

## **Olerup SSP<sup>®</sup> KIR HLA Ligand**

<b>Product number:</b>	<b>104.201-12 – including <i>Taq</i> polymerase</b> <b>104.201-12u – without <i>Taq</i> polymerase</b>
<b>Lot number:</b>	<b>26E</b>
<b>Expiry date:</b>	<b>2010-February-01</b>
<b>Number of tests:</b>	<b>12</b>
<b>Number of wells per test:</b>	<b>5</b>
<b>Storage - pre-aliquoted primers:</b>	<b>dark at -20°C</b>
- PCR Master Mix:	<b>-20°C</b>
- Adhesive PCR seals	<b>RT</b>
- Product Insert	<b>RT</b>

**This Product Description is only valid for Lot No. 26E.**

### **CHANGES COMPARED TO THE PREVIOUS *OLERUP SSP*<sup>®</sup> KIR HLA LIGAND LOT**

The KIR HLA Ligand specificity and interpretation tables have been updated for the HLA-A, HLA-B and HLA-Cw alleles described since the previous *Olerup SSP*<sup>®</sup> KIR HLA Ligand lot was made (**Lot No. X32**).

The KIR HLA Ligand primer set is unchanged compared to the previous lot.

## PRODUCT DESCRIPTION

### KIR HLA Ligand SSP typing

#### CONTENT

The primer set contains 5'- and 3'-primers for determining KIR HLA Ligands; HLA-Cw alleles with Asparagine or Lysine at position 80, HLA-B<sup>Bw4+</sup> alleles with Isoleucine or Threonine at position 80 and HLA-A<sup>Bw4+</sup> alleles.

#### PLATE LAYOUT

Each test consists of 5 PCR reactions in an 8 well cut PCR plate. Wells 6 to 8 are empty.

1	2	3	4	5	empty	empty	empty
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Wells 1 and 2: HLA-Cw KIR ligand primers

Wells 3 and 4: HLA-B KIR ligand primers

Well 5: HLA-A KIR ligand primers.

The 8 well cut PCR plate is marked with 'KIR Lig'.

Well No. 1 is marked with '26E'.

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

**Please note:** When removing each 8 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

The HLA-A, HLA-B and HLA-Cw alleles recognized by the HLA Nomenclature Committee in January 2008<sup>1</sup> have been considered in the Specificity and Interpretation Tables.

<sup>1</sup>HLA-A, HLA-B and HLA-Cw alleles listed on the IMGT/HLA web page 2008-January-11, release 2.20.0, [www.ebi.ac.uk/imgt/hla](http://www.ebi.ac.uk/imgt/hla).

## SPECIFICITY TABLE

### KIR HLA Ligand SSP typing

Specificities and sizes of the PCR products of the 5 primer mixes used for KIR HLA Ligand SSP.

Primer Mix	Size of spec. PCR product <sup>1</sup>	Size of control band <sup>2</sup>	KIR HLA Ligand epitope	Amplified HLA alleles
1	340 bp	800 bp	HLA-Cw <sup>Asparagine80</sup>	Cw*010201-0113, 0115-0119, 0212, 030201-0306, 0308-0314, 0316-0344, 0346, 0347, 0411, 0429, 0611, 070101-0706, 0708, 0710-0748, 0750-0753, 080101-0809, 0811-0815, 120201-120305, 1206-1208, 1210-1220, 140201-1403, 1405-1409, 1507, 1521, 160101-160103, 160401, 1606-1608, 1610
2	340 bp	800 bp	HLA-Cw <sup>Lysine80</sup>	Cw*0114, 020201-0211, 0213-0220, 0307, 0315, 0345, 04010101-040104, 0403-0410, 0412-0428, 0430, 0431, 050101-0517, 06020101-0610, 0612-0616N, 0707, 0709, 0749, 0810, 120401-1205, 1209, 1221, 1404, 150201-1506, 1508-1520, 1602, 1609, 1701-1704, 1801-1803
3	350 bp	800 bp	HLA-B <sup>Bw4+Threonine80</sup>	B*0802, 1301-1304, 1306-1308Q, 1310-1321, 1536, 1589, 9515, 1809, 2701, 3710, 380201-3804, 3808, 3815, 4047, 44020101-4405, 4407, 4408, 4410, 4412-4417, 4419N-4424, 4426-4445, 4447-4449, 4451-4459, 4704, 4902, 5309, 5311-5313, 5607
4	350 bp	1070 bp	HLA-B <sup>Bw4+Isoleucine80</sup>	B*0736, 0738, 0803, 1513, 1516-151702, 1523, 1524, 1567, 1587, 1595, 2702, 2730, 380101-380102,

				3805-3807, 3809-3814, 3816, 4013, 4019, 4406, 4418, 4425, 4450, 4818, 4901, 4903-4905, 510101- 5124, 5126-5146, 5148- 5152, 520101-5211, 530101-5302, 5304-5308, 5310, 5314, 5412, 5621, 570101-5711, 5713-5716, 5801, 5802, 5804-5816, 5901, 5902
<b>5</b>	450 bp	1070 bp	HLA-A <sup>Bw4+</sup>	A*0281, 0287, 9212, 9224, 9229, 9236, 2301-2317, 2319Q, 24020101-240302, 2405-2411N, 2413-2415, 2417, 2418, 2420-2425, 2427, 2429-2443, 2445N- 2484N, 250101-2506, 2913, 3107, 3108, 3110, 320101- 3215, 6836

<sup>1</sup>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 HLA Ligand 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.

<sup>2</sup>The 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-A\*01 subtyping. .

In addition, wells number 2 and 3 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.

## INTERPRETATION TABLE

### KIR HLA Ligand

	Well				
	1	2	3	4	5
Length of spec.	340	340	350	350	450
PCR product					
Length of int.	<b>800</b>	<b>800</b>	<b>800</b>	1070	1070
pos. control <sup>1</sup>					
5'-primer(s) <sup>2</sup>	1 <sup>st</sup>	1 <sup>st</sup>	1 <sup>st</sup>	1 <sup>st</sup>	28
	5'-CgA <sup>3'</sup>	5'-CgA <sup>3'</sup>	5'-CAG <sup>3'</sup>	5'-CAG <sup>3'</sup>	5'-TCg <sup>3'</sup>
					41
					5'-CTT <sup>3'</sup>
3'-primer <sup>3</sup>	302	302	309	309	317
	5'-ggC <sup>3'</sup>	5'-ggT <sup>3'</sup>	5'-gTg <sup>3'</sup>	5'-ATC <sup>3'</sup>	5'-ggA <sup>3'</sup>
Well No.	1	2	3	4	5
HLA allele					
Cw*010201-0113, 0115-0119, 0212, 030201-0306, 0308-0314, 0316-0344, 0346, 0347, 0411, 0429, 0611, 070101-0706, 0708, 0710-0748, 0750-0753, 080101-0809, 0811-0815, 120201-120305, 1206-1208, 1210-1220, 140201-1403, 1405-1409, 1507, 1521, 160101-160103, 160401, 1606-1608, 1610	1				
Cw*0114, 020201-0211, 0213-0220, 0307, 0315, 0345, 04010101-040104, 0403-0410, 0412-0428, 0430, 0531, 050101-0517, 06020101-0610, 0612-0616N, 0707, 0709, 0749, 0810, 120401-1205, 1209, 1221, 1404, 150201-1506, 1508-1520, 1602, 1609, 1701-1704, 1801-1803		2			
B*0802, 1301-1304, 1306-1308Q, 1310-1321, 1536, 1589, 9515, 1809, 2701, 3710, 380201-3804, 3808, 3815, 4047, 44020101-4405, 4407, 4408, 4410, 4412-4417, 4419N-4424, 4426-4445, 4447-4449, 4451-4459, 4704, 4902, 5309, 5311-5313, 5607	3				
B*0736, 0738, 0803, 1513, 1516-151702, 1523, 1524, 1567, 1587, 1595, 2702, 2730, 380101-380102, 3805-3807, 3809-3814, 3816, 4013, 4019, 4406, 4418, 4425, 4450, 4818, 4901, 4903-4905, 510101-5124, 5126-5146, 5148-5152, 520101-5211, 530101-5302, 5304-5308, 5310, 5314, 5412, 5621, 570101-5711, 5713-5716, 5801, 5802, 5804-5816, 5901, 5902				4	
A*0281, 0287, 9212, 9224, 9229, 9236, 2301-2317, 2319Q, 2402020101-240302, 2405-2411N, 241301-2415, 2417, 2418, 2420-2425, 2427, 2429-2443, 2445N-2484N, 250101-2506, 2913, 3107, 3108, 3110, 320101-3215, 6836	5				
HLA allele					
Well No.	1	2	3	4	5

<sup>1</sup>The 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-A\*01 subtyping. .

In addition, wells number 2 and 3 contain the primer pair giving rise to the shorter, 800 bp, internal positive control band in order to allow kit identification.

<sup>2</sup>The nucleotide position, in the 1<sup>st</sup> exon or the 1st intron, matching the specificity-determining 3'-end of the primer is given. Nucleotide numbering as on the [www.ebi.ac.uk/imgt/hla](http://www.ebi.ac.uk/imgt/hla) web site. The sequence of the 3 terminal nucleotides of the primer is given.

<sup>3</sup>The nucleotide position, in 3<sup>rd</sup> exon, matching the specificity-determining 3'-end of the primer is given in the anti-sense direction. Nucleotide numbering as on the [www.ebi.ac.uk/imgt/hla](http://www.ebi.ac.uk/imgt/hla) web site. The sequence of the 3 terminal nucleotides of the primer is given.

<b>CELL LINE VAL. SHEET</b>							
<b>KIR HLA Ligand primer set</b>							
			<b>Well</b>				
			<b>1</b>	<b>2</b>	<b>3</b>	<b>4</b>	<b>5</b>
			200843101	200843102	200843103	200843104	200843105
<b>IHWC cell line</b>							
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<sup>®</sup> KIR HLA Ligand SSP**

**Product number:** 104.201-12 – including *Taq* polymerase  
104.201-12u – without *Taq* polymerase  
**Lot number:** 26E  
**Expiry date:** 2010-February-01  
**Number of tests:** 12  
**Number of wells per test:** 5

#### **Well specifications:**

Well No.	Production No.
1	2008-431-01
2	2008-431-02
3	2008-431-03
4	2008-431-04
5	2008-431-05

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

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

**Date of approval:** 2008-February-15

**Approved by:**

**Quality Control, Supervisor**



Lot No.: **26E**

Lot-specific information

www.olerup.com

## Declaration of Conformity

**Product name:** *Olerup* SSP<sup>®</sup> KIR HLA Ligand  
**Product number:** 104.201-12, 104.201-12u  
**Lot number:** 26E

**Intended use:** Determination of HLA-Cw, HLA-B<sup>Bw4+</sup> and HLA-A<sup>Bw4+</sup> KIR ligand sequence motifs.

**Manufacturer:** *Olerup* SSP AB  
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**Fax:** +46-8-717 88 18

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-February-15

Olle Olerup  
Managing Director

KIR HLA Ligand  
104.201-12 – including *Taq* polymerase  
104.201-12u – without *Taq* polymerase

Product Insert

Page 10 of 12

Lot No.: **26E**

Lot-specific information

[www.olerup.com](http://www.olerup.com)





KIR HLA Ligand  
104.201-12 – including *Taq* polymerase  
104.201-12u – without *Taq* polymerase

Product Insert

Page 12 of 12

Lot No.: **26E**

Lot-specific information

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