101.841-12 – including *Taq* polymerase, IFU-01 101.841-12u – without *Taq* polymerase, IFU-02 Visit <u>www.olerup.com</u> for "Instructions for Use" (IFU)

Lot No.: 0H7

Lot-specific information www.olerup-ssp.com *Olerup* SSP<sup>®</sup> HLA-A\*24:09N

Product number:	101.841-12 – including <i>Taq</i> polymerase 101.841-12u – without <i>Taq</i> polymerase
Lot number:	0H7
Expiry date:	2021-04-01
Number of tests:	12
Number of wells per test:	2+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. 0H7.

Complete product documentation consists of generic Instructions for Use (IFU), lot specific Product Insert, Worksheet and Certificate.

## CHANGES COMPARED TO THE PREVIOUS OLERUP SSP® HLA-A\*24:09N LOT (2E4)

The HLA-A\*24:09N specificity and interpretation tables have been updated compared the previous *Olerup* SSP<sup>®</sup> HLA-A\*24:09N lot (Lot No. 2E4). The kit design is based on IMGT/HLA database 3.33.0.

The HLA-A\*24:09N primer set is unchanged compared to the previous *Olerup* SSP<sup>®</sup> HLA-A\*24:09N (Lot No. 2E4).

101.841-12 – including *Taq* polymerase, IFU-01 101.841-12u – without *Taq* polymerase, IFU-02 Visit <u>www.olerup.com</u> for "Instructions for Use" (IFU)

## Lot No.: **0H7** Lot-specific information www.olerup-ssp.com

Well **3** contains <u>Negative Control primer pairs</u>, that will amplify more than 95% of the *Olerup* SSP<sup>®</sup> HLA Class I, DRB, DQB1, DPB1 and DQA1 amplicons as well as all the amplicons generated by the control primer pairs matching the human growth hormone gene.

HLA-specific PCR product sizes range from 75 to 200 base pairs. The PCR product generated by the positive control primer pair is 430 base pairs.

Length of PCR	105	200	105	80	75	80	85
product							
5'-primer <sup>1</sup>	164	340	440	45	45	43	36
	5'-CAC3'	<sup>5</sup> '-Agg <sup>3'</sup>	<sup>5'</sup> -TTA3'	<sup>5′</sup> -Tgg <sup>3′</sup>	<sup>5</sup> '-Tgg <sup>3'</sup>	<sup>5′</sup> -Tgg <sup>3′</sup>	<sup>5'</sup> -TAC <sup>3'</sup>
							36
							<sup>5'</sup> -TAT <sup>3'</sup>
3'-primer <sup>2</sup>	231	2 <sup>nd</sup> I	507	59	58	57	47
	<sup>5'</sup> -TgC <sup>3'</sup>	<sup>5'</sup> -AAA <sup>3'</sup>	<sup>5′</sup> -TTg³′	<sup>5</sup> '-CTC <sup>3'</sup>	<sup>5'</sup> -ggC <sup>3'</sup>	<sup>5'</sup> -CTC <sup>3'</sup>	<sup>5'</sup> -ACA <sup>3'</sup>
							48
							<sup>5'</sup> -gCA <sup>3'</sup>
							48
							<sup>5'</sup> -gCC <sup>3'</sup>
							52
							<sup>5′</sup> -TgT³′
A*	+	+	+				
B*	+	+	+				
C*	+	+	+				
DRB1				+	+		
DRB3				+	+		
DRB5				+			
DQB1					+		
DPB1						+	
DQA1							+

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

<sup>2</sup>The nucleotide position for HLA class I genes and the codon for HLA class II genes, in the 2<sup>nd</sup> or 3<sup>rd</sup> exon or the 2<sup>nd</sup> intron, matching the specificity-determining 3'-end of the primer is given in the anti-sense direction.

Nucleotide and codon numbering as on the <u>www.ebi.ac.uk/imgt/hla</u> web site. The sequence of the 3 terminal nucleotides of the primer is given.

Visit www.olerup.com for

**101.841-12 – including Taq polymerase**, IFU-01 **101.841-12u – without Taq polymerase**, IFU-02

Lot No.: **0H7** 

Lot-specific information w

www.olerup-ssp.com

"Instructions for Use" (IFU)

# PRODUCT DESCRIPTION

# HLA-A\*24:09N SSP subtyping

## CONTENT

The primer set contains 5'- and 3'-primers for identifying the HLA-A\*24:09N allele.

## PLATE LAYOUT

Each test consists of 3 PCR reactions in an 8 well cut PCR plate. Wells 4 to 8 are empty.

1 2 NC empty empty empty empty empty

The 8 well cut PCR plate is marked with the Lot No. '0H7' in silver/gray ink. Well No. 1 is marked with the Lot No. '0H7'.

Wells 1 to 2 – HLA-A\*24:09N high resolution primers.

Well 3 – Negative Control (NC).

A faint row of numbers is seen between wells 1 and 2 or wells 7 and 8 of the PCR trays. These stem from the manufacture of the trays, and should be disregarded. The PCR plates are heat-sealed with a PCR-compatible foil.

**Please note:** When removing each 8 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-A\*24:09N SSP subtypings will be influenced by the A\*02:17:01<sup>w</sup>-02:17:04<sup>w</sup>, the A\*11:139, the A\*23:14:01-23:14:02, most A\*24, the A\*26:16, A\*33:19, A\*33:119, A\*68:45 and the A\*68:117 alleles.

### UNIQUELY IDENTIFIED ALLELES

The HLA-A\*24:09N allele will give rise to a unique amplification pattern by the primers in the HLA-A\*24:09N kit<sup>1,2</sup>.

<sup>1</sup>HLA-A alleles listed on the IMGT/HLA web page 2018-July-11, release 3.33.0, <u>www.ebi.ac.uk/imgt/hla</u>. <sup>2</sup>Alleles that have been deleted from or renamed in the official WHO HLA Nomenclature up to and including the last IMGT/HLA database release can be retrieved from web page

http://hla.alleles.org/alleles/deleted.html.

Visit www.olerup.com for

**101.841-12 – including Taq polymerase**, IFU-01 **101.841-12u – without Taq polymerase**, IFU-02

Lot No.: **0H7** 

Lot-specific information
SPECIFICITY TABLE

www.olerup-ssp.com

"Instructions for Use" (IFU)

# HLA-A\*24:09N SSP subtyping

Specificities and sizes of the PCR products of the 2+1 primer mixes used for HLA-A\*24:09N SSP subtyping

Primer Mix	Size of spec. PCR product <sup>1</sup>	Size of control band <sup>2</sup>	Amplified HLA-A alleles <sup>3</sup>
1	370 bp	800 bp	*24:09N
2	175 bp, 205 bp	1070 bp	*02:17:01 <sup>w</sup> -02:17:04 <sup>w</sup> , 11:139, 23:14:01-23:14:02, 24:02:01:01- 24:11N, 24:13:01-24:13:02, 24:17- 24:50, 24:54-24:56, 24:58-24:63, 24:66-24:91, 24:93, 24:95-24:113, 24:115-24:137, 24:139-24:187, 24:189-24:210, 24:212-24:221, 24:223-24:227, 24:229-24:290, 24:292-24:295, 24:297-24:303N, 24:305-24:315, 24:317-24:323N, 24:325-24:405, 26:16, 33:19, 33:119, 68:45, 68:117
<b>3</b> <sup>4</sup>	-	-	Negative Control

<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 HLA-A\*24:09N SSP typings.

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 inherit 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 internal positive control bands are 1070 or 800 base pairs respectively, well distribution as outlined in the table. Well number 1 contains the shorter, 800 bp, internal positive control band. The well distribution of the internal controls can help in orientation of the kit on gel photo, as well as allow for kit identification. In the presence of a specific amplification the intensity of the control band often decreases.

<sup>3</sup>For several HLA Class I alleles 1<sup>st</sup> and/or 4<sup>th</sup> exon(s) and beyond, as well as intron nucleotide sequences, are not available. In these instances it is not known whether some of the primers of the SSP sets are completely matched with the target sequences or not. Assumption is made that unknown sequences in these regions are conserved within allelic groups.

<sup>4</sup>Primer mix 3 contains a negative control, which will amplify more than 95% of HLA amplicons as well as the amplicons generated by the control primer pairs matching the human growth hormone gene. HLA-specific PCR product sizes range from 75 to 200 base pairs and the PCR product generated by the HGH positive control primer pair is 430 base pairs. 'w', may be weakly amplified.



Visit www.olerup.com for

www.olerup-ssp.com

**101.841-12 – including Taq polymerase**, IFU-01 **101.841-12u – without Taq polymerase**, IFU-02

"Instructions for Use" (IFU)

Lot No.: **0H7** 

## Lot-specific information PRIMER SPECIFICATION

Well No.	1	2
Length of spec.	370	175
PCR product		205
Length of int.	800	1070
pos. control <sup>1</sup>		
5'-primer(s) <sup>2</sup>	742	98
	5' -ACT 3'	<sup>5'</sup> -CTC <sup>3'</sup>
		368
		<sup>5'</sup> -gTT <sup>3'</sup>
3'-primer(s) <sup>3</sup>	971	259
	<sup>5'</sup> -CAg <sup>3'</sup>	
		502
		<sup>5'</sup> -CTT <sup>3'</sup>
		539
		<sup>5'</sup> -TCT <sup>3'</sup>
Well No.	1	2

<sup>1</sup>The internal positive control primer pairs amplify segments of the human growth hormone gene. The internal positive control bands are 1070 or 800 base pairs respectively, well distribution as outlined in the table. Well number 1 contains the shorter, 800 bp, internal positive control band. The well distribution of the internal controls can help in orientation of the kit on gel photo, as well as allow for kit identification. In the presence of a specific amplification the intensity of the control band often decreases.

<sup>2</sup>The nucleotide position matching the specificity-determining 3'-end of the primer is given. Nucleotide numbering as on the <u>www.ebi.ac.uk/imgt/hla</u> web site. The sequence of the 3 terminal nucleotides of the primer is given.

<sup>3</sup>The nucleotide position matching the specificity-determining 3'-end of the primer is given in the anti-sense direction. Nucleotide numbering as on the <u>www.ebi.ac.uk/imgt/hla</u> web site. The sequence of the 3 terminal nucleotides of the primer is given.



101.841-12 – including *Taq* polymerase, IFU-01 101.841-12u – without *Taq* polymerase, IFU-02

Lot No.: **0H7** 

Lot-specific information

"Instructions for Use" (IFU)

Visit <u>www.olerup.com</u> for

www.olerup-ssp.com

		ILA-A24:0	914 22L	' KIT		
					W	ell
					1	2
				Prod No.:	201899001	201899002
	IHW	/C cell line <sup>1</sup>	<b>A</b> *	<b>A</b> *		
1	9001	SA	*24:02		-	+
2	9280	LK707	*02:01		-	-
3	9011	E4181324	*01:01		-	-
4	9275	GU373	*30:01		-	-
5	9009	KAS011	*01:01		-	-
6	9353	SM	*02:01	*26:03	-	-
7	9020	QBL	*26:01		-	-
8	9025	DEU	*31:01		-	-
9		YAR	*26:01		-	-
10		LKT3	*24:02		- 1	+
11		PITOUT	*29:02		-	-
12	9052		*02:01		- 1	-
13		JESTHOM	*02:01		1_	-
14		OLGA	*31:01		_	_
15			*24:02		_	-
_	9075				-	+
16		SWEIG007	*29:02	*00.04	-	-
17		CTM3953540	*03:01	*80:01	-	-
18		32367	*33:03	*74:01	-	-
19		BM16	*02:01		-	-
20		SLE005	*02:01		-	-
21		AMALA	*02:17		-	W
22	9056	KOSE	*02:01		-	-
23	9124	IHL	*02:01	*34:01	-	-
24	9035	JBUSH	*32:01		-	-
25	9049	IBW9	*33:01		-	-
26	9285	WT49	*02:05		-	-
27	9191	CH1007	*24:10	*29:01	-	+
28	9320	BEL5GB	*02:01	*29:02	-	-
29		MOU	*29:02		-	-
30	9021	RSH	*30:01	*68:02	- 1	-
31		DUCAF	*30:02		-	-
32		HAG	*02:01		- 1	-
33		MT14B	*31:01		-	-
33 34	9104		*31:01		1_	-
34 35		SSTO	*32:01			-
				*11.04	-	-
36		KT17	*02:06	*11:01	-	-
37		HHKB	*03:01			•
38	9099		*02:17		-	w
39	9315		*01:01	*03:01	-	-
40		WHONP199	*02:07	*30:01	-	-
41	9055	H0301	*03:01		-	-
42	9066	TAB089	*02:07		-	-
43	9076	T7526	*02:06	*02:07	-	-
44	9057	TEM	*66:01		-	-
45	9239	SHJO	*23:01	*24:02	-	+
46		SCHU	*03:01		-	-
47		TUBO	*02:16	*03:01	- 1	-
48		TER-ND	*02:01	*11:01	L_	

#### HLA-A\*24:09N

#### **Product Insert**

#### 101.841-12 – including *Taq* polymerase, IFU-01 101.841-12u – without *Taq* polymerase, IFU-02

Visit <u>www.olerup.com</u> for "Instructions for Use" (IFU)

#### Lot No.: 0H7

#### Lot-specific information

#### www.olerup-ssp.com

<sup>1</sup>The provided cell line HLA specificities are retrieved from the <u>http://www.ihwg.org/hla</u> web site. The specificity of an individual cell line may thus be subject to change.

<sup>2</sup>The specificity of each primer solution in the kit has been tested against 48 well characterized cell line DNAs and where applicable, additional cell line DNAs.

Additional primers in primer mix 2 were tested by separately adding one additional 5'-primer, respectively one additional 3'-primer.



101.841-12 – including *Taq* polymerase, IFU-01 101.841-12u – without *Taq* polymerase, IFU-02 Visit <u>www.olerup.com</u> for "Instructions for Use" (IFU)

Lot No.: 0H7 ADDRESSES:

Lot-specific information

www.olerup-ssp.com

Manufacturer: Olerup SSP AB, Franzengatan 5, SE-112 51 Stockholm, Sweden. Tel: +46-8-717 88 27 Fax: +46-8-717 88 18 E-mail: olerup-se@caredx.com Web page: http://www.olerup.com

Distributed by: Olerup GmbH, Löwengasse 47 / 6, AT-1030 Vienna, Austria. *Tel:* +43-1-710 15 00 *Fax:* +43-1-710 15 00 10 *E-mail:* <u>olerup-at@caredx.com</u> *Web page:* <u>http://www.olerup.com</u>

Olerup Inc., 901 S. Bolmar St., Suite R, West Chester, PA 19382 *Tel:* 1-877-OLERUP1 *Fax:* 610-344-7989 *E-mail:* <u>olerup-us@caredx.com</u> *Web page:* <u>http://www.olerup.com</u>

For information on Olerup distributors worldwide, contact Olerup GmbH.

