

101.331-24/06 – including *Taq* polymerase101.331-24u/06u – without *Taq* polymeraseLot No.: **11E**

Lot-specific information

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Olerup SSP[®] DPA1

Product number: 101.331-24/06 – including *Taq* pol.
101.331-24u/06u – without *Taq* pol.

Lot number: 11E

Expiry date: 2010-February-01

Number of tests: 24 test – Product No. 101.331-24
6 tests – Product No. 101.331-06

Number of Wells per test: 16

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. 11E.

CHANGES COMPARED TO THE PREVIOUS OLERUP SSP[®] DPA1 LOT

The DPA1 specificity and interpretation tables have been updated for the DPA1 alleles described since the previous *Olerup SSP[®] DPA1* lot was made (**Lot No. X42**).

The primers of the Wells detailed below have been added, exchanged or modified.

Well	5'-primer	3'-primer	rationale
6	-	Added	Primer added for the DPA1*0204 allele.
16	Modiified	-	Increased specificity of specific primer pair.

PRODUCT DESCRIPTION

DPA1 SSP subtyping

CONTENT

The primer set contains 5'- and 3'-primers for identifying the DPA1*0103 to DPA1*0401 alleles.

PLATE LAYOUT

Each test consists of 16 PCR reactions in a 16 well cut PCR plate.

1	2	3	4	5	6	7	8
9	10	11	12	13	14	15	16

The 16 well cut PCR plate is marked with 'DPA1'.

Well No. 1 is marked with '11E'.

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

Only DPA1 alleles will be amplified by the DPA1 typing kit. Thus, the interpretation of DPA1 typings is not influenced by the DPA2 gene.

UNIQUELY IDENTIFIED ALLELES

All the phenotypically different DPA1 alleles, i.e. **DPA1*0103 to DPA1*0109, DPA1*0201 to DPA1*0204, DPA1*0301 to DPA1*0303 and DPA1*0401**, recognized by the HLA Nomenclature Committee in January 2008¹ will give rise to unique amplification patterns by the primers in the DPA1 typing kit.

The DPA1 typing kit cannot distinguish the DPA1*010301 and DPA1*010302 alleles, the DPA1*020101 to DPA1*020106 alleles and the DPA1*020201 and DPA1*020203 alleles.

¹DPA1 alleles listed on the IMGT/HLA web page 2008-January-11, release 2.20.0, www.ebi.ac.uk/imgt/hla.

RESOLUTION IN HOMO- AND HETEROZYGOTES

The 15 DPA1 alleles give rise to 16 different amplification patterns, that can be combined in 136 homozygous and heterozygous combinations. Fourteen of these genotypes do not give rise to unique amplification patterns.

++++----	-++-+-	0103, 0303 = 0104, 0301
+++-----	-----+	0103, 0107 = 0107, 0107
+++-----	-----+	0103, 0109 = 0109, 0109
++-+----	-----+	0104, 0108 = 0108, 0108
+---+----	---+----	0105, 0401 = 0401, 0401
-++-++++	+-----	0106, 0202 = 0106, 0204
----+++-	+-----	0202, 0204 = 0204, 0204

0103 = 010301 and 010302

SPECIFICITY TABLE**DPA1 SSP typing**

Specificities and sizes of the PCR products of the 16 primer mixes used for DPA1 SSP typing

Primer Mix	Size of spec. PCR product ¹	Size of control band ²	Amplified DPA1 ³ alleles
1⁴	85 bp	515 bp	010301-010302, 0104, 0105, 0107, 0108, 0109, 0401
2	250 bp	515 bp	010301-0104, 0106-0109
3	200 bp	430 bp	010301-010303, 0106, 0107, 0109, 0301, 0302
4⁴	115 bp	430 bp	0104, 0108, 0303
5⁴	105 bp	430 bp	0105, 020101-0204, 0401
6⁵	155, 195 bp	515 bp	0106, 0204
7⁴	100 bp	430 bp	0106, 020101-020106
8⁴	100 bp	430 bp	020201-020203, 0204
9	205 bp	430 bp	020201-020203, 0204, 0302
10⁴	85 bp	515 bp	010303, 0203, 0301-0303
11⁴	90 bp	515 bp	0301, 0303
12	205 bp	430 bp	0401
13	135 bp	430 bp	0107
14	140 bp	430 bp	0108
15	245 bp	430 bp	010303, 0301-0303
16	220 bp	430 bp	0109

¹Alleles 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 DR low resolution 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.

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 430 base pairs, for most wells, or a band of 515 base pairs, for some Wells.

Well number 1 contains the primer pair giving rise to the longer, 515 bp, internal positive control

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band in order to help in the correct orientation of the DPA1 typing.

In addition, wells number 2, 6, 10 and 11 contain the primer pair giving rise to the longer, 515 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.

³For some DPA1 alleles only partial 2nd exon nucleotide sequences are 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. We assume that unknown sequences are conserved within allelic groups.

⁴Specific PCR fragments shorter than 125 bp are less intense and less sharp compared to longer specific PCR fragments.

⁵Primer mix 6: Specific PCR fragment of 155 bp in the DPA1*0204 allele. Specific PCR fragment of 195 bp in the DPA1*0106.

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INTEPRETATION TABLE								
DPA1 SSP typing								
Amplification patterns of the DPA1 alleles								
	Well⁵							
	1	2	3	4	5	6	7	8
Length of spec.	85	250	200	115	105	155	100	100
PCR product						195		
Length of int.	515	515	430	430	430	515	430	430
pos. control¹								
5'-primer(s)²	15	11	28	4	84	31	11	11
	5'-ACg ^{3'}	5'-C gC ^{3'}	5'-gAA ^{3'}	5'-Cg g ^{3'}	5'-AAT ^{3'}	5'-g CA ^{3'}	5'-C gC ^{3'}	5'-C AT ^{3'}
3'-primer(s)³	31	83	83	28	intr⁶	69	31	31
	5'-CAT ^{3'}	5'-ggT ^{3'}	5'-ggT ^{3'}	5'-TC g ^{3'}	5'-ggC ^{3'}	5'-gTC ^{3'}	5'-CTg ^{3'}	5'-CTg ^{3'}
						83		
						5'-ggT ^{3'}		
Well No.	1	2	3	4	5	6	7	8
DPA1 allele⁴								
*010301-010302	1	2	3					
*010303		2	3					
*0104	1	2		4				
*0105	1				5			
*0106		2	3			6	7	
*0107	1	2	3					
*0108	1	2		4				
*0109	1	2	3					
*020101-020106					5		7	
*020201-020203					5			8
*0203					5			
*0204					5	6		8
*0301			3					
*0302			3					
*0303				4				
*0401	1				5			
DPA1 allele⁴								
Well No.	1	2	3	4	5	6	7	8

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INTERPRETATION TABLE								
DPA1 SSP typing								
Amplification patterns of the DPA1 alleles								
Well ⁵								
9	10	11	12	13	14	15	16	
205	85	90	205	135	140	245	220	Length of spec.
								PCR product
430	515	515	430	430	430	430	430	Length of int.
								pos. control ¹
11	15	66	18	51	50	15	23	5'-primer(s) ²
5'-C AT ^{3'}	5'-ACC ^{3'}	5'-A TC ^{3'}	5'-gA A ^{3'}	5'-CA T ^{3'}	5'-C Cg ^{3'}	5'-ACC ^{3'}	5'-T AC ^{3'}	
	15					15		
	5'-ACC ^{3'}					5'-ACC ^{3'}		
66	31	83	73	83	83	83	83	3'-primer(s) ³
5'-T CA ^{3'}	5'-CAT ^{3'}	5'-ggT ^{3'}	5'-AgC ^{3'}	5'-ggT ^{3'}	5'-ggT ^{3'}	5'-ggT ^{3'}	5'-ggT ^{3'}	
9	10	11	12	13	14	15	16	Well No.
								DPA1 allele ⁴
								*010301-010302
	10					15		*010303
								*0104
								*0105
								*0106
				13				*0107
					14			*0108
							16	*0109
								*020101-020106
9								*020201-020203
	10							*0203
9								*0204
	10	11				15		*0301
9	10					15		*0302
	10	11				15		*0303
			12					*0401
								DPA1 allele ⁴
9	10	11	12	13	14	15	16	Well No.

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¹The 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 430 base pairs, for most wells, or a band of 515 base pairs, for some wells.

Well number 1 contains the primer pair giving rise to the longer, 515 bp, internal positive control band in order to help in the correct orientation of the DPA1 typing.

In addition, wells number 2, 6, 10 and 11 contain the primer pair giving rise to the longer, 515 bp, internal positive control band in order to allow kit identification.

²The codon, in the 2nd exon, matching the specificity-determining 3'-end of the primer is given. 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. Empty spaces indicate codon boundaries.

³The codon, in the 2nd exon, matching the specificity-determining 3'-end of the primer is given in the anti-sense direction. 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. Empty spaces indicate codon boundaries.

⁴The sequence of the DPA1*0101 allele has been shown to be identical to DPA1*0103.

The sequence of the DPA1*0102 allele has been shown to be identical to DPA1*0103.

⁵Primer mix 6: Specific PCR fragment of 155 bp in the DPA1*0204 allele. Specific PCR fragment of 195 bp in the DPA1*0106.

⁶'intr', matching sequences in the 2nd intron.

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CELL LINE VALIDATION SHEET																				
DPA1 SSP kit																				
					Well															
					1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
				Lot No.:	200730601	200730602	200730603	200730604	200730605	200841306	200730607	200730608	200730609	200730610	200730611	200730612	200730613	200730614	200730615	200841316
	IHWC cell line		DPA1																	
1	9001	SA	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
2	9280	LK707	*0201	*0202	-	-	-	-	+	-	+	+	+	-	-	-	-	-	-	-
3	9011	E4181324	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
4	9275	GU373	*0201	*0401	+	-	-	-	+	-	+	-	-	-	-	+	-	-	-	-
5	9009	KAS011	*0103	*0201	+	+	+	-	+	-	+	-	-	-	-	-	-	-	-	-
6	9353	SM	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
7	9020	QBL	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
8	9007	DEM	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
9	9026	YAR	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
10	9107	LKT3	*0202		-	-	-	-	+	-	-	+	+	-	-	-	-	-	-	-
11	9051	PITOUT	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
12	9052	DBB	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
13	9067	BTB	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
14	9071	OLGA	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
15	9075	DKB	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
16	9037	SWEIG007	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
17	9008	WILJON	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
18	9257	32367	*0103	*0301	+	+	+	-	-	-	-	-	-	+	+	-	-	-	+	-
19	9038	BM16	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
20	9059	SLE005	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
21	9064	AMALA	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
22	9056	KOSE	*0103	*0201	+	+	+	-	+	-	+	-	-	-	-	-	-	-	-	-
23	9124	IHL	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
24	9035	JBUSH	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
25	9049	IBW9	*0201		-	-	-	-	+	-	+	-	-	-	-	-	-	-	-	-
26	9285	WT49	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
27	9191	CH1007	*0103	*0401	+	+	+	-	+	-	-	-	-	-	-	+	-	-	-	-
28	9320	BEL5GB	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
29	9050	MOU	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
30	9021	RSH	*0202	*0301	-	-	+	-	+	-	-	+	+	+	+	-	-	-	+	-
31	9019	DUCAF	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
32	9297	HAG	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
33	9098	MT14B	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
34	9104	DHIF	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
35	9302	SSTO	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
36	9024	KT17	*0202		-	-	-	-	+	-	-	+	+	-	-	-	-	-	-	-
37	9065	HHKB	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
38	9099	LZL	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
39	9315	CML	*0103	*0201	+	+	+	-	+	-	+	-	-	-	-	-	-	-	-	-
40	9134	WHONP199	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
41	9055	H0301	*0201		-	-	-	-	+	-	+	-	-	-	-	-	-	-	-	-
42	9066	TAB089	*0202		-	-	-	-	+	-	-	+	+	-	-	-	-	-	-	-
43	9076	T7526	*0401		+	-	-	-	+	-	-	-	-	-	-	+	-	-	-	-
44	9057	TEM	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
45	9239	SHJO	*0103	*0301	+	+	+	-	-	-	-	-	-	+	+	-	-	-	+	-
46	9013	SCHU	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
47	9045	TUBO	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-
48	9303	TER-ND	*0103		+	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-

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CERTIFICATE OF ANALYSIS

Olerup SSP® DPA1 SSP

Product number:

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101.331-24u/06u – without *Taq* pol.

Lot number:

11E

Expiry date:

2010-February-01

Number of tests:

24 test – Product No. 101.331-24

6 tests – Product No. 101.331-06

Number of Wells per test:

16

Well specifications:

Well No.	Production No.	Well No.	Production No.
1	2007-306-01	9	2007-306-09
2	2007-306-02	10	2007-306-10
3	2007-306-03	11	2007-306-11
4	2204-306-04	12	2007-306-12
5	2007-306-05	13	2007-306-13
6	2008-413-06	14	2007-306-14
7	2007-306-07	15	2007-306-15
8	2007-306-08	16	2008-413-16

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

No DNAs carrying the allele to be amplified by primer solutions 6, 13, 14 and 16 were available. The specificities of the primers in primer solutions 6 and 14 were tested by separately adding one additional 5'-primer and one additional 3'-primer, respectively. In primer solutions 13 and 16 it was only possible to test the 3'-primers, the 5'-primers were not possible to test.

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

Date of approval: 2008-February-08

Approved by:

Approved by:

Quality Control, Supervisor

Declaration of Conformity

Product name: Olerup SSP[®] DPA1
Product number: 101.331-24/06, 101.331-24u/06u
Lot number: 11E

Intended use: HLA-DPA1 high resolution histocompatibility testing

Manufacturer: Olerup SSP AB
Hasselstigen 1
SE-133 33 Saltsjöbaden, Sweden
Phone: +46-8-717 88 27
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, ISO 17025:1999 and ISO 13485:2000, following the provisions of the 98/79/EC Directive on *in vitro* diagnostic medical devices, Annex III.

The Technical Construction 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-08

Olle Olerup
Managing Director

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www.olerup.com**ADDRESSES:****Manufacturer:****Olerup SSP AB**, Hasselstigen 1, SE-133 33 Saltsjöbaden, Sweden.**Tel:** +46-8-717 88 27**Fax:** +46-8-717 88 18**E-mail:** olle.olerup@ssp-primers.se**Distributed by:****QIAGEN Vertriebs GmbH**, Löwengasse 47 / 6, AT-1030 Vienna, Austria.**Tel:** +43-1-710 15 00**Fax:** +43-1-710 15 00 10**E-mail:** office-at@qiagen.com**Web page:** <http://www.qiagen.com/goto/HLA>**Qiagen Inc.**, 27220 Turnberry Lane, Suite 200, Valencia, CA 91355**Customer Care:** 800-426-8157**Technical Service:**, 800-DNA-PREP (888-362-7737)**Fax:** 800-718-2056**Customer Care (Ordering):** customercare-us@qiagen.com**Literature Request:** literature-us@qiagen.com**Web page:** <http://www.qiagen.com/goto/HLA>

For information on *Olerup* SSP distributors worldwide, contact **QIAGEN Vertriebs GmbH**.