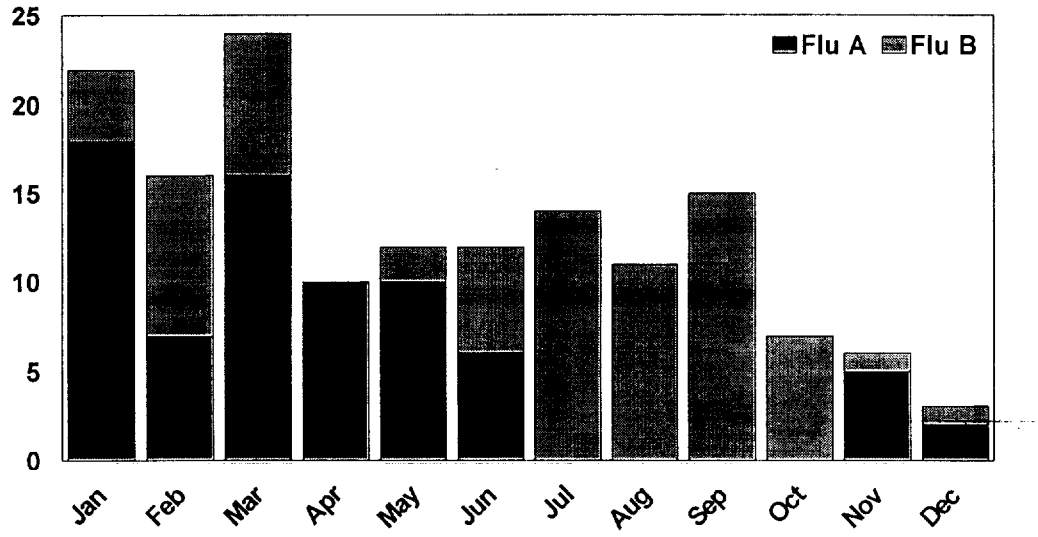
Figure 1.4



Thailand

Influenza was present throughout the year. Influenza A(H3N2) and influenza B viruses co-circulated for much of the year however only influenza B viruses were isolated between July and October. There were sporadic isolates of A(H1N1) viruses.

Figure 1.5
Influenza in Thailand 1999



2. INFLUENZA A (H1N1) STRAINS

A total of 40 isolates were analysed (Table 2.1). By antigenic and genetic analysis all isolates received were of the A/Beijing/262/95 lineage with no A/Bayern/7/95 lineage viruses. Antigenic analysis indicated that the viruses involved in the New Caledonia outbreak had undergone significant antigenic drift from the existing vaccine strain A/Beijing/262/95 (Table 2.2) and this was confirmed by sequence analysis of the HA-1 region of the viral haemagglutinins (Figure 2.1). A/New Caledonia/20/99 was selected as the representative reference strain for this new drift variant.

Sporadic isolates late in the year from Australia, New Zealand and Thailand were antigenically and genetically similar to A/New Caledonia/20/99 (Table 2.3) as were all of the influenza A viruses received late in the year from the Philippines and an isolate from Singapore (Table 2.4 and Figure 2.1).

TABLE 2.1

Summary - Characterization of Influenza A(H1N1) Isolates Received January 1999 - January 2000

Country	January - August 1999		September 1999 - January 2000	
	A/Bayern/7/95 lineage strains	A/Beijing/262/95 lineage strains	A/Bayern/7/95 lineage strains	A/Beijing/262/95 lineage strains
Australia	0	3	0	7
Malaysia	0	0	0	0
New Caledonia	0	9	0	0
New Zealand	0	1	0	1
Philippines	0	0	0	13
Singapore	0	0	0	1
South Africa	0	0	0	0
Thailand	0	4	0	1
TOTAL	**0**	**17**	**0**	**23**

TABLE 2.2
Haemagglutination-Inhibition Reactions of Influenza A(H1N1)

AUGUST 17TH 1999						
			ANTISERA			
ANTISERA NO.	F255-13D	F480-13D	F485-13D	F589-14D	F594-14D	PASSAGE HISTORY
	A	B	C	D	E	
	TEX/36	JOH/82	BEIJ/262	PHIL/8	N.CAL/20	
REF. Ag						
A. A/TEXAS/36/91	**>2560**	640	80	<20	80	SCK1,SE1,E8
B. A/JOHANNESBURG/82/96	1280	**>2560**	160	80	160	E5
C. A/BEIJING/262/95	80	80	**320**	320	320	E7
D. A/PHILIPPINES/8/99	160	160	320	**320**	320	E3
E. A/NEW CALEDONIA/20/99	<20	40	40	<20	**640**	E3
TEST Ag						
A/NEW CALEDONIA/18/99	<20	40	40	160	640	MDCKX,MDCK1
A/NEW CALEDONIA/20/99	<20	40	80	160	640	MDCKX,MDCK1
A/SYDNEY/223/99	<20	<20	<20	80	640	MDCK2
A/BANGKOK/68/99	<20	<20	40	80	640	MDCK5
A/AUCKLAND/176/99	<20	80	80	80	640	E2
A/SYDNEY/219/99	<20	<20	<20	20	640	MDCK1

TABLE 2.3
Haemagglutination-Inhibition Reactions of Influenza A(H1N1) Isolates

AUGUST 27TH 1999						
			ANTISERA			
ANTISERA NO.	F480-13D	F485-13D	F546-13D	F577-15D	F594-14D	PASSAGE HISTORY
	A	B	C	D	E	
	JOH/82	BEIJ/262	SING/9	PHIL/8	N.CAL/20	
REF. Ag						
A. A/JOHANNESBURG/82/96	**2560**	80	80	80	80	E5
B. A/BEIJING/262/95	80	**320**	320	320	320	E5
C. A/SINGAPORE/9/98	<20	20	**80**	20	80	X,MDCK4
D. A/PHILIPPINES/8/99	80	80	160	**160**	160	E3
E. A/NEW CALEDONIA/20/99	80	40	80	40	**640**	E3
TEST Ag						
A/BANGKOK/367/98	40	80	320	160	640	MDCK4
A/SYDNEY/219/99	<20	<20	40	<20	320	MDCK2
A/SYDNEY/223/99	<20	40	160	40	320	MDCK3
A/BRISBANE/108/99	20	40	160	80	640	MDCKX,MDCK1
A/SYDNEY/250/99	<20	<20	80	<20	640	MDCK1
A/BANGKOK/165/99	<20	20	160	20	640	P3,MDCK1
A/AUCKLAND/176/99	80	80	160	80	640	E3

TABLE 2.4
Haemagglutination-Inhibition Reactions of Influenza A(H1N1) Isolates

JANUARY 21ST 2000				
		ANTISERA		
ANTISERA NO.	F480-13D	F485-13D	F594-14D	PASSAGE HISTORY
	A	B	C	
	JOH/82	BEIJ/262	N.CAL/20	
REF. AG				
A. A/JOHANNESBURG/82/96	**1280**	80	80	E5
B. A/BEIJING/262/95	80	**320**	640	E5
C. A/NEW CALEDONIA/20/99	40	40	**640**	E5
TEST Ag				
A/WISCONSIN/6/99	20	20	320	C1C2MDCK1
A/SINGAPORE/78/99	40	40	640	MDCK3
A/PHILIPPINES/323/99	40	40	640	MDCKX,MDCK1
A/PHILIPPINES/320/99	80	80	1280	MDCKX,MDCK1

FIGURE 2.1
Phylogenetic comparisons of Influenza A(H1N1) Haemagglutinins

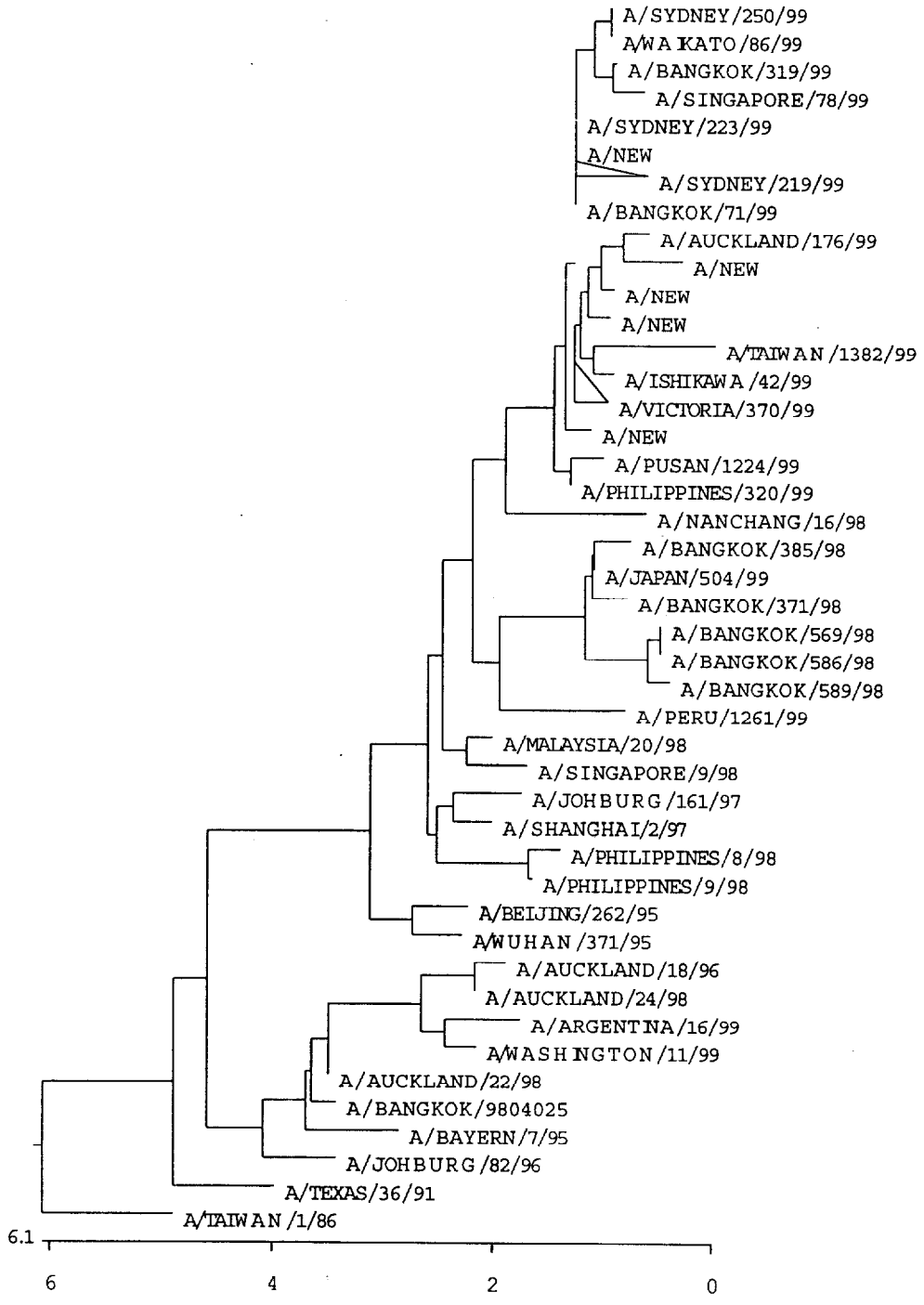


TABLE 2.5
Influenza H1 Amino Acid Differences
Comparison of A/Beijing/262/95 HA Sequence and 1999 Isolates

Isolate	Amino Acid Position																				
	10	11	14	35	40	69	72	128	132	141	152	162	167	169	182	188	190	220	270	297	309
A/Beijijng/262/95	**N**	**N**	**D**	**D**	**K**	**S**	**S**	**V**	**T**	**S**	**E**	**N**	**N**	**E**	**S**	**R**	**I**	**G**	**N**	**V**	**T**
A/Malaysia/20/98	-	-	-	-	-	-	-	-	S	-	K	-	-	-	-	-	-	D	D	-	A
A/Philippines/8/98 e	-	-	-	-	-	-	-	-	S	-	-	-	-	K	-	K	-	-	D	I	A
A/Singapore/9/98	-	-	-	-	-	-	F	-	S	-	K	-	D	-	-	-	-	D	D	-	A
A/Bangkok/71/99	-	-	-	-	-	L	-	-	S	-	G	K	-	-	P	-	L	D	D	-	-
A/New Caledonia/18/99 e	-	-	-	-	-	L	-	-	S	-	G	K	-	-	P	-	L	D	D	-	A
A/New Caledonia/19/99	-	H	-	-	-	L	-	-	S	-	G	K	-	-	P	-	L	D	D	-	-
A/New Caledonia/20/99 e	-	-	-	-	-	L	-	-	S	-	G	K	-	-	P	-	L	D	D	-	A
A/Sydney/219/99	-	-	-	-	-	L	-	I	S	R	G	K	-	-	P	-	L	D	D	-	A
A/Sydney/223/99	-	-	-	-	-	L	-	-	S	-	G	K	-	-	P	-	L	D	D	-	A
A/Auckland/176/99 e	-	-	-	N	-	L	-	-	S	-	G	K	-	-	P	-	L	D	D	-	A

3. INFLUENZA A (H3N2) STRAINS

A total of 1206 viruses were analysed (Table 3.1). All strains were classified as A/Sydney/5/97-like viruses by antigenic and genetic characterization, however:

- a substantial proportion of isolates showed a lowered reaction (4 fold or more) with A/Sydney antisera and antisera to other recent strains (classified as low-reactors)

- approximately a quarter of all isolates reacted more strongly with antiserum to the recent isolate A/Moscow/10/99 than with A/Sydney antiserum.

Table 3.2 shows a range of reactivities observed with isolates including occasional very low reacting strains (A/Waikato/21/99). Table 3.3 shows a recent assay comparing US isolates and Australia-Pacific isolates and includes antisera to the A/Panama strains and the higher growth reassortants Nib-41 and Nib-43. Table 3.4 compares a number of viruses received recently from Singapore with UK isolates.

Differentiation into the three groups was not clear cut and their relative proportions were similar early and late in the season (Table 3.1). There were no consistent sequence changes in the haemagglutinins which correlated with the reactivity of viruses in the HI test and no clear line of genetic drift (Figure 3.1 and Table 3.5).

Our studies have suggested a number of potential reasons for the high level of A/Sydney low reacting strains seen at the Australian Centre:

- Differing specificities of the A/Sydney antisera in use at CDC and in Australia (the Australian sera appear more discriminating) – data not shown.

- Where viruses were isolated into both eggs and cell culture the egg viruses generally showed a reactivity profile more typical of A/Sydney with the antiserum panel (ie. a possible artefact from the cell culture system) – Table 3.6.

- Electron microscope studies on a small number of samples suggested that the low-reacting strains were highly aggregated.

TABLE 3.1

Summary - Characterization of Influenza A(H3N2) Isolates Received January 1999 - January 2000

Country	January - August 1999			September 1999 - January 2000		
	A/Sydney/5/97 lineage strains	A/Sydney/5/97 low-reacting strains*	A/Moscow/10/99 lineage strains	A/Sydney/5/97 lineage strains	A/Sydney/5/97 low-reacting strains*	A/Moscow/10/99 lineage strains
Australia	196	183	97	71	73	63
Malaysia	3	3	0	0	0	0
New Caledonia	2	3	5	0	0	0
New Zealand	93	76	59	86	51	44
Philippines	2	3	0	0	1	1
Singapore	2	8	3	0	4	1
South Africa	8	2	3	0	0	0
Thailand	54	0	2	2	0	2
TOTAL	**360**	**278**	**169**	**159**	**129**	**111**

* Isolates showing a 4-fold or more reduction in reaction with A/Sydney/5/97 antiserum and no reaction with a monoclonal antibody Mab-105.

TABLE 3.2
Haemagglutination-Inhibition Reactions of Influenza A(H3N2) Isolates

AUGUST 31ST 1999				ANTISERA				
ANTISERA NO.	F535/14D	F526/13D	F536/15D	F570/15D	F578/15D	F576/15D	F579/14D	PASSAGE HISTORY
	A	B	C	D	E	F	G	
REF. Ag	NAN/933	SYD/5	DAR/1	SICH/346	CNIC/149	MOSC/10	DAR/5	
A A/NANCHANG/933/95	**1280**	160	320	320	160	160	320	EXE4
B A/SYDNEY/5/97	160	**1280**	>2560	1280	1280	1280	640	CEK2E4
C A/DARWIN/1/98	160	640	**640**	640	320	640	320	MDCK7
D A/SICHUAN/346/98	80	160	160	**640**	80	80	160	E7
E A/CNIC/149/98	160	320	320	320	**320**	640	640	C1,C1,MDCK3
F A/MOSCOW/10/99	160	640	640	320	640	**>2560**	320	E7
G A/DARWIN/5/99	320	640	1280	>2560	1280	320	**>2560**	E4
TEST Ag.								
A/WELLINGTON/22/99	20	80	80	160	80	160	160	MEKX,MDCK4
A/WELLINGTON/35/99	160	640	640	640	640	1280	640	MEKX,MDCK2
A/WELLINGTON/42/99	640	1280	640	1280	1280	640	10240	MEKX,MDCK2
A/WAIKATO/15/99	80	160	320	320	160	320	160	MEKX,MDCK3
A/WAIKATO/21/99	<20	<20	40	40	<20	80	80	MDCKX,MDCK3
A/WAIKATO/23/99	20	80	80	80	80	160	160	MEKX,MDCK2
A/WAIKATO/34/99	160	320	320	320	160	320	320	MDCKX,MDCK1
A/TOWNSVILLE/9/99	40	160	160	160	160	320	320	MDCK2
A/TOWNSVILLE/10/99	20	80	80	80	80	160	160	MDCK2
A/TOWNSVILLE/11/99	20	160	160	160	160	320	320	MDCK2
A/BRISBANE/85/99	20	80	80	80	80	160	160	MDCKX,MDCK1
A/CHRISTCHURCH/28/99	80	320	320	320	320	320	320	MDCK5
A/CHRISTCHURCH/42/99	160	640	1280	640	1280	1280	640	MDCK4
A/VICTORIA/377/99	80	320	640	320	640	640	320	MDCK1
A/VICTORIA/382/99	160	1280	1280	320	>2560	1280	640	MDCK1
A/VICTORIA/388/99	40	1280	1280	1280	>2560	>2560	640	MDCK1
A/VICTORIA/361/99	80	640	1280	320	320	640	320	MDCK1
A/VICTORIA/385/99	40	160	160	160	160	320	320	MDCKX,MDCK1
A/VICTORIA/522/99	20	80	80	80	80	160	160	MDCKX,MDCK1
A/SYDNEY/256/99	160	320	640	640	320	640	320	MDCK2
A/SYDNEY/258/99	640	>2560	>2560	>2560	>2560	>2560	>2560	MDCK2

TABLE 3.3
Haemagglutination-Inhibition Reactions of Influenza A(H3N2) Isolates

JANUARY 14TH 2000

	ANTISERA NO.	F526/13D	F514/13D IVR108	F536/15D	F576/15D	F579/14D	F608/18D	F620/13D	F618/13D NIB41	F619/13D NIB43	PASSAGE HISTORY
						ANTISERA					
		A	B	C	D	E	F	G	H	I	
		SYD/5	SYD/5	DAR/1	MOSC/10	DAR/5	VIC/358	PAN/2007	PAN/2007	PAN/2007	
A	A/SYDNEY/5/97	**1280**	1280	1280	640	320	640	640	640	640	CEK2E4
B	A/SYDNEY5/97 IVR 108	640	**1280**	1280	640	160	320	640	320	640	
C	A/DARWIN/1/98	640	640	**640**	640	320	640	640	320	160	MDCK7
D	A/MOSCOW/10/99	640	640	640	**>2560**	160	>2560	640	320	640	E9
E	A/DARWIN/5/99	320	640	640	320	**640**	640	640	320	320	E5
F	A/VICTORIA/358/99	320	320	160	1280	<20	**1280**	320	320	320	E4
G	A/PANAMA/2007/99	160	160	160	160	80	320	**640**	640	320	E6
H	A/PANAMA/2007/99 NIB 41	1280	640	320	640	320	640	1280	**1280**	1280	
I	A/PANAMA/2007/99 NIB 43	1280	1280	640	1280	320	640	1280	1280	**1280**	
	ANTIGENS										
	A/NEW HAMPSHIRE/4/99	160	160	160	160	40	80	80	80	80	MX,C2,MDCK1
	A/ALASKA/25/99	160	160	160	160	80	160	160	160	160	M1/C2/MDCK1
	A/ALASKA/27/99	160	160	160	160	80	160	160	160	160	M1/C2/MDCK1
	A/NEW YORK/94/99	80	80	80	160	40	80	160	80	160	MX,C1/MDCK1
	A/NORTH CAROLINA/8/99	160	160	160	160	160	160	160	160	80	M1/C1/MDCK1
	A/WASHINGTON/17/99	160	80	80	160	80	80	160	80	80	M1/C1/MDCK1
	A/SINGAPORE/96/99	80	40	80	160	160	160	160	160	80	MDCK3
	A/TENNESSEE/3/99	160	160	160	160	80	160	160	160	320	X2/C1/MDCK1
	A/MASSACHUSETTS/7/99	160	80	80	160	80	160	160	80	80	X1/C1/MDCK1
	A/SOUTH AUSTRALIA/115/99	160	160	160	320	320	320	320	160	160	MDCK3
	A/SOUTH AUSTRALIA/132/99	80	80	80	160	160	160	160	80	80	MDCKX,MDCK1
	A/SOUTH AUSTRALIA/147/99	160	80	40	160	160	160	160	80	80	MDCKX,MDCK2
	A/SOUTH AUSTRALIA/22/99	80	40	40	80	80	80	80	80	40	MDCK2
	A/TOWNSVILLE/19/99	80	40	40	80	80	80	80	80	40	MDCK3
	A/PHILIPPINES/22/99	160	160	160	160	80	320	320	160	320	E7
	A/PHILIPPINES/26/99	160	80	80	80	20	160	160	160	160	E4
	A/VICTORIA/614/99	80	80	80	160	160	160	160	80	80	X,MDCK2
	A/SINGAPORE/94/99	40	40	40	80	80	80	80	40	20	MDCK3

*IVR-108 is a high-growth reassortant of A/Sydney/5/97

TABLE 3.4
Haemagglutination-Inhibition Reactions of Influenza A(H3N2) Isolates

	JANUARY 22ND 2000				ANTISERA				
	ANTISERA NO.	F535/14D	F527/13D	F570/15D	F578/15D	F576/15D	F579/14D	F608/18D	PASSAGE HISTORY
		A	B	C	D	E	F	G	
	REF. Ag	NAN/933	SYD/5	SICH/346	CNIC/149	MOSC/10	DAR/5	VIC/358	
A	A/NANCHANG/933/95	**640**	80	160	<20	80	40	160	E4
B	A/SYDNEY/5/97	80	**320**	320	320	160	160	160	CEK2E4
C	A/SICHUAN/346/98	40	80	**640**	80	40	80	80	E7
D	A/CNIC/149/99	40	320	320	**640**	640	320	320	C1,C2,MDCK3
E	A/MOSCOW/10/99	80	160	160	320	**1280**	80	640	E9
F	A/DARWIN/5/98	40	80	320	160	80	**320**	160	E5
G	A/VICTORIA/358/99	80	160	80	160	640	<20	**640**	E4
	TEST Ag								
	A/ENGLAND/635/99	40	80	160	160	320	80	320	C1C2MDCK1
	A/ENGLAND/648/99	640	1280	640	>2560	1280	640	640	X,MDCK1
	A/ENGLAND/651/99	40	80	160	320	320	160	160	X,MDCK2
	A/SINGAPORE/61/99	40	80	160	160	160	160	160	MDCK3
	A/SINGAPORE/69/99	20	40	80	80	160	160	160	MDCK3
	A/SINGAPORE/59/99	40	40	80	160	160	160	160	MDCK5
	A/SINGAPORE/68/99	40	160	320	640	320	320	320	MDCK3
	A/SINGAPORE/62/99	40	80	160	160	160	160	160	MDCK3
	A/SINGAPORE/67/99	20	40	80	80	80	40	80	MDCK3
	A/SOUTH AUSTRALIA/155/99	40	160	320	640	320	320	320	MDCKX,MDCK2

TABLE 3.5

Influenza H3 Amino Acid Differences

Comparison of A/Sydney/5/97 and 1999 Isolates in date order of isolation

AA Number	Sydney/5/97	Townsville/1/99	Singapore/2/99	Singapore/14/99	Singapore/45/99	Darwin/1/98	Sydney/3/99	Victoria/1/99	New CaleCaledonia/4/99	New Caledonia/5/99 e	New Caledonia/9/99 e	Darwin/5/99	New Caledonia/11/99	Victoria/3/99	Victoria/3/99 e	Sydney/208/99	Auckland/218/99	Auckland/28/99	Townsville/2/99	Victoria/516/99	Auckland/16/99	Townsville/4/99	Bangkok/135/99	Brisbane/28/99	Bangkok/146/99	Victoria/522/99	Auckland/80/99	Victoria/314/99	Brisbane/33/99	Sth Australia/50/99	Auckland/139/99	Johannesburg/72/99	Victoria/527/99	Auckland/167/99	Victoria/536/99	Victoria/536/99	Victoria/538/99	Victoria/343/99	Sydney/220/99	Victoria/546/99	Victoria/552/99	Victoria/358/99	Victoria/559/99	Victoria/567/99	Victoria/578/99
Reactivity n=normal l=low		N	VL	N	VL	N	N	VL	L	N	N	L	N	L	N	N	N	L	L	N	L	N	N	L	N	N	L	N	N	N	N	N	L	N	L	L	L	L	L	L	L	N	L	L	L
Clinical Spec. Date		10/01/1999	12/01/1999	21/01/1999	3/02/1999	22/02/1999	24/02/1999	25/02/1999	8/03/1999	10/03/1999	10/03/1999	20/03/1999	17/04/1999	21/04/1999	21/04/1999	27/04/1999	3/05/1999	6/05/1999	6/05/1999	6/05/1999	14/05/1999	14/05/1999	24/05/1999	31/05/1999	1/06/1999	8/06/1999	9/06/1999	9/06/1999	16/06/1999	16/06/1999	18/06/1999	18/06/1999	24/06/1999	30/06/1999	2/07/1999	2/07/1999	3/07/1999	5/07/1999	6/07/1999	9/07/1999	12/07/1999	17/07/1999	17/07/1999	21/07/1999	26/07/1999
2																								N													N				N				R
3	I	L	L		L	L	L	L	L	L	L	L	L	L	L	L	L	L	L		L	L	L		L	L	L	-	L	L	L	L	L	L	L	L		L	L	L	L	L	L	L	L
5	G																V			E																									
21	P													L	L																														
24	T																																												
31	N	S																																											
33	Q																H																												
45	S													N	N																														
48	T																																												A
53	D									N														N		N		N											N	N			N		N
55	P								H		H		H																								H								
57	R		Q		Q		Q	Q	Q	Q	Q		Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
65	T			A																																									
92	K																T																												
94	Y																																												
103	P																																												
106	A																	V																											
121	N																										D																		
124	S						N										I																												
133	N																										D																		
137	Y	F	S	N	S		S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
140	K									E																																			
142	S	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R
144	I																				N		N		N		N			N	N			N	S	S					N				
145	K																																												
156	Q																																												
157	L																																		S	S									
158	K																																												
160	K		R		R		R	R	R		R		R			R		R		R				R								R	R		R	R		R							
167	T																																									A			
172	D									E				E	E		E		E		E	E	E	E	E	E	E	E	E	E			E					E	E	E	E	E	E	E	E
176	D																													N																
182	V									F																																			
183	H																																												
185	P																						S																						
186	S																																												
189	S																																	N											
192	T									I							I		I		I	I	I	I	I	I	I	I	I	I		I	I				I	I	I	I	I	I	I	I
193	S																																												
194	I	L	L	L	L	L	L	L	L	L		L	L	L		L	L	L	L	L	L	L	L	L	L	L	L	L	L	L		L	L	L	L	L	L	L	L	L	L	L	L	L	L
196	A																																						T						
206	T																										S																		
207	K			Q																																									
218	G																																												
219	S																																												
220	R										G																					S													
224	R																																												
226	I	V	V	V	V	V	V	V	V		V	V	V	V	V	V		V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V
229	R			I	I										I																	I													
233	H	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y		Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y
246	N																																						K						
268	M			I																																									
269	R	K																																											
272	D																N																												
274	I																													S	S			S												
279	S												Y																																
291	D																																												

FIGURE 3.1
Phylogenetic Relationships of Influenza A(H3N2) Haemagglutinins

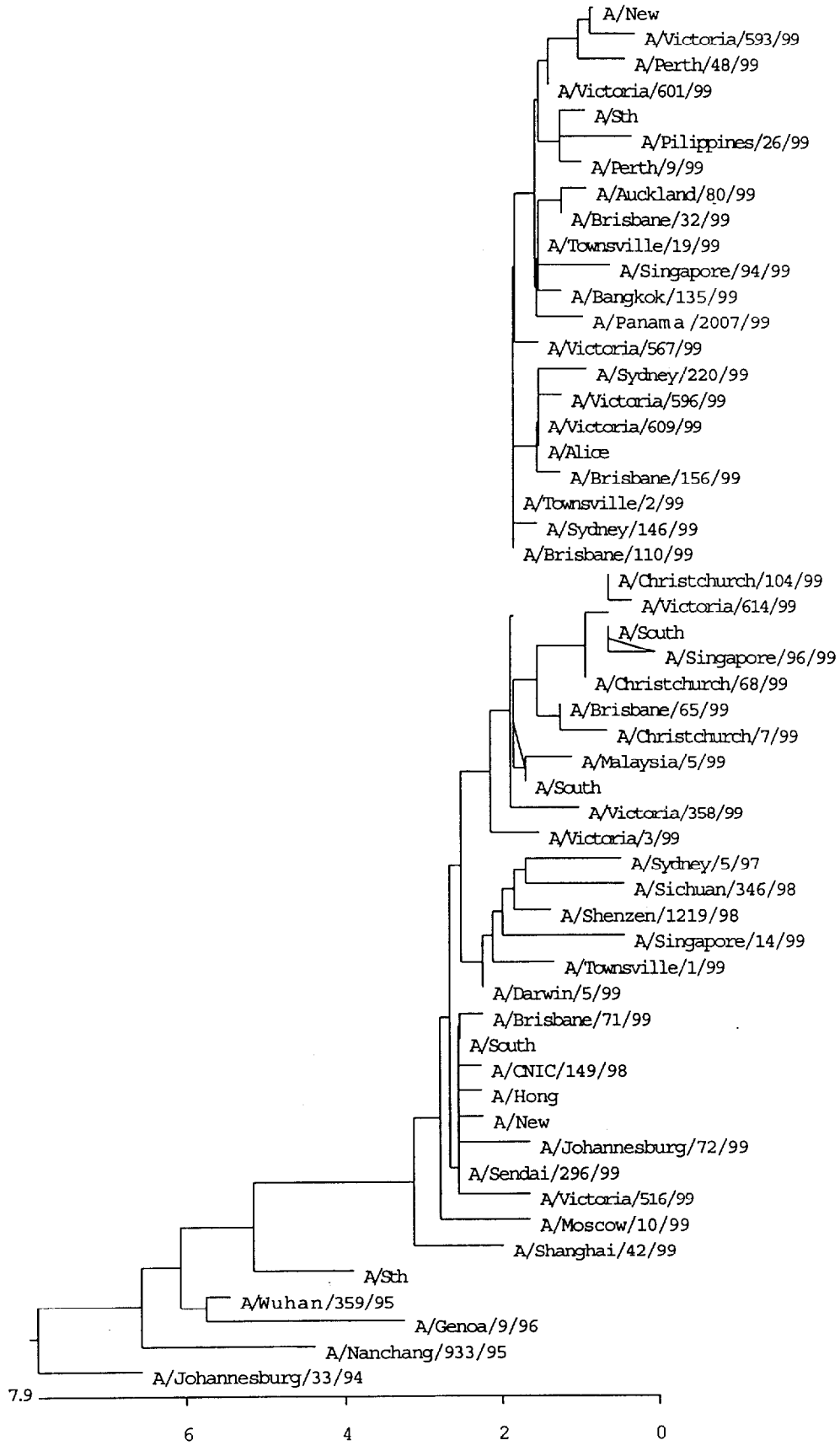


TABLE 3.6
HAI Comparison of H3 Cell-Egg Pairs

SEPTEMBER 14TH 1999	ANTISERA								
ANTISERA NO.	F535/14D	F526/13D	F578/15D	F576/15D	F579/14D	F597-14D	F598-14D	AS 363	PASSAGE HISTORY
ANTIGENS	NAN/933	SYD/5	CNIC/149	MOSC/10	DAR/5	VIC/577	BRIS/65	SYD/5	
A/SYDNEY/5/97	320	1280	>2560	>2560	640	640	640	>2560	CEK2,E4
A/SYDNEY/5/97	160	320	320	320	320	320	320	320	X,MDCK4
A/AUCKLAND/28/99	320	320	1280	320	1280	320	1280	>2560	E3
A/AUCKLAND/28/99	40	80	160	320	320	160	160	<20	MDCKX,MDCK1
A/BRISBANE/64/99	80	160	320	320	320	640	320	>2560	E4
A/BRISBANE/64/99	40	80	160	320	320	160	160	40	MDCKX,MDCK1
A/BRISBANE/65/99	160	320	640	640	640	640	640	>2560	E2
A/BRISBANE/65/99	40	80	80	160	160	80	160	<20	MDCKX,MDCK1
A/DARWIN/5/99	320	320	1280	320	1280	320	640	>2560	E4
A/DARWIN/5/99	40	80	160	320	320	160	160	<20	MDCK2
A/MALAYSIA/7/99	320	1280	>2560	>2560	640	640	640	>2560	E3
A/MALAYSIA/7/99	80	320	640	640	640	320	320	640	X,MDCK1
A/VICTORIA/3/99	320	1280	1280	1280	1280	640	1280	>2560	E4
A/VICTORIA/3/99	40	80	40	160	160	160	160	20	MDCK3
A/VICTORIA/522/99	320	320	640	320	1280	320	640	>2560	E4
A/VICTORIA/522/99	40	80	80	160	160	80	80	<20	MDCKX,MDCK1
A/VICTORIA/553/99	160	320	640	1280	320	320	640	>2560	CEK2,E2
A/VICTORIA/553/99	80	320	320	320	160	320	320	>2560	MDCKX,MDCK1
A/VICTORIA/546/99	160	320	320	>2560	160	320	320	>2560	E4
A/VICTORIA/546/99	160	320	640	640	320	320	320	640	MDCKX,MDCK1
A/VICTORIA/577/99	320	640	1280	>2560	640	1280	640	>2560	E2
A/VICTORIA/577/99	40	80	80	160	160	80	80	<20	MDCKX,MDCK1
A/VICTORIA/585/99	160	1280	>2560	1280	640	640	640	>2560	E2
A/VICTORIA/585/99	40	160	160	320	320	160	320	20	MDCKX,MDCK1
A/VICTORIA/588/99	80	320	640	640	320	640	640	>2560	E2
A/VICTORIA/588/99	40	80	80	160	160	80	160	<20	MDCKX,MDCK1

4. INFLUENZA B STRAINS

A total of 293 isolates were analysed. By antigenic and genetic analysis 282 (96%) of these were B/Beijing/184/93 lineage viruses with 11 isolates of B/Shangdong/7/97 lineage viruses from Thailand, all of these isolated in the January-March 1999 period (Table 4.1). The majority of the B/Beijing/184/93 lineage viruses were strongly inhibited by antisera to B/Beijing/184/93 and to the current vaccine strain B/Yamanashi/166/97. Occasional strains showed a 4-fold reduction in reaction with B/Yamanashi antiserum – these viruses were more strongly inhibited by antiserum to the recent isolate B/Johannesburg/5/99 (Tables 4.2 and 4.3).

Sequence analysis of the viral haemagglutinin indicates ongoing genetic drift away from the B/Yamanashi/166/95 vaccine strain (Figure 4.1 and Table 4.5).

The few B/Shangdong/7/97-lineage viruses were antigenically and genetically close to B/Shangdong/7/97 and the earlier reference strain B/Beijing/243/97 (Table 4.4).

TABLE 4.1

Summary - Characterization of Influenza B Isolates Received January 1999 - January 2000

Country	January - August 1999		September 1999 - January 2000	
	B/Beijing/184/93 lineage strains	B/Shangdong/7/97 lineage strains	B/Beijing/184/93 lineage strains	B/Shangdong/7/97 lineage strains
Australia	35	0	95	0
Malaysia	2	0	0	0
New Caledonia	0	0	2	0
New Zealand	14	0	59	0
Philippines	3	0	0	0
Singapore	2	0	0	0
South Africa	11	0	6	0
Thailand	21	11	32	0
TOTAL	**88**	**11**	**194**	**0**

TABLE 4.2
Haemagglutination-Inhibition Reactions of Influenza B Isolates

October 29th 1999									
					ANTISERA				
ANTISERA NO.	F488/24D	F551-25D	F433/27D	F429/27D	F428/27D	F575/27D	F604-26D	F585-29D	PASSAGE HISTORY
	A	B	C	D	E	F	G	H	
	BEIJ/243	SHAN/7	HAR/7	BEIJ/184	IND/1	YAM/166	STHA./5	JOH/5	
REF. Ag									
A. B/BEIJING/243/97	**640**	640	80	80	40	40	80	<20	E6
B. B/SHANGDONG/7/97	640	**1280**	80	80	40	40	80	<20	E9
C. B/HARBIN/7/94	20	80	**640**	640	80	320	320	640	EX VW2297,E1
D. B/BEIJING/184/93	10	40	320	**320**	160	320	320	320	EXVW2955,E1
E. B/INDIANA/1/95	20	40	320	320	**640**	320	320	320	X,C3,MDCK3
F. B/YAMANASH/166/98	20	40	320	320	80	**320**	320	640	E9
G. B/SOUTH AUSTRALIA/5/99	20	40	320	320	160	320	640	640	E4
H. B/JOHANNESBURG/5/99	40	40	320	320	160	320	640	640	E7
TEST Ag									
B//WAIKATO/8/99	80	160	640	640	640	320	1280	1280	MDCKX,MDCK1
B/WAIKATO/6/99	40	80	320	320	320	160	640	640	MDCKX,MDCK1
B/WAIKATO/5/99	40	80	320	320	320	320	640	640	MDCKX,MDCK1
B/WELLINGTON/4/99	40	40	320	320	320	160	640	640	MDCKX,MDCK1
B/BRISBANE/28/99	80	80	320	320	320	160	640	640	MDCKX,MDCK1
B/BRISBANE/2999	40	80	320	320	320	320	320	320	MDCKX,MDCK1
B/BRISBANE/30/99	40	80	320	320	320	160	640	640	MDCKX,MDCK1
B/PERTH/6/99	20	40	160	160	160	80	320	320	MDCKX,MDCK1
B/PERTH/7/99	20	40	160	320	80	160	320	320	MDCKX,MDCK1
B/CHRISTCHURCH/34/99	40	80	320	640	320	640	640	640	MDCK3
A/PERTH/60/99	40	80	320	320	160	320	640	640	MDCK1
B/CANBERRA/1/99	20	80	320	320	320	160	320	320	MKX,MDCK1

TABLE 4.3
Haemagglutination-Inhibition Reactions of Influenza B Isolates

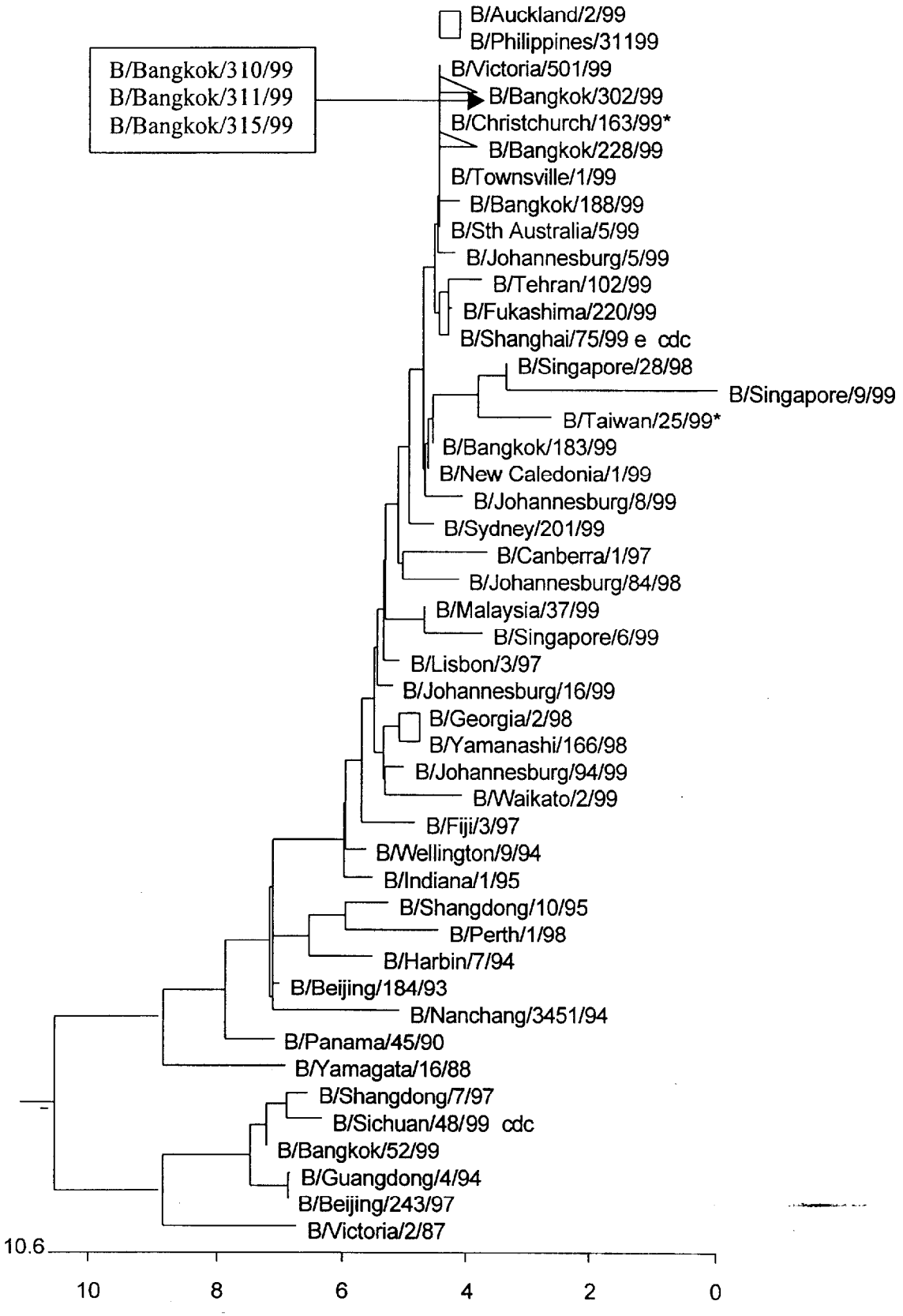
13TH DECEMBER 1999									
					ANTISERA				
	ANTISERA NO.	F488/24D	F551-25D	F433/27D	F429/27D	F428/27D	F575/27D	585/28D	PASSAGE HISTORY
		A	B	C	D	E	F	G	
		BEIJ/243	SHAN/7	HAR/7	BEIJ/184	IND/1	YAM/166	JOH/5	
	REF. Ag								
A.	B/BEIJING/243/97	**1280**	1280	80	80	80	40	160	E6
B.	B/SHANGDONG/7/97	1280	**1280**	<20	40	<20	<20	80	E9
C.	B/HARBIN/7/94	20	40	**320**	640	80	320	320	VW2297+E1
D.	B/BEIJING/184/93	20	40	160	**320**	160	320	640	E10
E.	B/INDIANA/1/95	40	80	320	640	**640**	320	640	X,C3,MDCK3
F.	B/YAMANASH/166/98	20	40	640	640	320	**640**	640	E9
G.	B/JOHANNESBURG/5/99	20	40	160	320	160	320	**640**	E6
	TEST Ag								
	B/BANGKOK/303/99	40	40	160	320	320	160	640	MDCK3
	B/BANGKOK/205/99	80	80	320	640	320	320	640	MDCK3
	B/BANGKOK/206/99	40	40	320	320	320	320	640	MDCK3
	B/BANGKOK/212/99	40	40	320	320	320	320	320	MDCK4
	B/BANGKOK/222/99	40	40	320	320	320	320	1280	MDCK4
	B/BANGKOK/223/99	40	40	160	320	320	160	640	MDCK3
	B/BANGKOK/229/99	80	80	320	320	320	160	640	MDCK3
	B/BANGKOK/269/99	40	40	80	80	80	80	80	MDCK4
	B/BANGKOK/270/99	40	40	320	320	320	320	640	MDCK4
	B/BANGKOK/280/99	40	40	320	320	320	320	640	MDCK4
	B/BANGKOK/282/99	40	40	320	320	320	160	320	MDCK3
	B/BANGKOK/288/99	40	40	160	160	160	160	640	MDCK4
	B/BANGKOK/290/99	40	40	160	320	320	160	640	MDCK4
	B/BANGKOK/301/99	40	40	160	320	320	320	640	MDCK4
	B/BANGKOK/302/99	20	20	160	160	160	160	640	MDCK4
	B/BANGKOK/310/99	40	40	320	320	320	320	640	MDCK4
	B/BANGKOK/311/99	20	20	160	160	160	160	640	MDCK4
	B/TASMANIA/1/99	80	80	320	320	320	320	640	MDCK1
	B/BANGKOK/323/99	80	80	320	640	640	320	640	MDCK4
	B/BANGKOK/309/99	40	40	320	320	160	160	640	MDCK4
	B/VICTORIA/512/99	80	80	320	320	320	320	640	MDCKX,MDCK1
	B/VICTORIA/511/99	40	40	160	320	320	160	640	MDCKX,MDCK1
	A/CHRISTCHURCH/163/99	40	40	160	320	320	160	640	MDCK2

B/CHRISTCHURCH/270/99	80	80	320	320	320	160	640	MDCK3

TABLE 4.4
Haemagglutinin-Inhibition Reactions of Influenza B Isolates

APRIL 28TH 1999	ANTISERA							
ANTISERA No.	F348/26D	F488/24D	F551-25D	F433/27D	F428/27D	F489/28D	LOT98-65	PASSAGE HISTORY
	A	B	C	D	E	F	G	
REF. Ag	GUA/4	BEIJ/243	SHAN/7	HAR/7	IND/1	LISB/3	YAM/166	
A. B/GUANGDONG/4/94	**320**	320	160	40	40	40	40	E8
B. B/BEIJING/243/95	320	**640**	320	40	40	40	40	E6
C. B/SHANGDONG/7/97	320	640	**640**	20	20	20	20	E8
D. B/HARBIN/7/94	<20	20	20	**160**	80	160	320	E13
E. B/INDIANA/1/95	40	80	20	320	**320**	320	320	X1/C3/MDCK4
F. B/LISBON/3/96	20	20	20	320	80	**320**	320	E8
G. B/YAMANASH/166/98	<20	20	20	320	160	320	**640**	E8
TEST Ag								
B/BANGKOK/33/99	80	160	160	40	40	40	40	MDCK4
B/BANGKOK/51/99	640	640	320	20	20	20	40	MDCK4
B/BANGKOK/50/99	320	320	160	80	80	80	80	MDCK3
B/PHILIPPINES/1/99	40	40	20	320	320	320	320	MDCKX,MDCK4
B/TEHRAN/102/99	<20	20	20	320	80	320	320	EXE2
B/BRISBANE/2/99	20	40	20	320	320	320	320	MDCKX,MDCK1

FIGURE 4.1
Phylogenetic Relationships of Influenza B Haemagglutinins



5. SEROLOGY

Antibody assays were performed using panels of pre- and post-immunization sera from young adults and older adults immunised in Australia, Europe and USA during 1999. In all cases vaccinees received a single dose of vaccine containing:

an A/Beijing/262/95 (H1N1)-like strain
an A/Sydney/5/97 (H3N2)-like strain
a B/Beijing/184/93-like strain

The actual B/Beijing/184/93-like strains were:

- For Australia – B/Harbin/7/94
- For Europe and USA – B/Yamanashi/166/99

Results of assays performed in September 1999 and January 2000 are shown.

TABLE 5.1

Haemagglutination Inhibition Antibody Responses
Influenza Type A(H1N1) Vaccine Component
Young Adults – September 1999

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Young Adult 9/99	30	A/BEIJING/262/95	E7	93	14.8	206.2	17	93	0	67
		A/JOHANNESBURG/82/96	E5	83	42.9	265.9	67	100	3	73
		A/NEW CALEDONIA/20/99	E4	87	7.2	72.9	3	70	0	33
		A/NANCHANG/16/99	EXE4	83	8.1	78.2	7	8	0	40
American Young Adult 9/99	32	A/BEIJING/262/95	E7	42	20.6	67.3	38	88	4	29
		A/JOHANNESBURG/82/96	E5	25	46.2	100.8	75	88	25	46
		A/NEW CALEDONIA/20/99	E4	42	17.8	46.2	38	71	8	13
		A/NANCHANG/16/99	EXE4	38	18.3	58.2	38	79	4	25
British Young Adult 9/99	24	A/BEIJING/262/95	E7	96	20.6	329.3	29	100	0	79
		A/JOHANNESBURG/82/96	E5	88	53.4	403.0	83	100	13	88
		A/NEW CALEDONIA/20/99	E4	96	8.2	103.7	4	96	0	54
		A/NANCHANG/16/99	EXE4	96	12.6	201.5	21	96	0	63

TABLE 5.2

Haemagglutination Inhibition Antibody Responses
Influenza Type A(H1N1) Vaccine Component
Young Adults – January 2000

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Young Adult 1/2000	30	A/BEIJING/262/95	E3E4	86	20.0	160.0	28	97	0	59
		A/MICHIGAN/27/99	E4E1	48	17.0	54.0	10	79	0	24
		A/SHENZEN/510/99	E2E2E1	45	65.0	188.0	93	100	24	79
American Young Adult 1/2000	32	A/BEIJING/262/95	E3E4	33	29.1	77.7	54	96	8	29
		A/MICHIGAN/27/99	E4E1	25	13.7	29.1	13	50	0	4
		A/SHENZEN/510/99	E2E2E1	25	24.5	54.9	50	79	4	17
British Young Adult 1/2000	24	A/BEIJING/262/95	E3E4	96	28.3	276.9	46	100	0	79
		A/MICHIGAN/27/99	E4E1	71	18.3	67.3	17	92	0	13
		A/SHENZEN/510/99	E2E2E1	42	73.3	207.4	100	100	25	92

TABLE 5.3

Haemagglutination Inhibition Antibody Responses
Influenza Type A(H1N1) Vaccine Component
Older Adults – September 1999

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Older Adult 9/99	30	A/BEIJING/262/95	E7	50	29.6	108.0	50	97	3	33
		A/JOHANNESBURG/82/96	E5	40	74.6	196.9	87	100	37	63
		A/NEW CALEDONIA/20/99	E4	53	12.9	44.9	13	63	0	25
		A/NANCHANG/16/99	EXE4	43	24.6	80.0	43	83	3	30
American Older Adult 9/99	32	A/BEIJING/262/95	E7	22	15.0	31.7	17	39	4	22
		A/JOHANNESBURG/82/96	E5	4	54.9	75.5	83	96	30	35
		A/NEW CALEDONIA/20/99	E4	4	12.2	21.2	22	30	4	9
		A/NANCHANG/16/99	EXE4	4	18.3	28.3	26	39	4	9
British Older Adult 9/99	24	A/BEIJING/262/95	E7	79	26.7	155.4	50	100	0	67
		A/JOHANNESBURG/82/96	E5	33	41.3	103.7	67	92	4	38
		A/NEW CALEDONIA/20/99	E4	83	8.2	47.6	4	71	0	8
		A/NANCHANG/16/99	EXE4	71	14.1	73.3	25	83	0	38

TABLE 5.4

Haemagglutination Inhibition Antibody Responses
Influenza Type A(H1N1) Vaccine Component
Older Adults – January 2000

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Older Adult 1/2000	30	A/BEIJING/262/95	E3E4	40	34.8	98.5	63	90	10	33
		A/MICHIGAN/27/99	E4E1	37	17.8	44.9	7	67	3	13
		A/SHENZEN/510/99	E2E2E1	30	68.0	142.5	97	100	13	53
American Older Adult 1/2000	32	A/BEIJING/262/95	E3E4	26	28.3	58.2	39	87	17	26
		A/MICHIGAN/27/99	E4E1	22	8.7	17.3	13	26	0	0
		A/SHENZEN/510/99	E2E2E1	17	16.3	34.6	17	52	0	17
British Older Adult 1/2000	24	A/BEIJING/262/95	E3E4	63	31.7	106.8	63	88	4	54
		A/MICHIGAN/27/99	E4E1	50	18.9	58.2	17	75	4	21
		A/SHENZEN/510/99	E2E2E1	25	89.8	179.5	100	100	25	79

TABLE 5.5

Haemagglutination Inhibition Antibody Responses
Influenza Type A(H3N2) Vaccine Component
Young Adults – September 1999

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Young Adult 9/99	30	A/SYDNEY/5/97	E5	67	14.5	72.9	23	80	0	43
		A/MOSCOW/10/99	E7	67	24.1	105.5	37	97	0	47
		A/SHANGHAI/42/99	E5	50	15.5	49.2	10	73	0	13
		A/PANAMA/2007/99	E5	43	19.5	62.0	17	83	0	27
American Young Adult 9/99	24	A/SYDNEY/5/97	E5	33	15.4	41.2	29	75	0	4
		A/MOSCOW/10/99	E7	33	16.3	43.6	25	67	0	13
		A/SHANGHAI/42/99	E5	17	8.2	15.9	4	25	0	0
		A/PANAMA/2007/99	E5	29	10.9	26.7	8	46	0	4
British Young Adult 9/99	24	A/SYDNEY/5/97	E5	79	17.8	87.2	13	96	0	29
		A/MOSCOW/10/99	E7	58	36.7	116.4	79	100	0	50
		A/SHANGHAI/42/99	E5	61	17.8	54.9	4	92	0	8
		A/PANAMA/2007/99	E5	42	24.5	71.3	33	100	0	17

TABLE 5.6

Haemagglutination Inhibition Antibody Responses
Influenza Type A(H3N2) Vaccine Component
Young Adults – January 2000

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Young Adult 1/2000	30	A/SYDNEY/5/97	CEK2E4	69	20.5	110.5	31	90	7	55
		A/VICTORIA/358/99	E3	80	26.4	81.9	55	97	0	45
		A/PHILIPPINES/26/98	E5	52	7.6	24.1	3	48	0	3
American Young Adult 1/2000	24	A/SYDNEY/5/97	CEK2E4	42	21.2	58.2	46	71	4	25
		A/VICTORIA/358/99	E3	33	12.6	30.8	13	58	0	4
		A/PHILIPPINES/26/98	E5	21	5.8	10.0	0	4	0	0
British Young Adult 1/2000	24	A/SYDNEY/5/97	CEK2E4	83	29.1	164.6	50	100	0	79
		A/VICTORIA/358/99	E3	38	42.4	100.8	79	96	0	54
		A/PHILIPPINES/26/98	E5	71	6.3	24.5	0	33	0	0

TABLE 5.7

Haemagglutination Inhibition Antibody Responses
Influenza Type A(H3N2) Vaccine Component
Older Adults – September 1999

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Older Adult 9/99	30	A/SYDNEY/5/97	E5	47	20.5	56.6	27	80	3	23
		A/MOSCOW/10/99	E7	30	31.0	71.3	63	83	3	27
		A/SHANGHAI/42/99	E5	23	14.1	28.3	7	50	0	7
		A/PANAMA/2007/99	E5	30	17.8	41.9	7	60	3	13
American Older Adult 9/99	24	A/SYDNEY/5/97	E5	26	10.9	23.1	17	30	0	9
		A/MOSCOW/10/99	E7	26	11.9	25.9	17	43	4	9
		A/SHANGHAI/42/99	E5	8	7.7	14.1	9	17	0	0
		A/PANAMA/2007/99	E5	17	9.2	18.9	17	26	0	4
British Older Adult 9/99	24	A/SYDNEY/5/97	E5	67	16.8	82.3	25	92	4	33
		A/MOSCOW/10/99	E7	46	38.9	127.0	71	100	4	54
		A/SHANGHAI/42/99	E5	38	20.0	46.2	17	88	0	0
		A/PANAMA/2007/99	E5	29	35.6	82.3	67	100	4	25

TABLE 5.8

Haemagglutination Inhibition Antibody Responses
Influenza Type A(H3N2) Vaccine Component
Older Adults – January 2000

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Older Adult 1/2000	30	A/SYDNEY/5/97	CEK2E4	30	33.2	80.0	57	97	7	27
		A/VICTORIA/358/99	E3	27	33.2	71.3	70	93	3	23
		A/PHILIPPINES/26/98	E5	37	7.8	20.9	3	30	0	7
American Older Adult 1/2000	24	A/SYDNEY/5/97	CEK2E4	17	14.6	30.0	26	43	0	13
		A/VICTORIA/358/99	E3	9	10.3	20.0	13	30	0	4
		A/PHILIPPINES/26/98	E5	9	5.5	8.2	0	9	0	0
British Older Adult 1/2000	24	A/SYDNEY/5/97	CEK2E4	58	25.9	89.8	42	88	4	38
		A/VICTORIA/358/99	E3	46	30.8	87.2	63	96	4	42
		A/PHILIPPINES/26/98	E5	63	7.5	21.8	4	29	0	0

TABLE 5.9

Haemagglutination Inhibition Antibody Responses
Influenza Type B Vaccine Component
Young Adults – September 1999

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Young Adult 9/99	30	B/YAMANASHI/166/99	E9	80	32.5	285.0	60	100	10	80
		B/JOHANNESBURG/5/99	E7	80	19.5	188.0	30	97	3	60
		B/SOUTH AUSTRALIA/5/99	E4	83	14.1	139.3	13	93	3	50
American Young Adult 9/99	32	B/YAMANASHI/166/99	E9	33	69.2	219.8	79	100	33	79
		B/JOHANNESBURG/5/99	E7	29	42.4	134.5	71	100	21	50
		B/SOUTH AUSTRALIA/5/99	E4	38	33.6	106.8	54	96	17	46
		B/SHANGDONG/7/97	E10	8	11.9	17.8	25	38	4	4
British Young Adult 9/99	24	B/YAMANASHI/166/99	E9	96	33.6	465.6	54	100	4	92
		B/JOHANNESBURG/5/99	E7	88	21.2	276.9	33	100	0	83
		B/SOUTH AUSTRALIA/5/99	E4	88	17.8	213.5	29	100	0	79
		B/SHANGDONG/7/97	E10	46	14.6	40.0	25	58	0	17

TABLE 5.10

Haemagglutination Inhibition Antibody Responses
Influenza Type B Vaccine Component
Young Adults – January 2000

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Young Adult 1/2000	30	B/YAMANASHI/166/98	E7	76	19.5	167.5	31	97	3	55
		B/SHANGHAI/180/99	E8	72	18.2	152.7	28	97	3	52
		B/SHENZHEN/654/99	E6	62	13.8	62.0	14	86	0	24
		B/TENNESSEE/4/99	X2C1E1E1	90	20.0	242.4	38	97	3	79
American Young Adult 1/2000	32	B/YAMANASHI/166/98	E7	29	43.6	130.7	67	96	25	63
		B/SHANGHAI/180/99	E8	29	40.0	100.8	67	96	17	38
		B/SHENZHEN/654/99	E6	33	18.9	44.9	33	71	8	17
		B/TENNESSEE/4/99	X2C1E1E1	29	65.3	174.4	75	100	25	67
British Young Adult 1/2000	24	B/YAMANASHI/166/98	E7	88	20.6	269.0	38	100	4	79
		B/SHANGHAI/180/99	E8	88	21.2	232.8	38	100	0	83
		B/SHENZHEN/654/99	E6	67	22.4	100.8	33	100	0	38
		B/TENNESSEE/4/99	X2C1E1E1	83	24.5	391.6	33	100	0	88

TABLE 5.11

Haemagglutination Inhibition Antibody Responses
Influenza Type B Vaccine Component
Older Adults – September 1999

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Older Adult 9/99	30	B/YAMANASHI/166/99	E9	20	76.4	136.1	87	100	30	43
		B/JOHANNESBURG/5/99	E7	13	43.9	76.4	63	90	20	30
		B/SOUTH AUSTRALIA/5/99	E4	13	34.8	65.0	57	83	17	27
American Older Adult 9/99	32	B/YAMANASHI/166/99	E9	9	67.3	116.4	78	96	30	52
		B/JOHANNESBURG/5/99	E7	17	38.9	69.2	65	88	17	26
		B/SOUTH AUSTRALIA/5/99	E4	13	25.2	46.2	48	74	9	17
		B/SHANGDONG/7/97	E10	0	14.6	16.3	27	32	5	5
British Older Adult 9/99	24	B/YAMANASHI/166/99	E9	71	34.6	195.8	58	92	21	79
		B/JOHANNESBURG/5/99	E7	63	18.9	113.1	33	92	0	63
		B/SOUTH AUSTRALIA/5/99	E4	75	15.9	92.4	29	92	0	46
		B/SHANGDONG/7/97	E10	17	11.6	21.8	17	33	0	4

TABLE 5.12

Haemagglutination Inhibition Antibody Responses
Influenza Type B Vaccine Component
Older Adults – January 2000

Population	N	Antigen	Passage History	% Rise	GMT		% ≥ 40		% ≥ 160	
					Pre	Post	Pre	Post	Pre	Post
Australian Older Adult 1/2000	30	B/YAMANASHI/166/98	E7	10	45.9	72.9	73	90	17	27
		B/SHANGHAI/180/99	E8	20	40.9	71.3	63	90	20	20
		B/SHENZHEN/654/99	E6	10	32.5	66.5	53	87	7	23
		B/TENNESSEE/4/99	X2C1E1E1	3	50.4	94.0	70	90	20	40
American Older Adult 1/2000	32	B/YAMANASHI/166/98	E7	9	41.2	69.2	65	78	17	30
		B/SHANGHAI/180/99	E8	4	37.7	54.9	65	78	13	17
		B/SHENZHEN/654/99	E6	17	17.3	28.3	35	39	4	13
		B/TENNESSEE/4/99	X2C1E1E1	13	49.0	89.8	65	91	22	39
British Older Adult 1/2000	24	B/YAMANASHI/166/98	E7	63	24.5	103.7	54	92	4	54
		B/SHANGHAI/180/99	E8	58	21.2	82.3	42	88	0	46
		B/SHENZHEN/654/99	E6	42	24.5	75.5	42	92	0	33
		B/TENNESSEE/4/99	X2C1E1E1	63	25.2	134.5	54	88	8	63

6 HIGH GROWTH REASSORTANT VIRUSES FOR VACCINE PRODUCTION

A/Moscow/10/99

High-growth reassortant viruses prepared by CSL Limited were investigated for antigenic and genetic homology with the initial A/Moscow/10/99 isolate. The initial reassortant IVR-112 was found to have a deletion mutation in the viral haemagglutinin at position 197 and a change from ALANINE to PROLINE at the adjacent position 198 (Table 6.1).Antisera prepared against this reassortant were found to have reduced reactivity against a number of recent isolates compared with antiserum prepared against the virus isolate itself (Table 6.2).

Further work demonstrated that –

- all A/Moscow/10/99 reassortants prepared had genetic changes in their haemagglutinin
- these changes were associated with reduced immunogenicity against certain low-reacting isolates (Tables 6.2 and 6.3)
- on passage multiple species could be detected in the original isolate of A/Moscow/10/99
- samples of A/Moscow/10/99 in different laboratories appeared to have different predominant species and induce antisera of differing specificities

TABLE 6.1
Sequence Differences
A/Moscow/10/99 Reassortants

Virus	Amino Acid Position					
	142	145	190	193	197	198
A/Moscow/10/99 (CDC)	R	K	D	S	Q	A
A/Moscow/10/99 (Aust)	R	N	D	R	Q	A
IVR-112	R	N	D	S	-	P
IVR-117	R	N	D	S	-	P
IVR-118	R	N	D	R	Q	A
IVR-119	R	N	V	S	Q	A
IVR-122	G	R	D	S	Q	A
RESVIR-15	R	N	V	S	Q	A

TABLE 6.2
Haemagglutination-Inhibition Reactions of Influenza A(H3N2) Isolates

OCTOBER 6TH 1999

ANTISERA NO.	F527/13D	F536/15D	F570/15D	F578/15D	F579/14D	F576/15D	F595/13D	F602/12D	PASSAGE HISTORY
				ANTISERA					
	A	B	C	D	E	F	G	H	
REF. Ag	SYD/5	DAR/1	SICH/346	A/CNIC	DAR/5	MOSC/10	IVR-112	IVR-117	
A. A/SYDNEY/5/97	**1280**	1280	1280	1280	320	1280	640	320	CEK2E4
B. A/DARWIN/1/98	320	**640**	640	320	320	320	160	160	MDCK7
C. A/SICHUAN/346/99	160	160	**640**	160	160	80	80	80	E7
D. A/CNIC/149/98	320	640	320	**640**	320	640	320	160	C1C1MDCK3
E. A/DARWIN/5/99	320	640	>2560	640	**1280**	320	160	160	E4
F. A/MOSCOW/10/99	320	320	320	1280	320	**>2560**	640	640	E7
A/MOSCOW/10/99	320	320	320	640	320	>2560	**1280**	640	E6
G. IVR-112	160	320	320	640	160	1280	>2560	**1280**	IVR-112
H. IVR-117	160	320	160	640	160	>2560	>2560	>2560	IVR-117
TEST AG.									
A/SHANGHAI/42/99	160	320	320	640	640	160	160	160	E5
A/VICTORIA/358/99	160	160	160	320	160	1280	320	320	E3
A/BRISBANE/32/99	160	320	320	320	160	640	320	160	MDCKX,MDCK1
A/CHRISTCHURCH/63/99	160	160	320	320	320	320	160	160	MDCK3
A/SOUTH AUSTRALIA/81/99	80	160	160	160	320	320	160	160	MDCKX,MDCK1
A/VICTORIA/390/99	320	640	320	640	320	320	160	160	MDCK1
A/BRISBANE/156/99	320	320	640	640	640	1280	320	320	MDCKX,MDCK1
A/PERTH/46/99	160	320	320	320	160	640	320	320	X,MDCK1

TABLE 6.3
Haemagglutination-Inhibition Reactions of Influenza A(H3N2) Isolates

OCTOBER 26TH 1999									
				ANTISERA					
ANTISERA NO.	F527/13D	F536/15D	F570/15D	F578/15D	F576/15D	F579/14D	F610-14D (IVR-118)	F611-14D (IVR-118)	PASSAGE HISTORY
	B	C	D	E	F	G	J	K	
REF. Ag	SYD/5	DAR/1	SICH/346	A/CNIC	MOSC/10	DAR/5	MOSC/10	MOSC/10	
A. A/SYDNEY/5/97	**640**	640	640	640	640	160	80	80	CEK2E4
B. A/DARWIN/1/98	320	**640**	320	320	320	320	80	80	MDCK7
C. A/CNIC/149/98	160	320	320	**320**	320	320	160	160	C1C1MDCK3
D. A/MOSCOW/10/99	320	320	320	640	**1280**	320	320	320	E9
E. A/DARWIN/5/99	320	320	1280	640	160	**1280**	160	160	E5
F. A/MOSCOW/10/99 IVR-118	320	320	320	1280	>2560	320	**1280**	1280	IVR-118
TEST AG.									
A/MOSCOW/10/99 IVR-119	320	320	320	640	1280	160	320	**320**	IVR-119
A/MOSCOW/10/99	320	320	320	640	1280	320	320	320	E6
A/MOSCOW/10/99	320	320	320	640	1280	320	320	640	E7
A/JOHANNESBURG/57/99 IVR-121	160	320	640	320	80	640	80	80	IVR-121
A/BRISBANE/156/99	320	640	640	1280	1280	1280	160	160	MDCKX,MDCK2
A/VICTORIA/601/99	80	80	80	80	160	160	80	80	MDCKX,MDCK1
A/PERTH/43/99	160	320	320	160	320	160	80	80	X,MDCK2