

Olerup SSP[®] KIR Genotyping

Product number:	104.101-12 – including <i>Taq</i> polymerase 104.101-12u – without <i>Taq</i> polymerase
Lot number:	71E
Expiry date:	2010-June-01
Number of tests:	12
Number of wells per test:	23 + 1
Storage - pre-aliquoted primers:	dark at -20°C
- PCR Master Mix:	-20°C
- Adhesive PCR seals	RT
- Product Insert	RT

This Product Description is only valid for Lot No. 71E.

**CHANGES COMPARED TO THE PREVIOUS *OLERUP SSP[®]*
KIR GENOTYPING LOT**

The KIR specificity and interpretation tables have been updated for the KIR alleles described since the previous *Olerup SSP[®]* KIR Genotyping lot was made (**Lot No. Y47**).

The primers of the tubes detailed below have been exchanged, added or modified compared to the previous lot.

Well	5'-primer	3'-primer	rationale
17	Exchanged	Exchanged	Primer pair exchanged to avoid co-amplification of 3DL1*009.

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Lot-specific information

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Well **24** contains Negative Control primer pairs, that will amplify more than 95% of the *Olerup* SSP[®] HLA Class I, DRB, DQB1 and DPB1 amplicons as well as the amplicons generated by control primer pairs.

PCR product sizes range from 75 to 430 base pairs.

The PCR product generated by the control primer pair is 430 base pairs.

Length of PCR product	105	200	105	80	75	80
5'-primer¹	164	340	440	45	45	43
	5'-CAC ^{3'}	5'-Agg ^{3'}	5'-TTA ^{3'}	5'-Tg g ^{3'}	5'-Tg g ^{3'}	5'-Tg g ^{3'}
3'-primer²	231	2nd I	507	59	58	57
	5'-TgC ^{3'}	5'-AAA ^{3'}	5'-TTg ^{3'}	5'-CTC ^{3'}	5'-ggC ^{3'}	5'-CTC ^{3'}
A*	+	+	+			
B*	+	+	+			
Cw*	+	+	+			
DRB1				+	+	
DRB3				+	+	
DRB5				+		
DQB1					+	
DPB1						+

¹The nucleotide position for HLA class I genes and the codon for HLA class II genes, in the 2nd or 3rd exon, matching the specificity-determining 3'-end of the primer is given. Nucleotide and codon numbering as on the www.ebi.ac.uk/imgt/hla web site. The sequence of the 3 terminal nucleotides of the primer is given.

²The nucleotide position for HLA class I genes and the codon for HLA class II genes, in the 2nd or 3rd exon or the 2nd intron, matching the specificity-determining 3'-end of the primer is given in the anti-sense direction. Nucleotide and codon numbering as on the www.ebi.ac.uk/imgt/hla web site. The sequence of the 3 terminal nucleotides of the primer is given.

PRODUCT DESCRIPTION

KIR Genotyping SSP typing

CONTENT

The primer set contains 5'- and 3'-primers for KIR Genotyping.

PLATE LAYOUT

Each test consists of 24 PCR reactions in a 24 well cut PCR plate.

1	2	3	4	5	6	7	8
9	10	11	12	13	14	15	16
17	18	19	20	21	22	23	24

Wells 1 to 23 – KIR Genotyping primers.

Well 24 – Negative Control.

The 24 well cut PCR plate is marked with 'KIR'.

Well No. 1 is marked with the Lot No. '71E'.

The PCR plates are covered with a PCR-compatible foil.

Please note: When removing each 24 well PCR plate, make sure that the remaining plates stay covered. Use a scalpel or a similar instrument to carefully cut the foil between the plates.

UNIQUELY IDENTIFIED ALLELES

All the KIR alleles available in the IPD KIR Sequence Database in January 2008¹ will be amplified by the primers in the KIR Genotyping SSP kit.

¹KIR alleles listed on the IPD KIR web page 2008-January-18, release 2.0.0, www.ebi.ac.uk/ipd/kir.

SPECIFICITY TABLE

KIR Genotyping SSP typing

Specificities and sizes of the PCR products of the 23 primer mixes used for KIR SSP Genotyping SSP.

Primer Mix	Size of spec. PCR product ¹	Size of control band ²	KIR Gene	Amplified KIR ³ alleles
1	145 bp	800 bp	2DL1	001-010
2	145 bp	800 bp	2DL2	001-005
3	520 bp	1070 bp	2DL3	001-007
4	200 bp	800 bp	2DL4	00101-012
5⁵	190 bp	800 bp	2DL5A, 2DL5B	0010101-0010102, 0050101-005010102 0020101-004, 00601- 009
6⁶	1650 bp	430 bp	2DL5A	0010101-0010102, 0050101-005010102
7⁶	1650 bp	515 bp	2DL5B	0020101-004, 00601- 000
8⁴	100 bp	800 bp	2DS1	001-004
9	205 bp	800 bp	2DS2	0010101-005
10	160 bp	800 bp	2DS3	00101-004
11	215 bp	800 bp	2DS4	0010101-00103
12	200 bp	800 bp	2DS4	003, 004, 006, 007, 009
13⁴	110 bp	800 bp	2DS5	001-008
14	130 bp	800 bp	3DL1	00101-002, 00401-009, 01501-044, 056, 057
15⁴	95 bp	800 bp	3DL2	00101-021
16⁴	115 bp	800 bp	3DL3	00101-031
17	130 bp	800 bp	3DS1	010-014, 045-049N, 055
18	165 bp	800 bp	2DP1	00101-003
19⁴	125 bp	800 bp	3DP1	001-006
20	235 bp	800 bp	3DP1	00301-00301, 004-006 [?]
21	145 bp	800 bp	2DS1	001
22	145 bp	800 bp	2DS1	0020101-004
23⁴	80 bp	800 bp	3DL1	00401-00402

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Lot-specific information

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¹ Alleles are assigned by the presence of specific PCR product(s). However, the sizes of the specific PCR products may be helpful in the interpretation of KIR SSP typings.

When the primers in a primer mix can give rise to specific PCR products of more than one length this is indicated if the size difference is 20 base pairs or more. Size differences shorter than 20 base pairs are not given. For high resolution SSP kits the respective length of the specific PCR product(s) of the alleles amplified by these primer mixes are given.

Nonspecific amplifications, i.e. a ladder or a smear of bands, may sometimes be seen. GC-rich primers have a higher tendency of giving rise to nonspecific amplifications than other primers.

PCR fragments longer than the control bands may sometimes be observed. Such bands should be disregarded and do not influence the interpretation of the SSP typings.

PCR fragments migrating faster than the control bands, but slower than a 400 bp fragment may be seen in some gel read-outs. Such bands can be disregarded and do not influence the interpretation of the SSP typings.

Some primers may give rise to primer oligomer artifacts. Sometimes this phenomenon is an inherent feature of the primer pair(s) of a primer mix. More often it is due to other factors such as too low amount of DNA in the PCR reactions, taking too long time in setting up the PCR reactions, working at elevated room temperature or using thermal cyclers that are not pre-heated.

² The internal positive control primer pairs amplify segments of the human growth hormone gene. The three different control primer pairs give rise to either an internal positive control band of 800 base pairs, for most wells, or a band of 1070 base pairs or 430 base pairs, for some wells.

Well number 3 contains the primer pair giving rise to the 1070 bp internal positive control band, well number 6 contains the primer pair giving rise to the 430 base pair internal positive control band and well number 7 contains the primer pair giving rise to the 515 base pair internal positive control band in order to help in the correct orientation and kit identification of the KIR Genotyping typing.

In the presence of a specific amplification the intensity of the control band often decreases.

³ KIR alleles listed on the IPD KIR web page 2008-January-18, release 2.0.0, www.ebi.ac.uk/ipd/kir.

⁴ Short specific PCR fragments are less intense and not as sharp as longer specific bands.

⁵ Primer mix 5 may yield somewhat less specific PCR product than the other KIR SSP primer mixes.

⁶ The specific PCR product generated by primer mixes 6 and 7 are longer than the internal positive control band.

'?', the 2nd intron sequence of the primer matching region is not known.

INTERPRETATION TABLE												
KIR SSP Genotyping												
Amplification patterns of the KIR alleles												
	Well											
	1	2	3	4	5	6	7	8	9	10	11	12
Length of spec.	145	145	520	200	190	1650	1650	100	205	160	215	200
PCR product												
Length of int.	800	800	1070	800	800	430	515	800	800	800	800	800
pos. control ¹												
5'-primer(s) ²	130	208	344	208	226	-16	-16	165	140	226	229	234
	5'-gAA ^{3'}	5'-CCA ^{3'}	5'-CTg ^{3'}	5'-CCg ^{3'}	5'-CCA ^{3'}	5'-TCA ^{3'}	5'-TCg ^{3'}	5'-gAg ^{3'}	5'-gTA ^{3'}	5'-CCT ^{3'}	5'-CTA ^{3'}	5'-TCT ^{3'}
								165				
								5'-gAA ^{3'}				
3'-primer(s) ³	165	243	350	262	276	27	27	185	195	266	288	288
	5'-gCg ^{3'}	5'-ACA ^{3'}	5'-CAA ^{3'}	5'-ggA ^{3'}	5'-gAg ^{3'}	5'-ACA ^{3'}	5'-gTT ^{3'}	5'-gAC ^{3'}	5'-ATg ^{3'}	5'-CCT ^{3'}	5'-ggA ^{3'}	5'-ggA ^{3'}
			351									
			5'-ACC ^{3'}									
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
KIR allele ⁴												
2DL1*001-010	1											
2DL2*001-005		2										
2DL3*001-007			3									
2DL4*00101-012				4								
2DL5A*0010101-0010102, 0050101-05010102					5	6						
2DL5B*0020101-004, 00601-009					5		7					
2DS1*001								8				
2DS1*0020101-004								8				
2DS2*0010101-005									9			
2DS3*00101-004										10		
2DS4*0010101-00103											11	
2DS4*003, 004, 006, 007, 009												12
2DS5*001-008												
3DL1*00101-002, 00501-009, 01501-044, 056, 057												
3DL1*00401-00402												
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

INTERPRETATION TABLE											
KIR SSP Genotyping											
Amplification patterns of the KIR alleles											
Well											
13	14	15	16	17	18	19	20	21	22	23	
110	130	95	115	130	165	125	235	145	145	80	Length of spec.
											PCR product
800	800	800	800	800	800	800	800	800	800	800	Length of int.
											pos. control ¹
142	136	27	156	133	29	25	2 nd I	130	130	31	5'-primer(s) ²
5'-ACC3'	5'-CAA3'	5'-TCA3'	5'-CCC3'	5'-TCT3'	5'-CAT3'	5'-Tgg3'	5'-gCC3'	5'-gAA3'	5'-gAA3'	5'-TCA3'	
165	166	45	181	163	71	54	54	165	165	44	3'-primer(s) ³
5'-gTg3'	5'-CAA3'	5'-ggC3'	5'-gTA3'	5'-ggA3'	5'-TAC3'	5'-TAC3'	5'-TAC3'	5'-gCC3'	5'-gCT3'	5'-TCC3'	
13	14	15	16	17	18	19	20	21	22	23	Well No.
											KIR allele ⁴
											2DL1*001-010
											2DL2*001-005
											2DL3*001-007
											2DL4*00101-012
											2DL5A*0010101-0010102, 0050101-05010102
											2DL5B*0020101-004, 00601-009
								21	22		2DS1*001
									22		2DS1*0020101-004
											2DS2*0010101-005
											2DS3*00101-004
											2DS4*0010101-00103
											2DS4*003, 004, 006, 007, 009
13											2DS5*001-008
	14										3DL1*00101-002, 00501009, 01501-044, 056, 057
	14									23	3DL1*00401-00402
13	14	15	16	17	18	19	20	21	22	23	Well No.

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Lot-specific information

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Length of spec.	145	145	520	200	190	1650	1650	100	205	160	215	200
PCR product												
Well No.	1	2	3	4	5	6	7	8	9	10	11	12
3DL2*00101-021												
3DL3*00101-031												
3DS1*010-014, 045-049N, 055												
2DP1*00101-003												
3DP1*001-002												
3DP1*00301-00302												
3DP1*004, 005, 006												
KIR allele ⁴												
Well No.	1	2	3	4	5	6	7	8	9	10	11	12

¹The internal positive control primer pairs amplify segments of the human growth hormone gene. The three different control primer pairs give rise to either an internal positive control band of 800 base pairs, for most wells, or a band of 1070 base pairs or 430 base pairs, for some wells.

Well number 3 contains the primer pair giving rise to the 1070 bp internal positive control band and wells number 6 and 7 contain the primer pair giving rise to the 430 base pair internal positive control band in order to help in the correct orientation and kit identification of the KIR Genotyping typing.

110	130	95	115	130	165	125	235	145	145	80	Length of spec.
											PCR product
13	14	15	16	17	18	19	20	21	22	23	Well No.
		15									3DL2*00101-021
			16								3DL3*00101-031
				17							3DS1*010-014, 045-049N, 055
					18						2DP1*00101-003
						19					3DP1*001-002
						19	20				3DP1*00301-00302
						19	?				3DP1*004, 005, 006
											KIR allele⁴
13	14	15	16	17	18	19	20	21	22	23	Well No.

²The nucleotide position, in the 1st, 3rd, 4th, 5th or 7th exon or the 2nd intron matching the specificity-determining 3'-end of the primer is given. Nucleotide numbering as on the KIR web page 2006-November-10, release 1.3.0, www.ebi.ac.uk/ipd/kir. The sequence of the 3 terminal nucleotides of the primer is given.

³The nucleotide position, in the 3rd, 4th, 5th or 8th exon, matching the specificity-determining 3'-end of the primer. Nucleotide numbering as on the KIR web page 2006-November-10, release 1.3.0, www.ebi.ac.uk/ipd/kir. The sequence of the 3 terminal nucleotides of the primer is given in the anti-sense direction.

⁴KIR alleles listed on the IPD KIR web page 2008-January-18, release 2.0.0, www.ebi.ac.uk/ipd/kir. '?', the 2nd intron sequence of the primer matching region is not know.

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Lot-specific information

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CELL LINE VALIDATION SHEET																		
KIR Genotyping primer set																		
			Well															
			1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
			200505001	200505002	200733003	200505004	200505005	200505006	200505007	200505008	200505009	200505010	200505011	200505012	200505013	200505014	200505015	200505016
		IHWC cell line																
1	9001	SA	+	-	+	+	-	-	-	-	-	-	+	-	-	+	+	+
2	9280	LK707	-	+	-	+	+	-	+	+	+	-	-	+	+	+	+	+
3	9011	E4181324	+	-	+	+	-	-	-	-	-	-	+	+	-	+	+	+
4	9275	GU373	+	-	+	+	-	-	-	-	-	-	+	+	-	+	+	+
5	9009	KAS011	+	-	+	+	+	+	-	+	-	-	-	+	+	+	+	+
6	9353	SM	+	-	+	+	+	+	-	+	-	+	+	-	-	+	+	+
7	9020	QBL	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
8	9007	DEM	+	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+
9	9026	YAR	+	-	+	+	-	-	-	-	-	-	+	+	-	+	+	+
10	9107	LKT3	+	-	+	+	-	-	-	-	-	-	+	-	-	+	+	+
11	9051	PITOUT	+	+	+	+	-	-	-	-	+	-	-	+	-	+	+	+
12	9052	DBB	-	+	+	+	+	-	+	-	+	+	-	+	-	+	+	+
13	9067	BTB	+	-	+	+	-	-	-	-	-	-	-	+	-	+	+	+
14	9071	OLGA	+	-	+	+	+	+	-	+	-	-	-	+	+	+	+	+
15	9075	DKB	+	-	+	+	-	-	-	-	-	-	+	-	-	+	+	+
16	9037	SWEIG007	+	-	+	+	-	-	-	-	-	-	-	+	-	+	+	+
17	9008	WILJON	+	-	+	+	-	-	-	-	-	-	-	+	-	+	+	+
18	9257	32367	+	-	+	+	-	-	-	-	-	-	-	+	-	+	+	+
19	9038	BM16	+	-	+	+	-	-	-	-	-	-	-	+	-	+	+	+
20	9059	SLE005	+	-	+	+	-	-	-	-	-	-	+	+	-	+	+	+
21	9064	AMALA	+	+	+	+	+	+	-	+	+	-	+	-	+	+	+	+
22	9056	KOSE	+	+	+	+	-	-	-	-	+	-	-	+	-	+	+	+
23	9124	IHL	+	+	+	+	-	-	-	-	+	-	+	+	-	+	+	+
24	9035	JBUSH	+	-	+	+	-	-	-	-	-	-	+	+	-	+	+	+
25	9049	IBW9	+	-	+	+	-	-	-	-	-	+	+	+	-	+	+	+
26	9285	WT49	+	+	+	+	+	-	+	+	+	+	-	+	+	+	+	+
27	9191	CH1007	+	+	+	+	+	-	+	-	+	-	+	+	-	+	+	+
28	9320	BEL5GB	+	+	-	+	+	-	+	-	+	-	-	+	-	+	+	+
29	9050	MOU	+	-	+	+	-	-	-	-	-	-	-	+	-	+	+	+
30	9021	RSH	+	+	+	+	+	-	+	-	+	-	+	+	+	+	+	+
31	9019	DUCAF	+	-	+	+	-	-	-	-	-	-	-	+	-	+	+	+
32	9297	HAG	+	-	+	+	-	-	-	-	-	-	+	+	-	+	+	+
33	9098	MT14B	+	-	+	+	-	-	-	-	-	-	+	-	-	+	+	+
34	9104	DHIF	+		+	+	-	-	-	-	+	-	+	+	-	+	+	+
35	9302	SSTO	+	+	+	+	-	-	-	-	+	-	+	+	-	+	+	+
36	9024	KT17	+	-	+	+	+	+	-	+	-	+	-	+	-	+	+	+
37	9065	HHKB	+	+	+	+	+	+	-	+	+	-	-	+	+	+	+	+
38	9099	LZL	+	+	+	+	+	-	+	+	+	+	+	-	-	+	+	+
39	9315	CML		+	-	+	+	+	+	+	+	+	+	-	+	-	+	+
40	9134	WHONP199	+	-	+	+	-	-	-	-	-	-	+	-	-	+	+	+
41	9055	H0301	+	+	-	+	+	-	+	-	+	+	+	-	-	+	+	+
42	9066	TAB089	+	-	+	+	-	-	-	-	-	-	-	+	-	+	+	+
43	9076	T7526	+	-	+	+	+	+	-	+	+	-	+	-	+	+	+	+
44	9057	TEM	+	+	+	+	+	-	+	-	+	+	-	+	-	+	+	+
45	9239	SHJO	+	+	+	+	+	-	+	-	-	-	+	-	+	+	+	+
46	9013	SCHU	+	-	+	+	-	-	-	-	-	-	+	+	-	+	+	+
47	9045	TUBO	+	-	+	+	-	-	-	-	-	-	-	+	-	+	+	+
48	9303	TER-ND	+	-	+	+	-	-	-	-	-	-	-	+	-	+	+	+

Lot No.: **71E**

Lot-specific information

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CELL LINE VAL. SHEET									
KIR Genotyping primer set									
			Well						
			17	18	19	20	21	22	23
			200847517	200505018	200505019	200505020	200505021	200505022	200505023
IHWC cell line									
1	9001	SA	-	+	+	+	-	-	-
2	9280	LK707	-	-	+	-	-	+	-
3	9011	E4181324	+	+	+	+	-	-	+
4	9275	GU373	-	+	+	+	-	-	-
5	9009	KAS011	+	+	+	+	-	+	-
6	9353	SM	+	+	+	+	-	+	-
7	9020	QBL	+	+	+	+	-	+	+
8	9007	DEM	+	+	+	+	-	+	-
9	9026	YAR	-	+	+	+	-	-	-
10	9107	LKT3	-	+	+	+	-	-	-
11	9051	PITOUT	-	+	+	+	-	-	+
12	9052	DBB	-	+	+	+	-	-	+
13	9067	BTB	-	+	+	+	-	-	+
14	9071	OLGA	+	+	+	+	-	+	-
15	9075	DKB	-	+	+	+	-	-	-
16	9037	SWEIG007	-	+	+	+	-	-	+
17	9008	WILJON	-	+	+	+	-	-	+
18	9257	32367	-	+	+	+	-	-	+
19	9038	BM16	-	+	+	+	-	-	-
20	9059	SLE005	-	+	+	+	-	-	-
21	9064	AMALA	+	+	+	+	-	+	-
22	9056	KOSE	-	+	+	+	-	-	+
23	9124	IHL	-	+	+	+	-	-	+
24	9035	JBUSH	-	+	+	+	-	-	-
25	9049	IBW9	-	+	+	+	-	-	+
26	9285	WT49	-	+	+	+	-	+	-
27	9191	CH1007	-	+	+	+	-	-	-
28	9320	BEL5GB	-	+	+	+	-	-	+
29	9050	MOU	-	+	+	+	-	-	+
30	9021	RSH	-	+	+	+	-	-	-
31	9019	DUCAF	-	+	+	+	-	-	-
32	9297	HAG	-	+	+	+	-	-	-
33	9098	MT14B	-	+	+	+	-	-	-
34	9104	DHIF	-	+	+	+	-	-	-
35	9302	SSTO	-	+	+	+	-	-	-
36	9024	KT17	+	+	+	+	-	+	-
37	9065	HHKB	+	+	+	+	-	+	-
38	9099	LZL	-	+	+	+	-	+	-
39	9315	CML	+	+	+	+	-	+	+
40	9134	WHONP199	-	+	+	+	-	-	-
41	9055	H0301	-	+	+	+	-	-	-
42	9066	TAB089	-	+	+	+	-	-	-
43	9076	T7526	+	+	+	+	-	+	-
44	9057	TEM	-	+	+	+	-	-	-
45	9239	SHJO	-	+	+	+	-	-	-
46	9013	SCHU	-	+	+	+	-	-	-
47	9045	TUBO	-	+	+	+	-	-	-
48	9303	TER-ND	-	+	+	+	-	-	-

CERTIFICATE OF ANALYSIS**Olerup SSP® KIR Genotyping SSP****Product number:** 104.101-12 – including *Taq* polymerase104.101-12u – without *Taq* polymerase**Lot number:** 71E**Expiry date:** 2010-June-01**Number of tests:** 12**Number of wells per test:** 23 + 1**Well specifications:**

Well No.	Production No.	Well No.	Production No.	Well No.	Production No.
1	2005-050-01	9	2005-050-09	17	2008-475-18
2	2005-050-02	10	2005-050-10	18	2005-050-19
3	2007-330-03	11	2005-050-12	19	2005-050-20
4	2005-050-04	12	2005-050-13	20	2005-050-21
5	2005-050-05	13	2005-050-14	21	2005-050-22
6	2005-050-06	14	2005-050-15	22	2005-050-23
7	2005-050-07	15	2005-050-16	23	2005-050-24
8	2005-050-08	16	2005-050-17		

The specificity of each primer solution of the kit has been tested against 48 IHWC cell line DNAs.

The negative control primer pairs, **Production No. 2006-257-01**, can detect contamination with PCR products diluted 10^{-7} .

Results: No false positive or false negative amplifications were obtained.

Date of approval: 2008-June-03

Approved by:

Quality Control, Supervisor

Lot No.: **71E**

Lot-specific information

www.olerup.com

Declaration of Conformity

Product name: Olerup SSP® KIR Genotyping
Product number: 104.101-12, 104.101-12u
Lot number: 71E

Intended use: KIR Genotyping

Manufacturer: Olerup SSP AB
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We, Olerup SSP AB, hereby declare that this product, to which this Declaration of Conformity relates is in conformity with the following Standard(s) and other normative document(s) ISO 9001:2000 and ISO 13485:2003, following the provisions of the 98/79/EC Directive on *in vitro* diagnostic medical devices, Annex III, as transposed into the national laws of the Member States of the European Union.

The Technical Documentation File is maintained at Olerup SSP AB, Hasselstigen 1, SE-133 33 Saltsjöbaden, Sweden.

The Authorized Representative located within the Community is: Olerup SSP AB.

Saltsjöbaden, Sweden
2008-June-03

Olle Olerup
Managing Director

Lot No.: **71E**

Lot-specific information

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