

Olerup SSP[®] HLA-B*41

Product number:	101.542-06 – including <i>Taq</i> polymerase
Lot number:	11G
Expiry date:	2011-July-01
Number of tests:	6
Number of wells per test:	12
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. 11G.

CHANGES COMPARED TO THE PREVIOUS *OLERUP SSP*[®] HLA-B*41 LOT.

The HLA-B*41 specificity and interpretation tables have been updated for the HLA-B alleles described since the previous *Olerup SSP*[®] HLA-B*41 lot was made (**Lot No. 50E**).

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

Well	5'-primer	3'-primer	rationale
1	-	Modified	Modified primer to improve specificity.
4	-	Modified	Modified primer to improve specificity.
10	-	Modified	Modified primer to improve specificity.

PRODUCT DESCRIPTION

HLA-B*41 SSP typing

CONTENT

The primer set contains 5'- and 3'-primers for identifying the B*4101 to B*4108 alleles.

PLATE LAYOUT

Each HLA-B*41 test consists of 12 PCR reactions in a 16 well cut PCR plate. Wells 13 to 16 are empty.

1	2	3	4	5	6	7	8
9	10	11	12	empty	empty	empty	empty

The 16 well PCR plate is marked with ‘HLA-B*41’ in silver/gray ink.

Well No. 1 with the Lot No. ‘11G’.

The PCR plates are heat-sealed with a PCR-compatible foil.

Please note: When removing each 16 well PCR plate, make sure that the remaining plates stay sealed. Use a scalpel or a similar instrument to carefully cut the foil between the plates.

INTERPRETATION

The interpretation of HLA-B*41 SSP subtypings will be influenced by three B*07, most B*08, most B*13, ten B*15, most B*18, three B*27, four B*35, the B*37, four B*39, most B*40, the B*42, six B*44, the B*45, two B*46, the B*49, the B*50, most B*51, the B*52, the B*5315, the B*54, most B*55, most B*56, the B*5808, the B*59, the B*7301 and the B*78 alleles when present on the other haplotype. In addition, the Cw*070403 allele will be amplified by primer mix 12.

UNIQUELY IDENTIFIED ALLELES

All the HLA-B*41¹, i.e. **B*4101 to B*4108** recognized by the HLA Nomenclature Committee in May 2009² will be amplified by the primers in the HLA-B*41 SSP kit.

¹The B*4106 and B*4508 alleles give rise to identical amplification patterns with the HLA-B*41 high resolution kit. These two alleles can be distinguished by the HLA-B low resolution and/or HLA-B*45 kits.

²HLA-B alleles listed on the IMGT/HLA web page 2009-May-11, release 2.25.2, www.ebi.ac.uk/imgt/hla.

RESOLUTION IN HOMO- AND HETEROZYGOTES

The eight HLA-B*41 alleles give rise to 9 different amplification patterns that can be combined in 45 homozygous and heterozygous combinations. Fourteen of these genotypes do not give rise to unique amplification patterns.

+++--++-	-+++	4101,4104 = 4102,4107 = 4104,4107
+---+--+	-++-	4101,4107 = 4107,4107
+---+--+	-++-	4101,4105 = 4105,4105
-++-+---	-++-	4102,4104 = 410302,4104 = 4104,4104
-+-----	-++-	4102,4102 = 4102,410302
--+-----	-++-	410301,410301 = 410301,410302

SPECIFICITY TABLE

HLA-B*41 SSP subtyping

Specificities and sizes of the PCR products of the 12 primer mixes used for HLA-B*41 SSP subtyping

Primer Mix	Size of spec. PCR product ¹	Size of control band ²	Amplified HLA-B*41 alleles	Other amplified HLA Class I alleles ³
1	285 bp	800 bp	4101, 4105, 4107	0809, 4204, 5520
2	215 bp	1070 bp	4102, 4104	0704, 0719, 0725, 080101-0805, 0807, 0808N, 0810, 0811, 0814, 0815, 0817-0819N, 0821-0824, 0826-0838, 3587, 3709, 4201, 4202, 420501-4210
3 ^{4,5}	70 bp	1070 bp	410301	
4	265 bp	800 bp	4104, 4107	5315, 5520
5	255 bp	1070 bp	4105	
6	220 bp	1070 bp	4101, 4105-4107	0809, 1583, 4204, 4415, 4418, 4420, 4501-4509, 5108, 5120, 5136, 5144N, 5520, 5613
7 ⁶	295 bp	800 bp	4106	130201-1304, 1308Q, 1309, 1314-1316, 1318, 1319, 1504, 1516, 1542, 1567, 1583, 1595, 9537, 9555, 2714, 3537, 3560, 390601, 390602, 3933, 3934, 40060101-400602, 4044, 4053, 4070, 4075, 4083, 4086, 4093, 4095, 4096, 4415, 4418, 4420, 4447, 4501-4509, 4611, 4618, 490101-4905, 5001, 5002, 5004, 510101-5103, 5105, 5107-512402, 5126-5141N,

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Lot-specific information

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				5143, 5144N, 5148-5155, 5157, 5158, 5160, 5161, 5163, 5165, 520101-5214, 5401-5405N, 5407, 5408N, 5410-5417, 550101-5503, 5505, 5507, 5509-5513, 5515-5519, 5521-5534, 5601, 560501-5608, 5613-5617, 5619N-5624, 5808, 5901-5904, 7301, 7801-7806
8⁵	215 bp	1070 bp	4101-4108	1323, 1571, 180101-1803, 1805, 1806, 1808-1815, 1817N-1824, 1826-1828, 1830-1832, 2718, 2729, 3563, 370101-3714, 400101-400202, 400204-400602, 4009, 4011, 401401-4016, 4018-4020, 4022N-4039, 4042-4048, 4050-4057, 4059, 4060, 4062-4067, 4069-4074, 4076-4096, 4415, 4418, 4455, 4501-4505, 4507-4509, 490101-4905, 5001, 5002, 5004
9^{4,5}	105 bp	1070 bp	4108	0704, 0725, 420501-420502
10⁴	105 bp	800 bp	4101-4105, 4107	080101-0805, 0807-0812, 0814-0819N, 0821-0824, 0826-0830N, 0832-0838, 3587, 4201, 4202, 4204, 4206, 4207, 4209, 4210, 5520
11	245 bp	800 bp	4101, 4105-4107	0809, 1542, 1583, 3560, 390601, 390602, 3934, 4204, 4508, 4618, 5121, 5136, 5401, 5402, 5404, 5405N, 5407, 5408N, 5410-5417, 550101-

				5503, 5505, 5507, 5510-5513, 5515-5521, 5523, 5525-5534, 5623, 5901-5903
12	210 bp	1070 bp	4102-4104, 4108	0704, 0719, 0725, 080101-0805, 0807, 0808N, 0810-0812, 0814-0819N, 0821- 0824, 0826, 0828-0832, 0834-0838, 1551, 3587, 3709, 3712, 4201, 4202, 420501-4210, 4441, Cw*070403

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 HLA-B*41 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 lengths 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

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

Well number 1 contains the primer pair giving rise to the shorter, 800 bp, internal positive control band in order to help in the correct orientation of the HLA-B*41 subtyping.

In addition, wells number 4, 7, 10 and 11 contain the primer pair giving rise to the shorter, 800 bp, internal positive control band in order to allow kit identification.

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

³Due to the sharing of sequence motifs between HLA-B alleles some non-HLA-B*41 alleles will be amplified by primer mixes 1, 2, 4, 6 to 12. In addition, the Cw*070403 allele will be amplified by primer mix 12.

The B*4106 and B*4508 alleles give rise to identical amplification patterns with the HLA-B*41 high resolution kit. These two alleles can be distinguished by the HLA-B low resolution and/or HLA-B*45 kits.

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

⁵Primer mixes 3, 8 and 9 may give rise to nonspecific amplifications.

⁶Primer mix 7 has tendencies of giving rise to primer dimer artefacts.

INTERPRETATION TABLE								
HLA-B*41 SSP subtyping								
Amplification patterns of the HLA-B*4101 to B*4108 alleles								
	Well							
	1	2	3	4	5	6	7	8
Length of spec.	285	215	70	265	255	220	295	215
PCR product								
Length of int.	800	1070	1070	800	1070	1070	800	1070
pos. control¹								
5'-primer²	357	363	97	379	97	357	357	97
	5' -Tgg 3'	5' -AgC 3'	5' -TCC 3'	5' -ACC 3'	5' -TCC 3'	5' -Tgg 3'	5' -Tgg 3'	5' -TCC 3'
3'-primer³	603	538	126	603	312	538	610	272
	5' -gTg 3'	5' -gTC 3'	5' -TCT 3'	5' -gTg 3'	5' -AgC 3'	5' -gTC 3'	5' -CTg 3'	5' -Tgg 3'
Well No.	1	2	3	4	5	6	7	8
HLA-B allele								
*4101	1					6		8
*4102		2						8
*410301			3					8
*410302								8
*4104		2		4				8
*4105	1				5	6		8
*4106, 4508⁴						6	7	8
*4107	1			4		6		8
*4108								8
*0704, 0725, 420501, 420502		2						
*0719, 0831, 4208		2						
*080101-0805, 0807, 0808N, 0810, 0811, 0814, 0815, 0817-0819N, 0821-0824, 0826, 0828-0830N, 0832, 0834-0838, 3587, 4201, 4202, 4206, 4207, 4209, 4210		2						
*0809, 4204	1					6		
*0812, 0816								
*0827, 0833		2						
Well No.	1	2	3	4	5	6	7	8

INTERPRETATION TABLE				
HLA-B*41 subtyping				
Amplification patterns of the B*41 alleles				
Well				
9	10	11	12	
105	105	245	210	Length of spec.
				PCR product
1070	800	800	1070	Length of int.
				pos. control¹
540	540	357	369	5'-primer²
5'-gAC 3'	5'-gAC 3'	5'-Tgg 3'	5'-TAC 3'	
605	603	559	538	3'-primer³
5'-gCT 3'	5'-gTg 3'	5'-CgT 3'	5'-gTC 3'	
9	10	11	12	Well No.
				HLA-B allele
	10	11		*4101
	10		12	*4102
	10		12	*410301
	10		12	*410302
	10		12	*4104
	10	11		*4105
		11		*4106, 4508⁴
	10	11		*4107
9			12	*4108
9			12	*0704, 0725, 420501, 420502
			12	*0719, 0831, 4208
	10		12	*080101-0805, 0807, 0808N, 0810, 0811, 0814, 0815, 0817-0819N, 0821-0824, 0826, 0828-0830N, 0832, 0834-0838, 3587, 4201, 4202, 4206, 4207, 4209, 4210
	10	11		*0809, 4204
	10		12	*0812, 0816
	10			*0827, 0833
9	10	11	12	Well No.

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Lot-specific information

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Length of spec.	285	215	70	265	255	220	295	215
PCR product								
Well No.	1	2	3	4	5	6	7	8
*130201-1304, 1308Q, 1309, 1314-1316, 1318, 1319, 1504, 1516, 1567, 1595, 9537, 9555, 2714, 3537, 3933, 4075, 4447, 4611, 510101-5103, 5105, 5107, 510901-5119, 5122-512402, 5126-5135, 5137-5141N, 5143, 5148-5155, 5157, 5158, 5160, 5161, 5163, 5165, 520101-5214, 5403, 5509, 5522, 5524, 5601, 560501-5608, 5614-5617, 5619N-5622, 5624, 5808, 5904, 7301, 7801-7806							7	
*1323, 1571, 180101-1803, 1805, 1806, 1808-1815, 1817N-1824, 1826-1828, 1830-1832, 2718, 2729, 3563, 370101-3708, 3710, 3711, 3713, 3714, 400101-400202, 400204-4005, 4009, 4011, 401401-4016, 4018-4020, 4022N-4039, 4042, 4043, 4045-4048, 4050-4052, 4054-4057, 4059, 4060, 4062-4067, 4069, 4071-4074, 4076-4082, 4084, 4085, 4087-4092, 4094, 4455								8
*1542, 3560, 390601, 390602, 3934, 4618, 5121, 5401, 5402, 5404, 5405N, 5407, 5408N, 5410-5417, 550101-5503, 5505, 5507, 5510-5513, 5515-5519, 5521, 5523, 5525-5534, 5623, 5901-5903							7	
*1551, 4441, <i>Cw*070403</i>								
*1583, 5136						6	7	
*3709		2						8
*3712								8
*40060101-400602, 4044, 4053, 4070, 4083, 4086, 4093, 4095, 4096, 490101-4905, 5001, 5002, 5004							7	8
*4415, 4418, 4501-4505, 4507, 4509						6	7	8
*4420, 4506, 5108, 5120, 5144N, 5613						6	7	
*5315				4				
*5520	1			4		6		
Well No.	1	2	3	4	5	6	7	8

105	105	245	210	Length of spec. PCR product
9	10	11	12	Well No.
				*130201-1304, 1308Q, 1309, 1314-1316, 1318, 1319, 1504, 1516, 1567, 1595, 9537, 9555, 2714, 3537, 3933, 4075, 4447, 4611, 510101-5103, 5105, 5107, 510901-5119, 5122-512402, 5126-5135, 5137-5141N, 5143, 5148-5155, 5157, 5158, 5160, 5161, 5163, 5165, 520101-5214, 5403, 5509, 5522, 5524, 5601, 560501-5608, 5614-5617, 5619N-5622, 5624, 5808, 5904, 7301, 7801-7806
				*1323, 1571, 180101-1803, 1805, 1806, 1808-1815, 1817N-1824, 1826-1828, 1830-1832, 2718, 2729, 3563, 370101-3708, 3710, 3711, 3713, 3714, 400101-400202, 400204-4005, 4009, 4011, 401401-4016, 4018-4020, 4022N-4039, 4042, 4043, 4045-4048, 4050-4052, 4054-4057, 4059, 4060, 4062-4067, 4069, 4071-4074, 4076-4082, 4084, 4085, 4087-4092, 4094, 4455
		11		*1542, 3560, 390601, 390602, 3934, 4618, 5121, 5401, 5402, 5404, 5405N, 5407, 5408N, 5410-5417, 550101-5503, 5505, 5507, 5510-5513, 5515-5519, 5521, 5523, 5525-5534, 5623, 5901-5903
			12	*1551, 4441, <i>Cw</i> *070403
		11		*1583, 5136
			12	*3709
			12	*3712
				*40060101-400602, 4044, 4053, 4070, 4083, 4086, 4093, 4095, 4096, 490101-4905, 5001, 5002, 5004
				*4415, 4418, 4501-4505, 4507, 4509
				*4420, 4506, 5108, 5120, 5144N, 5613
				*5315
	10	11		*5520
9	10	11	12	Well No.

Lot No.: **11G**

Lot-specific information

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In addition, wells number 4, 7, 10 and 11 contain the primer pair giving rise to the shorter, 800 bp, internal positive control band in order to allow kit identification.

²The nucleotide position, in the 2nd or 3rd exons, matching the specificity-determining 3'-end of the primer is given. Nucleotide numbering as on the www.ebi.ac.uk/imgt/hla web site. The sequence of the 3 terminal nucleotides of the primer is given.

³The nucleotide position, in the 2nd or 3rd exons, matching the specificity-determining 3'-end of the primer is given. Nucleotide numbering as on the www.ebi.ac.uk/imgt/hla web site. The sequence of the 3 terminal nucleotides of the primer is given.

⁴The B*4106 and B*4508 alleles give rise to identical amplification patterns with the HLA-B*41 high resolution kit. These two alleles can be distinguished by the HLA-B low resolution and/or HLA-B*45 kits.

CELL LINE VALIDATION SHEET																
HLA-B*41 SSP primer set																
				Well												
				1	2	3	4	5	6	7	8	9	10	11	12	
				Prod. No.:	200962001	200845402	200845403	200962004	200845405	200845406	200845407	200845408	200962009	200962010	200845411	200845412
	IHC cell line			B*												
1	9001	SA	*0702		-	-	-	-	-	-	-	-	-	-	-	-
2	9280	LK707	*5201	*7301	-	-	-	-	-	-	+	-	-	-	-	-
3	9011	E4181324	*52011		-	-	-	-	-	-	+	-	-	-	-	-
4	9275	GU373	*1510	*5301	-	-	-	-	-	-	-	-	-	-	-	-
5	9009	KAS011	*3701		-	-	-	-	-	-	-	+	-	-	-	-
6	9353	SM	*3901	*5101	-	-	-	-	-	-	+	-	-	-	-	-
7	9020	QBL	*1801		-	-	-	-	-	-	-	+	-	-	-	-
8	9025	DEU	*3501		-	-	-	-	-	-	-	-	-	-	-	-
9	9026	YAR	*3801		-	-	-	-	-	-	-	-	-	-	-	-
10	9107	LKT3	*5401		-	-	-	-	-	-	+	-	-	-	+	-
11	9051	PITOUT	*4403		-	-	-	-	-	-	-	-	-	-	-	-
12	9052	DBB	*5701		-	-	-	-	-	-	-	-	-	-	-	-
13	9004	JESTHOM	*2705		-	-	-	-	-	-	-	-	-	-	-	-
14	9071	OLGA	*1501	*1520	-	-	-	-	-	-	-	-	-	-	-	-
15	9075	DKB	*4001		-	-	-	-	-	-	-	+	-	-	-	-
16	9037	SWEIG007	*4002		-	-	-	-	-	-	-	+	-	-	-	-
17	9282	CTM3953540	*0801	*5501	-	+	-	-	-	-	+	-	-	+	+	+
18	9257	32367	*1401	*5601	-	-	-	-	-	-	+	-	-	-	-	-
19	9038	BM16	*1801		-	-	-	-	-	-	-	+	-	-	-	-
20	9059	SLE005	*4001		-	-	-	-	-	-	-	+	-	-	-	-
21	9064	AMALA	*1501		-	-	-	-	-	-	-	-	-	-	-	-
22	9056	KOSE	*3503		-	-	-	-	-	-	-	-	-	-	-	-
23	9124	IHL	*4002	*5602	-	-	-	-	-	-	-	+	-	-	-	-
24	9035	JBUSH	*3801		-	-	-	-	-	-	-	-	-	-	-	-
25	9049	IBW9	*1402		-	-	-	-	-	-	-	-	-	-	-	-
26	9285	WT49	*5801		-	-	-	-	-	-	-	-	-	-	-	-
27	9191	CH1007	*0705	*5101	-	-	-	-	-	-	+	-	-	-	-	-
28	9320	BEL5GB	*4402	*4403	-	-	-	-	-	-	-	-	-	-	-	-
29	9050	MOU	*4403		-	-	-	-	-	-	-	-	-	-	-	-
30	9021	RSH	*4201		-	+	-	-	-	-	-	-	-	+	-	+
31	9019	DUCAF	*1801		-	-	-	-	-	-	-	+	-	-	-	-
32	9297	HAG	*4102		-	+	-	-	-	-	-	+	-	+	-	-
33	9098	MT14B	*4001		-	-	-	-	-	-	-	+	-	-	-	-
34	9104	DHIF	*3801		-	-	-	-	-	-	-	-	-	-	-	-
35	9302	SSTO	*4402		-	-	-	-	-	-	-	-	-	-	-	-
36	9024	KT17	*1501	*3501	-	-	-	-	-	-	-	-	-	-	-	-
37	9065	HHKB	*0702		-	-	-	-	-	-	-	-	-	-	-	-
38	9099	LZL	*1501		-	-	-	-	-	-	-	-	-	-	-	-
39	9315	CML	*0801	*2705	-	+	-	-	-	-	-	-	-	+	-	+
40	9134	WHONP199	*1302	*4601	-	-	-	-	-	-	-	-	-	-	-	-
41	9055	H0301	*1402		-	-	-	-	-	-	-	-	-	-	-	-
42	9066	TAB089	*4601		-	-	-	-	-	-	-	-	-	-	-	-
43	9076	T7526	*4601		-	-	-	-	-	-	-	-	-	-	-	-
44	9057	TEM	*3801		-	-	-	-	-	-	-	-	-	-	-	-
45	9239	SHJO	*4201	*5001	-	+	-	-	-	-	+	+	-	+	-	+
46	9013	SCHU	*0702		-	-	-	-	-	-	-	-	-	-	-	-
47	9045	TUBO	*5101		-	-	-	-	-	-	+	-	-	-	-	-
48	9303	TER-ND	*3501	*4403	-	-	-	-	-	-	-	-	-	-	-	-

CERTIFICATE OF ANALYSIS

Olerup SSP[®] HLA-B*41 SSP

Product number: 101.542-06 – including *Taq* polymerase
Lot number: 11G
Expiry date: 2011-July-01
Number of tests: 6
Number of wells per test: 12

Well specifications:

Well No.	Production No.	Well No.	Production No.
1	2009-620-01	9	2009-620-09
2	2008-454-02	10	2009-620-10
3	2008-454-03	11	2008-454-11
4	2009-620-04	12	2008-454-12
5	2008-454-05		
6	2008-454-06		
7	2008-454-07		
8	2008-454-08		

The specificity of each primer solution of the HLA-B*41 primer set has been tested against 48 well characterized IHWC cell line DNAs.

No DNAs carrying the alleles to be amplified by primer solutions 3, 4, 5 and 9 were available. The specificities of the primers in primer solutions 4 and 9 were tested by separately adding one additional 3'-primer, respectively one additional 5'-primer. In primer solution 3 and 5 it was only possible to test the 5'-primer, the 3'-primer was not possible to test.

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

Date of approval: 2009-July-11

Approved by:

Quality Control, Supervisor

Lot No.: **11G**

Lot-specific information

www.olerup.com

Declaration of Conformity

Product name: *Olerup* SSP® HLA-B*41
Product number: 101.542-06
Lot number: 11G

Intended use: HLA-B*41 high resolution histocompatibility testing

Manufacturer: *Olerup* SSP AB
Hasselstigen 1
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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 II List B, conformity assessed using Annex IV, 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.

Notified Body: Lloyd's Register Quality Assurance Limited, Hiramford, Middlemarch Office Village, Siskin Drive, Coventry CV3 4FJ, United Kingdom. (Notified Body number: 0088.)

Saltsjöbaden, Sweden
2009-July-11

Olle Olerup
Managing Director

Lot No.: **11G**

Lot-specific information

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