Evaluation of the automated HISTO SPOT HLA AB test in comparison to a Luminex SA Test

Institute for Transfusion Medicine, Hanover Medical School
Ing. Murielle Verboom

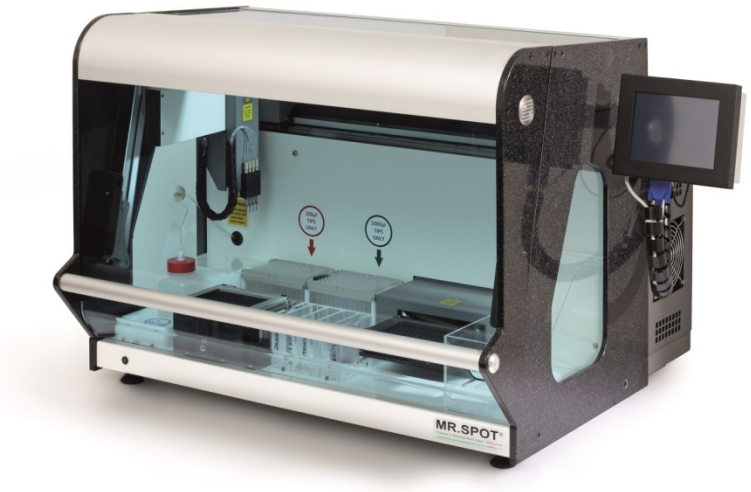
Medizinische Hochschule Hannover

Aim

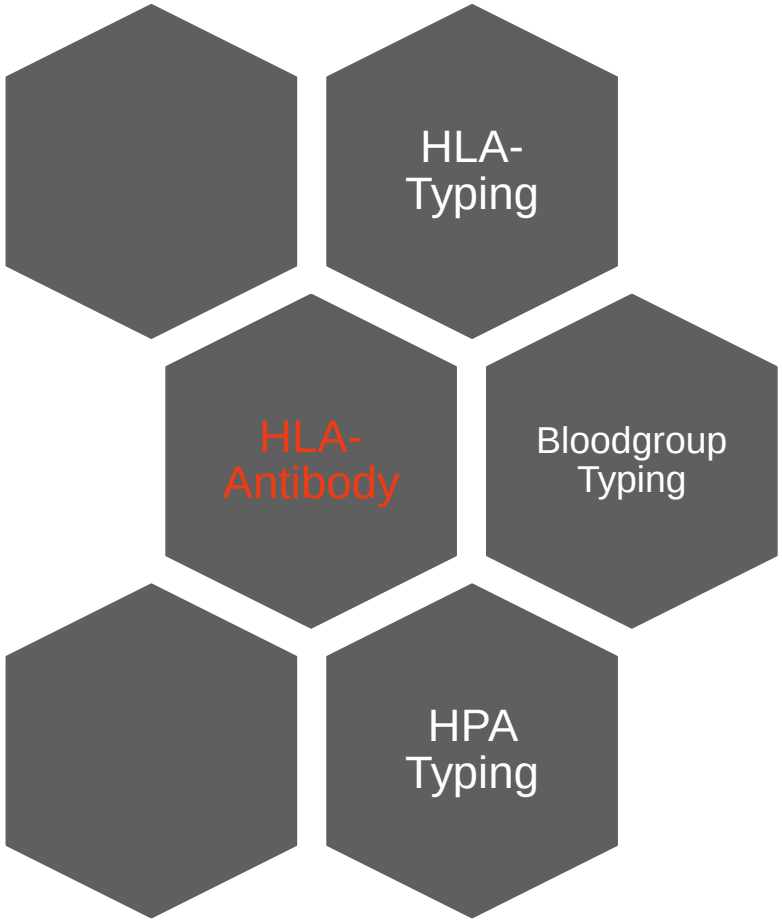
Aim of the current study was the evaluation of the automated HISTO SPOT HLA AB (BAG Health Care) test in comparison to a Luminex Single Antigen Test

Mr. SPOT®

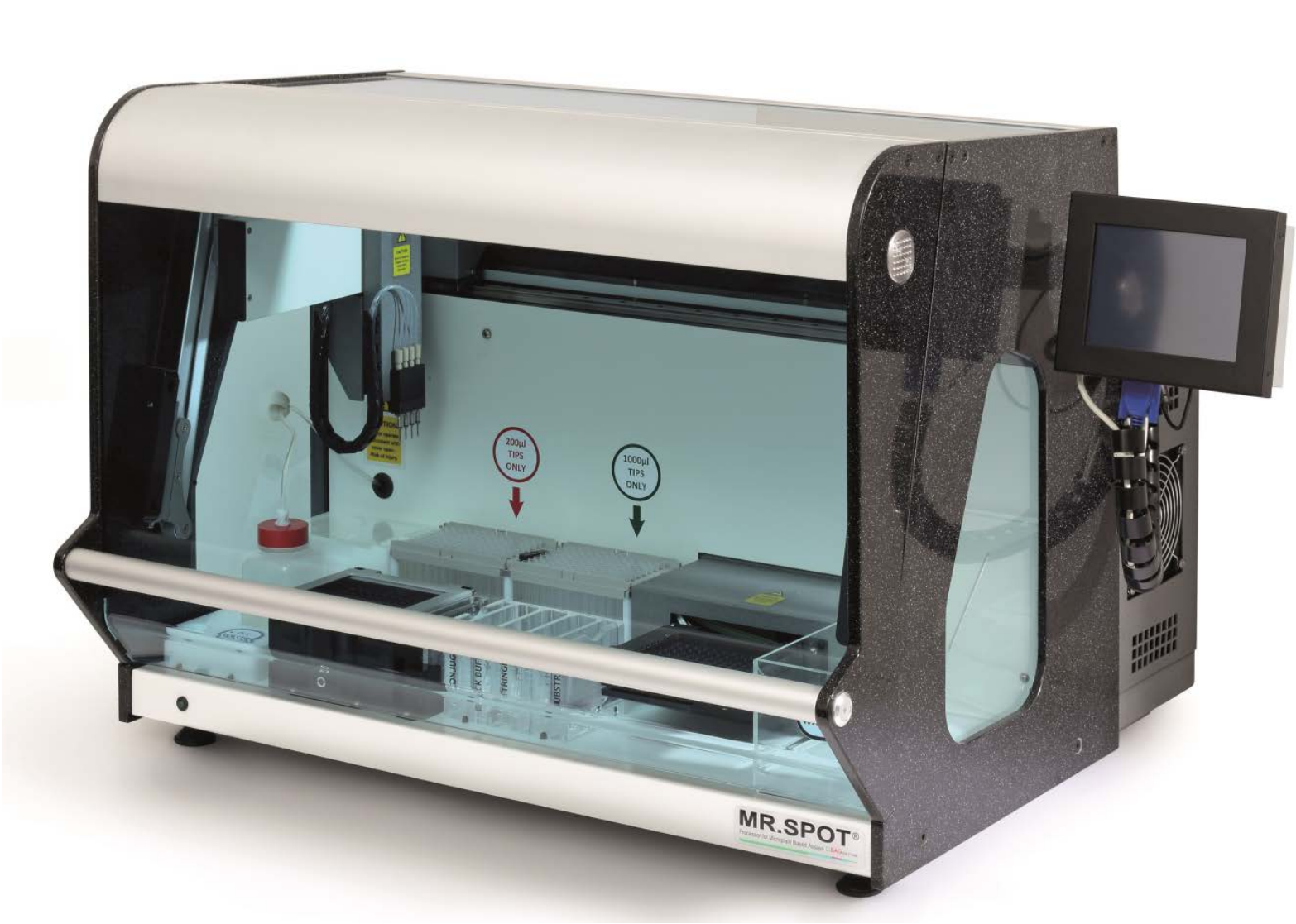
Automated PCR-SSO system for antigen typing and antibody detection



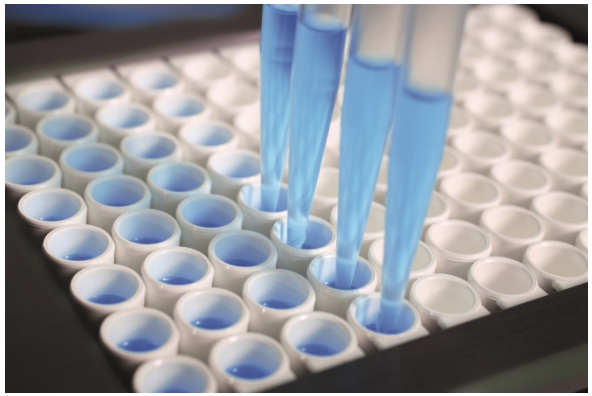
Product Overview



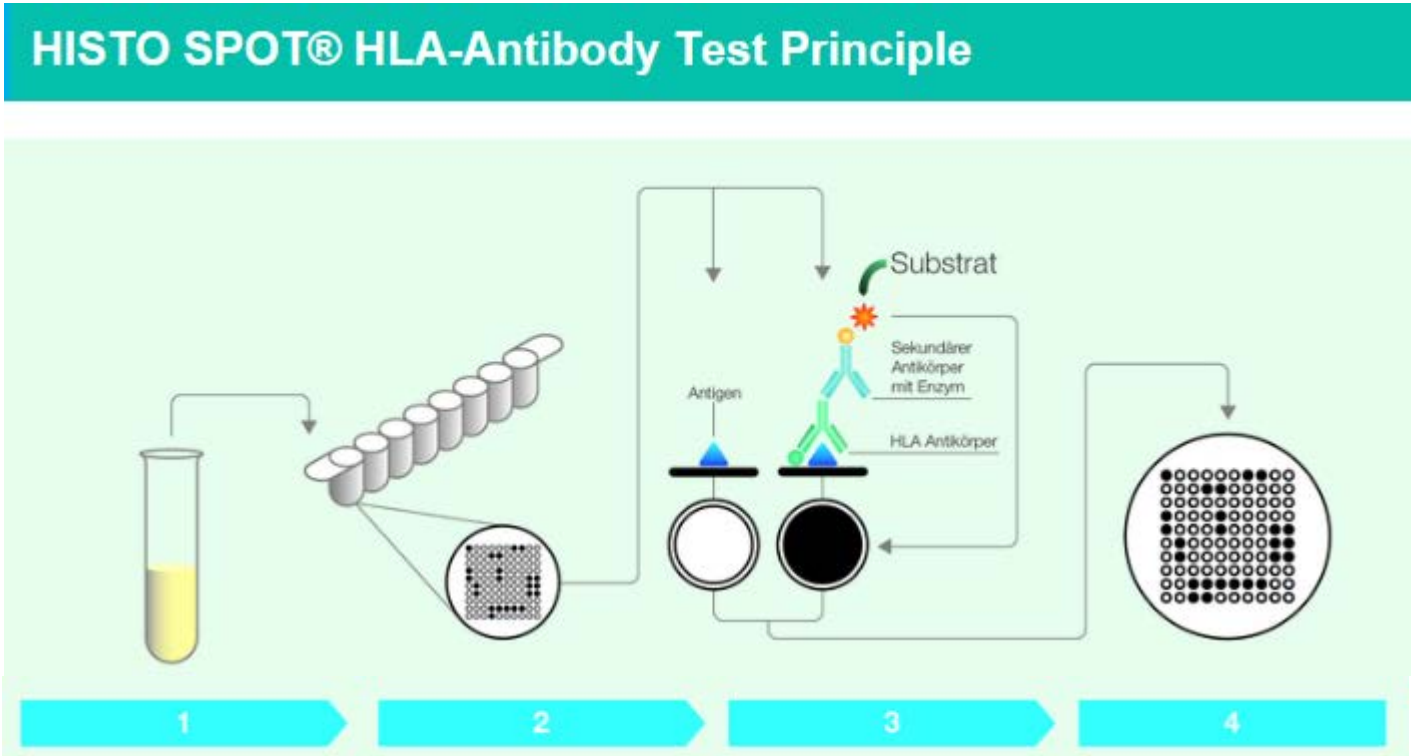
Mr. SPOT® Processor



Mr. SPOT® Processor

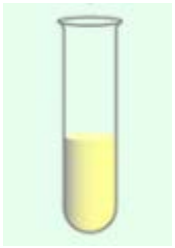


Principle of the Test



1	2	3	4
Sera dilution	Binding of the HLA-antibodies to the proteins on the bottom of the microtiterplate	Colorimetric detection of the positiv reactions	The presence and specificity of HLA-antibodies is determined in the reaction pattern

Workflow

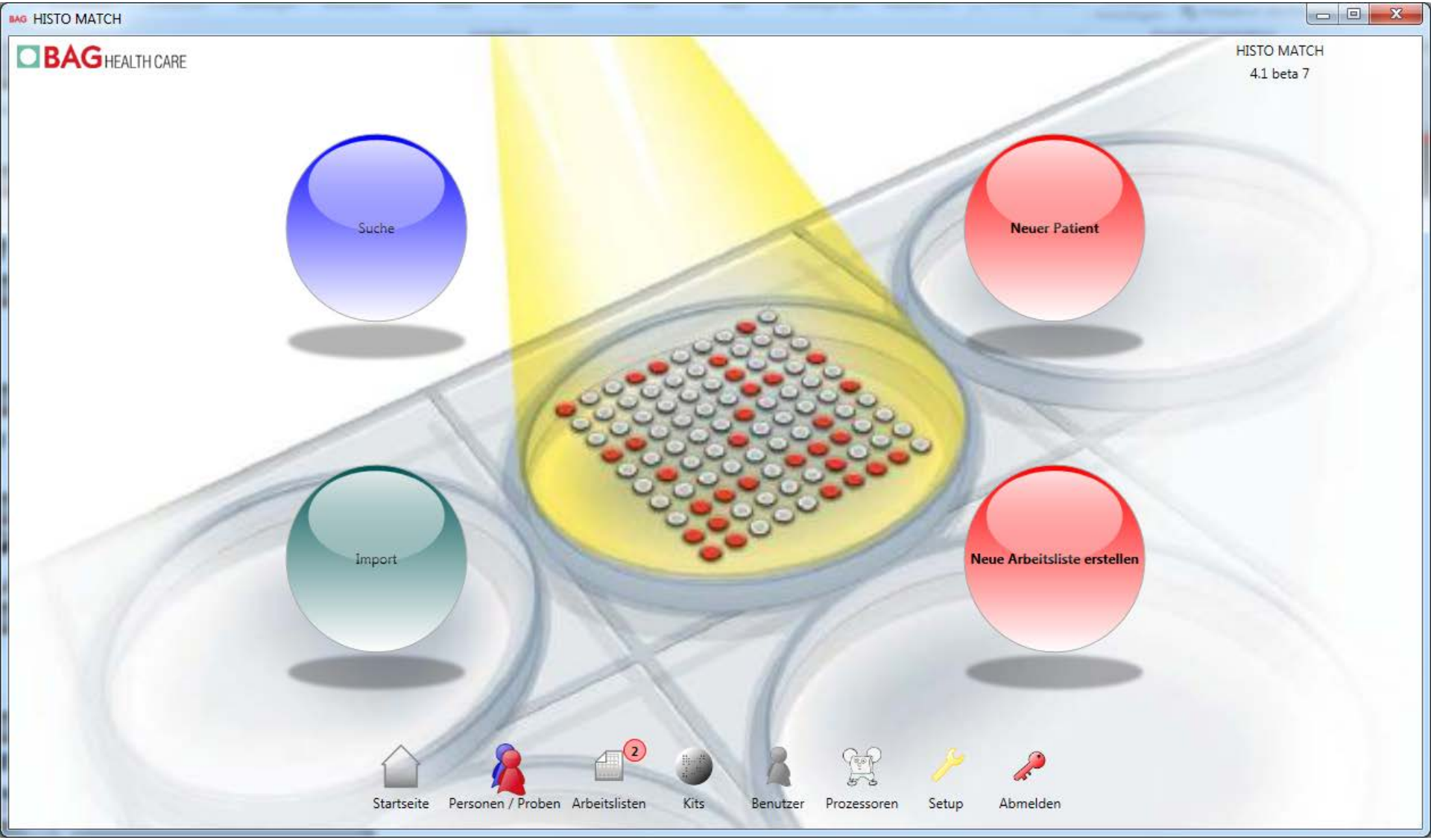


HLA AB ID Class I
5 µl Serum + 95 µl SampleBuffer

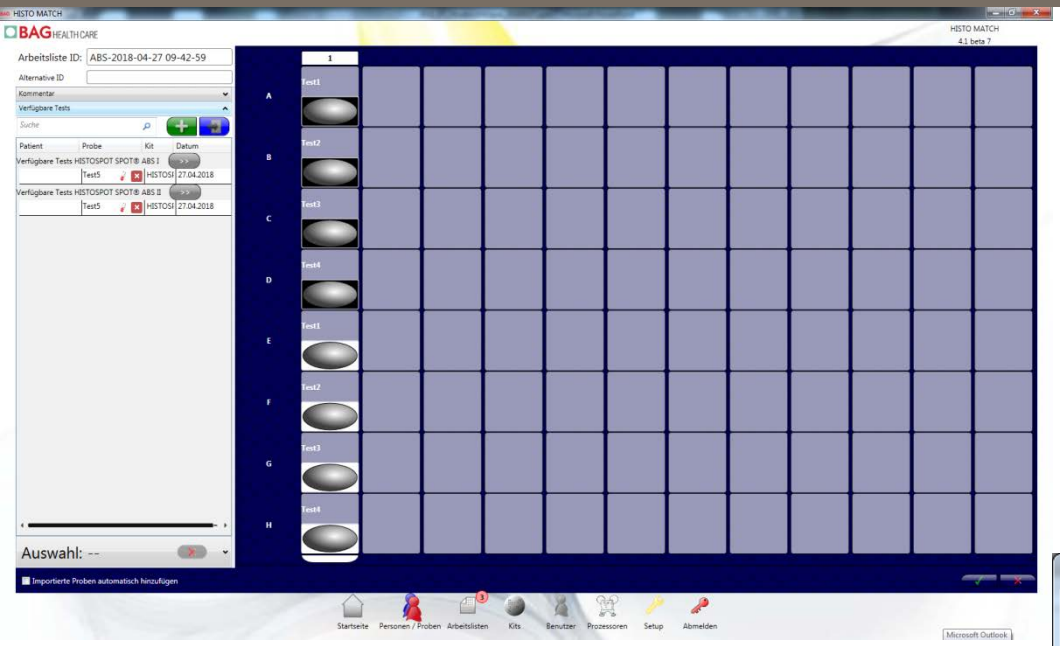
HLA AB ID Class II
2 µl Serum + 98 µl SampleBuffer

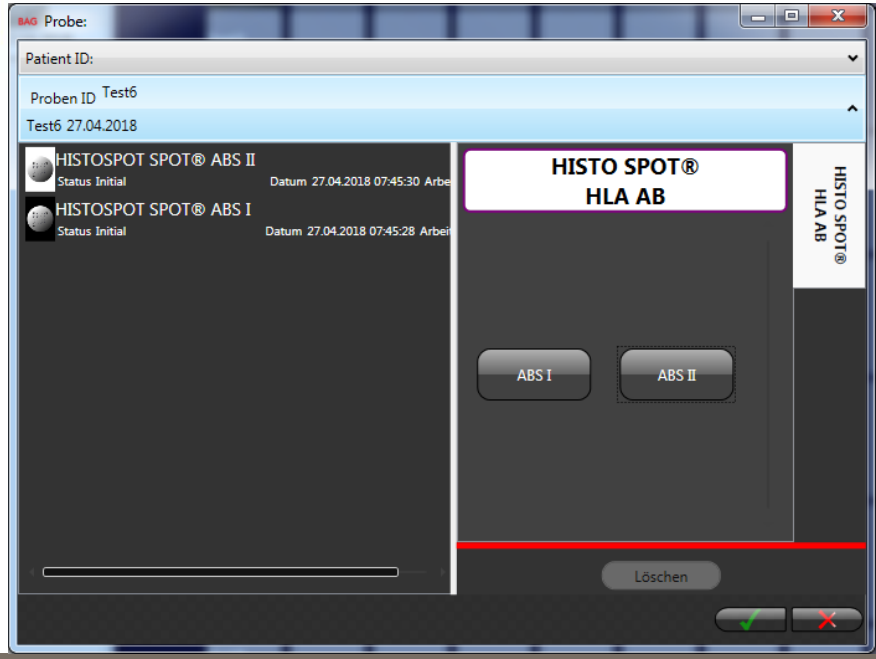
HistoMatch Software

HISTO MATCH Software

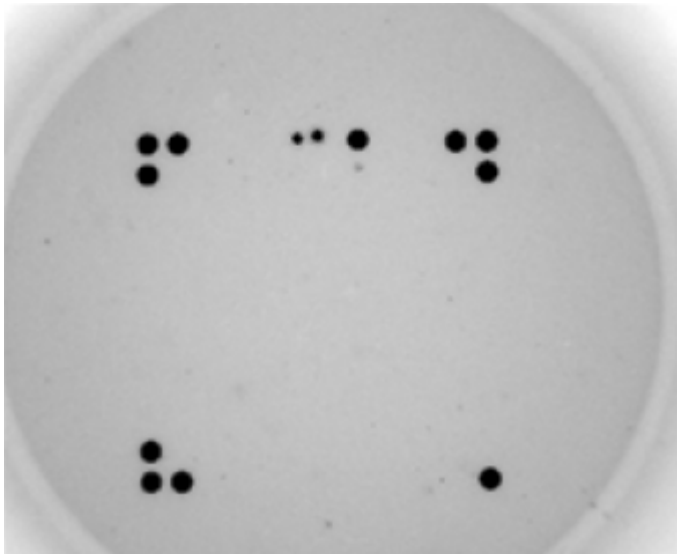


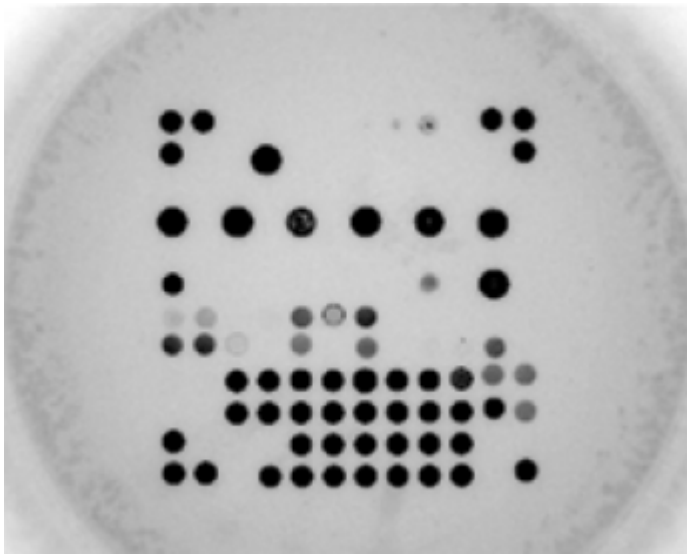
HISTO MATCH Software





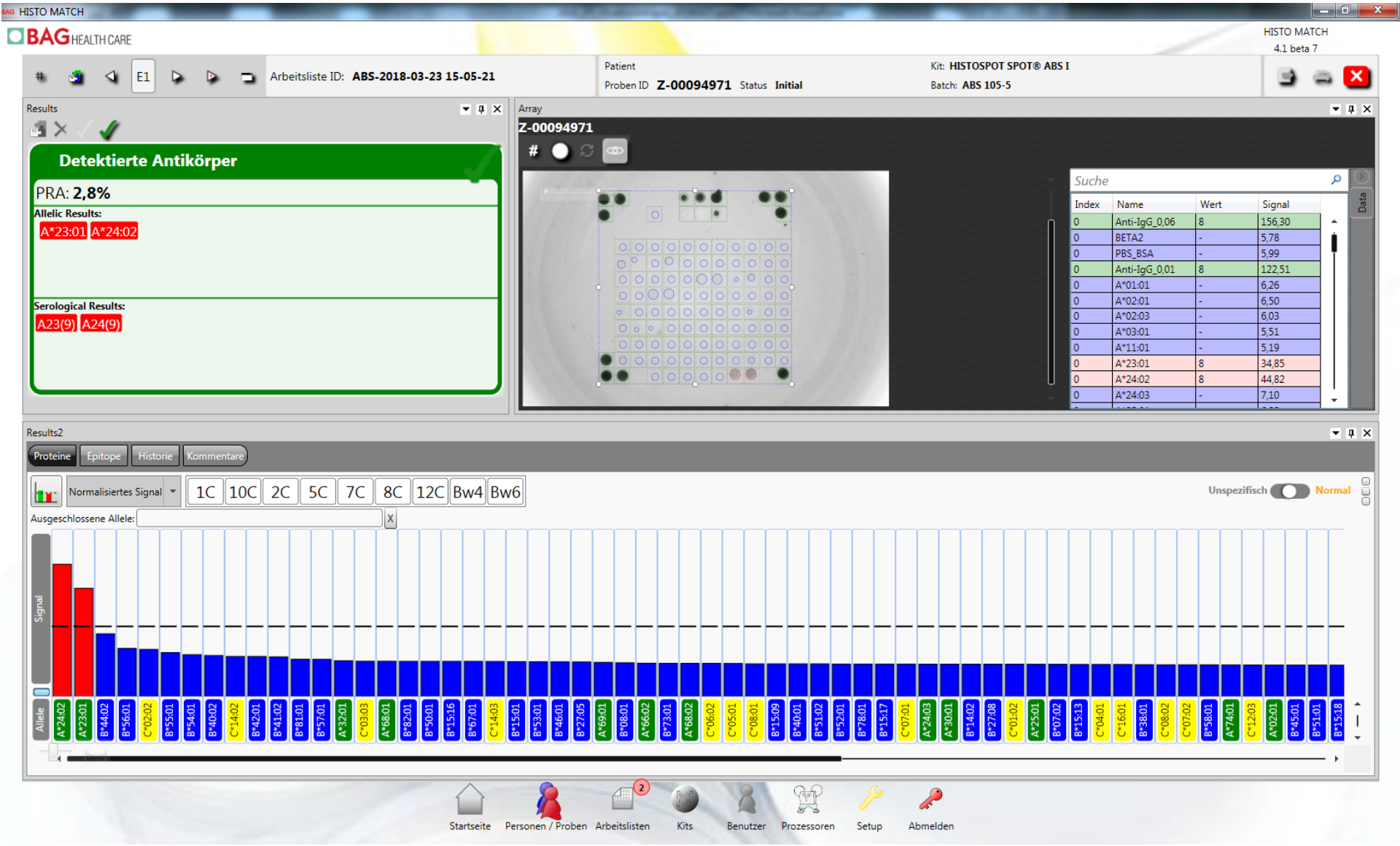
HISTO MATCH Software





- Positional Positions allow the software to locate the image
- IgG controls
- Positive and negativ sample reactions

HISTO MATCH Software



HISTO MATCH Report

HISTO MATCH Worklist BAG HEALTH CARE

Worklist: **ABS-2018-03-23 15-05-21**

Date: **3/23/2018**

Performed: **3/23/2018** By

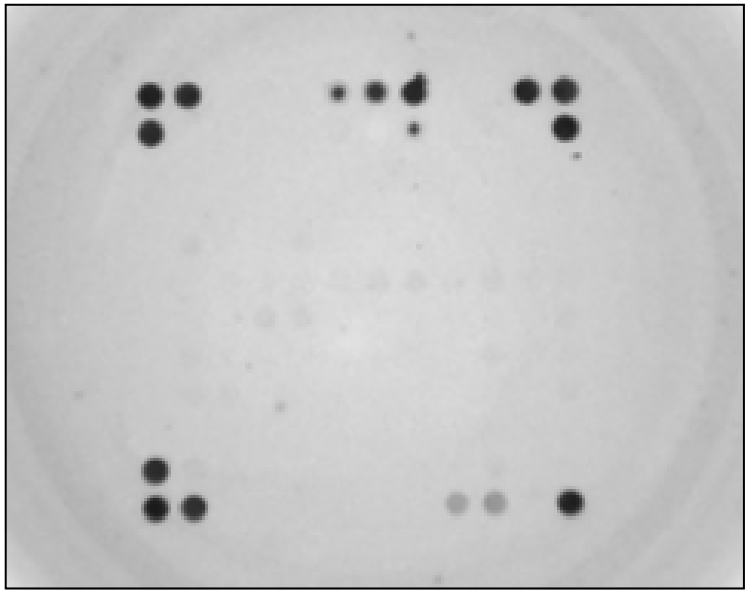
Imported: **3/25/2018** By **bag**

Machine: **56240** Protocol: **IBC Program V15 :**

PCRPlate: **-**

Reagents

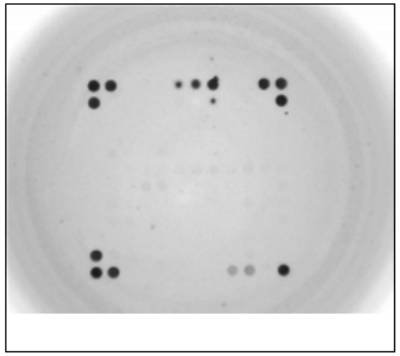
WASH1	1709WASH1/27-1
SAMBUF	1708SAMBUF/10-1
CONJBUF	1708CONJBUF/10-1
SUBSAB	1709SUBSAB/4-1
CONJAB	1705CONJAB/29-1



Sample: Z-00094971
Sample Date: 3/23/2018 3:05:47 PM

Date Performed
Analyzed By bag
Date Analyzed 4/17/2018 11:34:07 AM
Approved By bag
Date Approved 3/29/2018 7:25:36 AM
Authorized By
Date Authorized

Kit HISTOSPOT SPOT® ABS I
Batch: 601050516114
Expiry: 11/30/2018



Summary: A23;A24

PRA 2.79 Algorithm Normal

Specificity A23;A24

Excluded

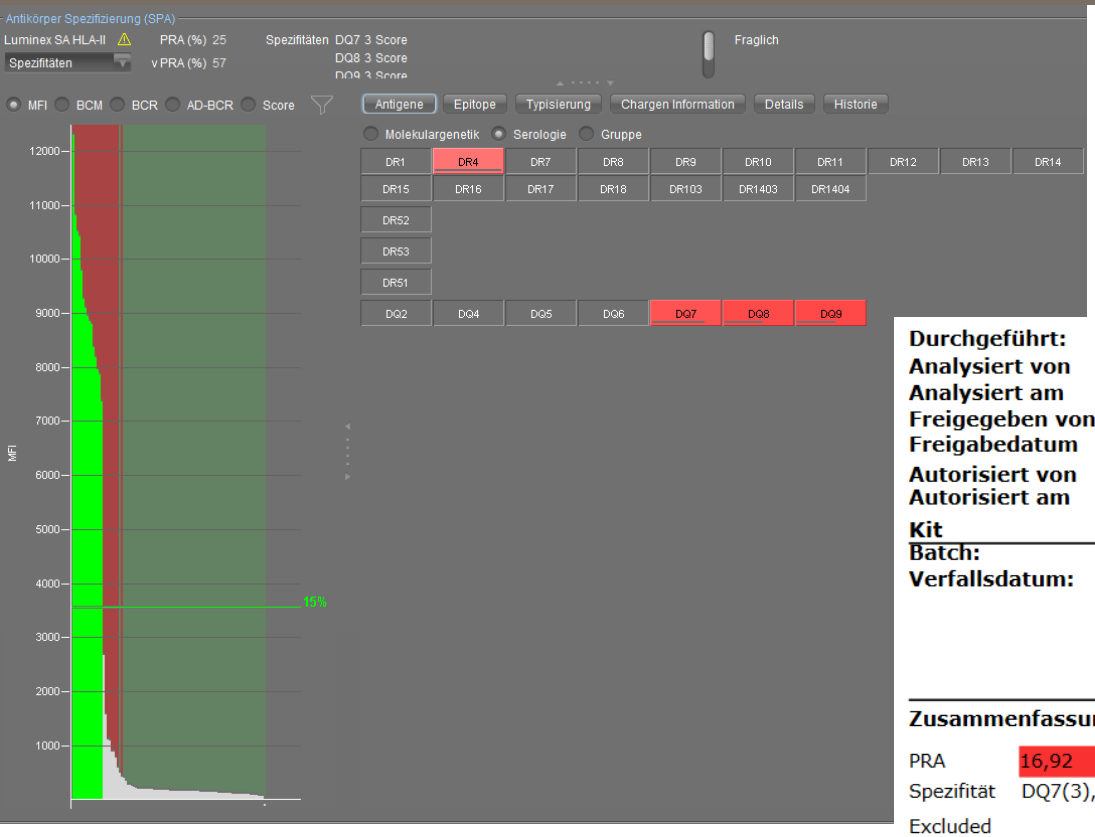
Name	Creg	Signal	Normalized Signal	CutOff	Value	Signal
A*24:02	1C,2C,Bw4	44.82	1.85	1.40	+	
A*23:01	1C,2C,Bw4	34.85	1.68	1.40	+	
B*44:02	12C,Bw4	12.47	1.35	1.40	-	
B*56:01	7C,Bw6	9.41	1.24	1.40	-	
C*02:02		9.10	1.24	1.40	-	
B*55:01	7C,Bw6	8.07	1.22	1.40	-	
B*54:01	7C,Bw6	7.39	1.20	1.40	-	
B*40:02	7C,12C,Bw6	7.99	1.19	1.40	-	
C*14:02		7.38	1.19	1.40	-	
B*42:01	7C,Bw6	7.27	1.19	1.40	-	
B*41:02	7C,12C,Bw6	7.64	1.18	1.40	-	
B*81:01	7C,Bw6	6.62	1.17	1.40	-	
B*57:01	2C,5C,Bw4	7.01	1.17	1.40	-	
A*32:01	10C,Bw4	7.64	1.16	1.40	-	
C*03:03		6.17	1.15	1.40	-	
A*68:01	10C,2C	6.69	1.15	1.40	-	
B*82:01	7C,Bw6	5.68	1.15	1.40	-	
B*50:01	5C,12C,Bw6	7.07	1.15	1.40	-	
B*15:16	5C,Bw4	7.18	1.15	1.40	-	
B*67:01	7C,8C,Bw6	7.16	1.15	1.40	-	
C*14:03		6.13	1.15	1.40	-	
B*15:01	5C,Bw6	6.13	1.15	1.40	-	
B*53:01	5C,Bw4	5.47	1.15	1.40	-	

Results HISTO SPOT® HLA AB compared to Luminex SA

HLA	Nr. of samples	MFI	Dropout
Class I	192	91,5%	0
Class II	87	91,4%	0

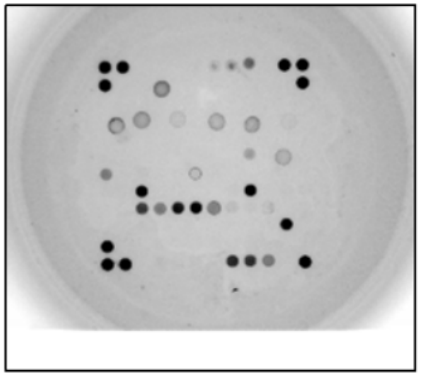
Agreement with matched antigens when compared to results obtained using Luminex Single Antigens LIFECODES LSA

Result



Durchgeführt:

Analysiert von	bag
Analysiert am	27.04.2018 10:53:03
Freigegeben von	bag
Freigabedatum	26.04.2018 10:52:13
Autorisiert von	
Autorisiert am	
Kit	HISTOSPOT SPOT® ABS II
Batch:	612052517014
Verfallsdatum:	31.01.2019



Zusammenfassung: DQ7(3),DQ8(3),DQ9(3),DQA2,DQA4,DR1,DR4,DR53

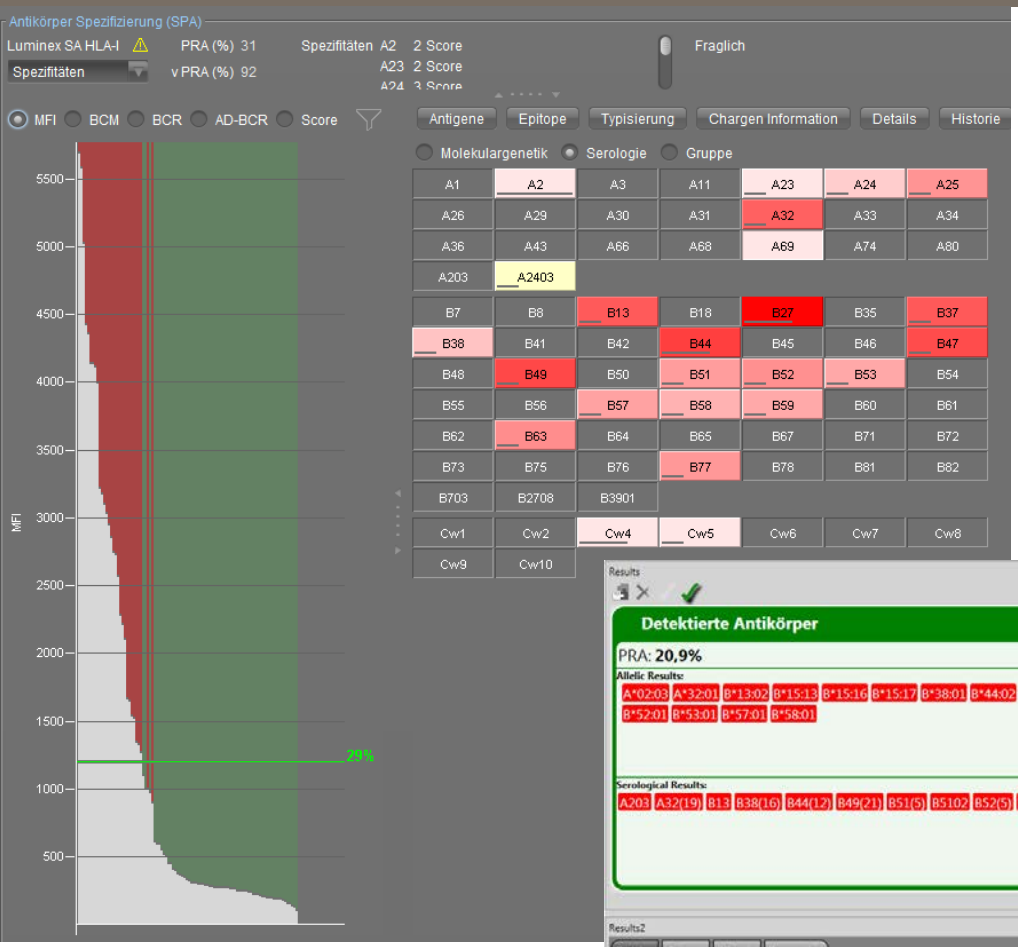
PRA 16,92 Algorithmus Normal

Spezifität DQ7(3),DQ8(3),DQ9(3),DQA2,DQA4,DR1,DR4,DR53

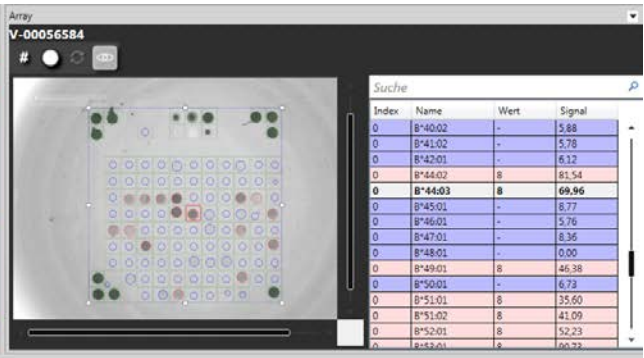
Excluded

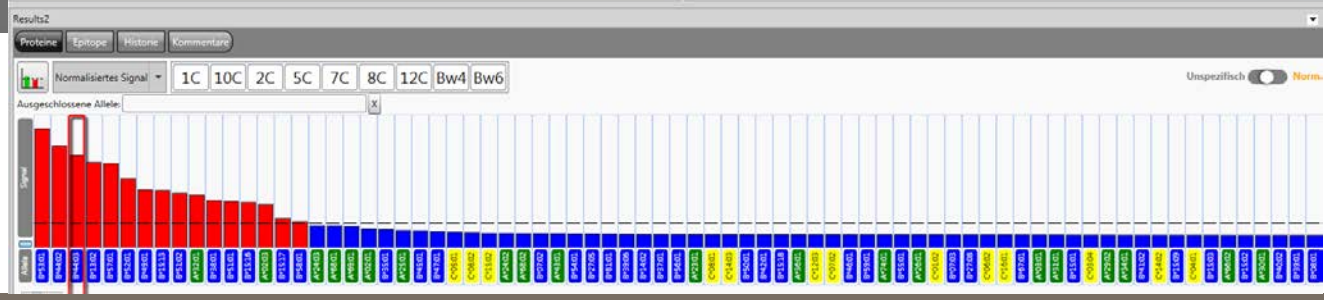
Name	Signal	Normalisiertes Signal	Schwellenwert	Wert Signal	
DQA1*02:01	165,84	3,77	1,43	+	
DQB1*03:02	165,84	3,77	1,43	+	
DRB1*04:01	146,76	3,45	1,43	+	
DQA1*02:01	169,69	3,35	1,43	+	
DQB1*03:03	169,69	3,35	1,43	+	
DQA1*02:01	128,19	3,06	1,43	+	
DQB1*03:01	128,19	3,06	1,43	+	
DRB1*04:02	126,31	2,73	1,43	+	
DRB4*01:01	61,65	2,03	1,85	+	
DRB1*01:01	61,65	2,03	1,85	+	
DRB1*04:01	61,65	2,03	1,85	+	
DRB1*04:05	59,08	1,83	1,43	+	
DRB1*08:01	47,84	1,79	1,85	-	

Result

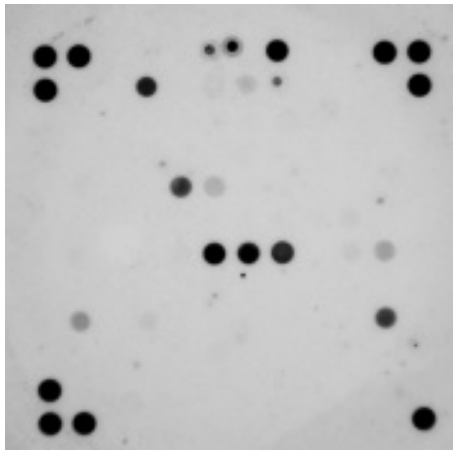

Antikörper Spezifizierung (SPA)
Luminex SA HLA-I
PRA (%) 31
Spezifitäten
v PRA (%) 92
Spezifitäten A2 2 Score
A23 2 Score
A24 3 Score
Fraglich
MFI BCM BCR AD-BCR Score
Antigene Epitope Typisierung Chargen Information Details Historie
Molekulargenetik Serologie Gruppe
A1 A2 A3 A11 A23 A24 A25
A26 A29 A30 A31 A32 A33 A34
A36 A43 A66 A68 A69 A74 A80
A203 A2403
B7 B8 B13 B18 B27 B35 B37
B38 B41 B42 B44 B45 B46 B47
B48 B49 B50 B51 B52 B53 B54
B55 B56 B57 B58 B59 B60 B61
B62 B63 B64 B65 B67 B71 B72
B73 B75 B76 B77 B78 B81 B82
B703 B2708 B3901
Cw1 Cw2 Cw4 Cw5 Cw6 Cw7 Cw8
Cw9 Cw10
MFI
5500
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
29%

Results
Detektierte Antikörper
PRA: 20,9%
Allelic Results:
A*02:03 A*32:01 B*13:02 B*15:13 B*15:16 B*15:17 B*38:01 B*44:02 B*44:03 B*49:01 B*51:01 B*51:02
B*52:01 B*53:01 B*57:01 B*58:01
Serological Results:
A203 A32(19) B13 B38(16) B44(12) B49(21) B51(5) B5102 B52(5) B53 B57(17) B58(17) B63(15) B77(15)

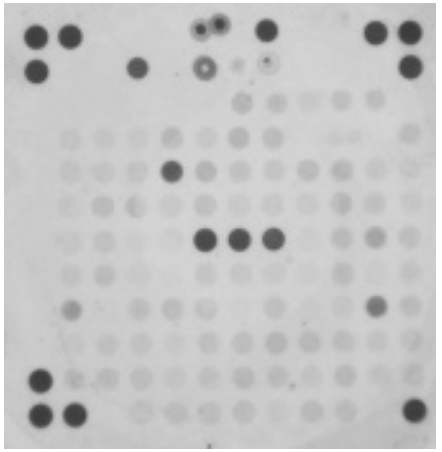

Array
V-00056584
Suche
Index Name Wert Signal
0 B*40:02 - 5,88
0 B*41:02 - 5,78
0 B*42:01 - 6,12
0 B*44:02 8 81,54
0 B*44:03 8 69,96
0 B*45:01 - 8,77
0 B*46:01 - 5,76
0 B*47:01 - 8,36
0 B*48:01 - 0,00
0 B*49:01 8 46,38
0 B*50:01 - 6,73
0 B*51:01 8 35,60
0 B*51:02 8 41,09
0 B*52:01 8 52,23


Results2
Proteine Epitope Historie Kommentare
Normalisiertes Signal
1C 10C 2C 5C 7C 8C 12C Bw4 Bw6
Unspezifisch
Norm.
Ausgeschlossene Allele:

Sera Treatment Heat Inactivation



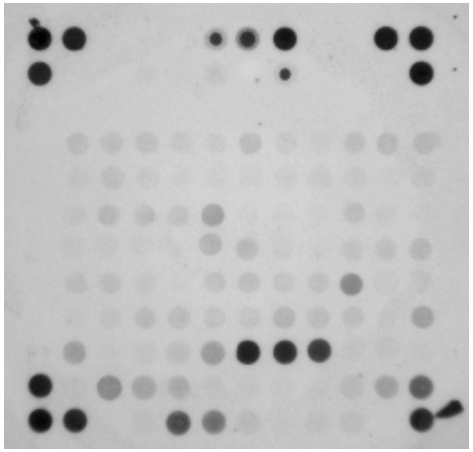
Untreated



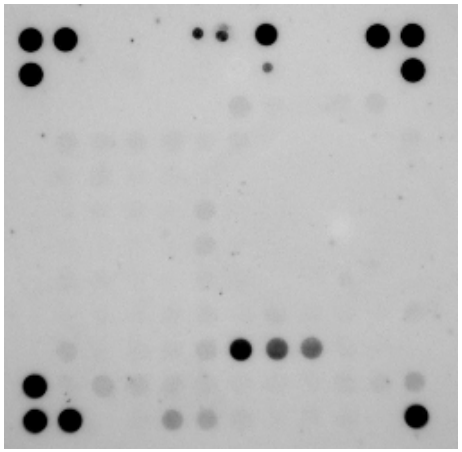
Heat inactivated

Heat inactivation of the sera increases unspecific background reactions with the HISTO SPOT HLA AB test and makes interpretation difficult or sometimes impossible.

Sera Treatment with SeraPure



Untreated



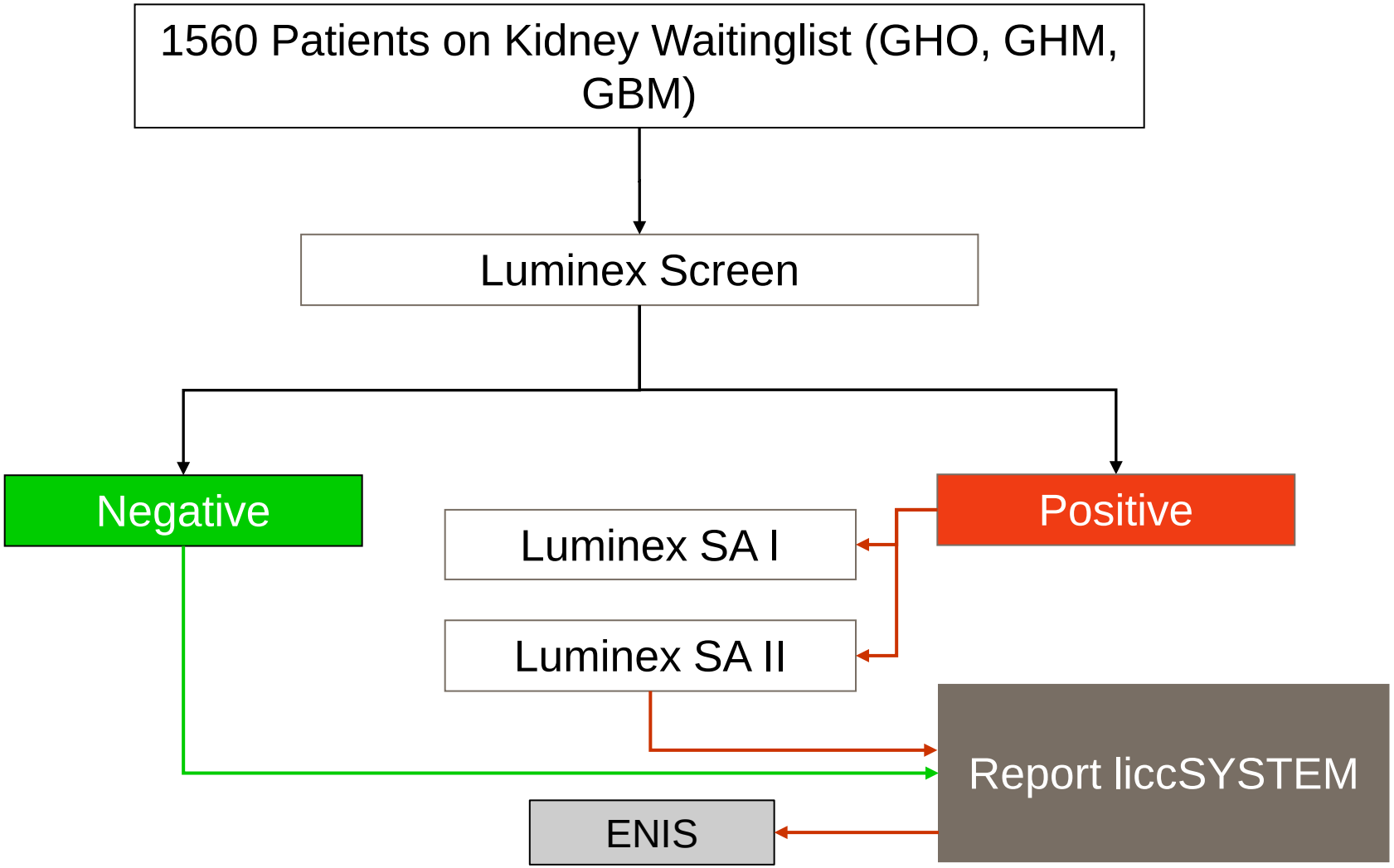
SeraPure Treatment

Sera treatment with SeraPure reduces unspecific background reactions with the HISTO SPOT HLA AB test and makes interpretation easier.

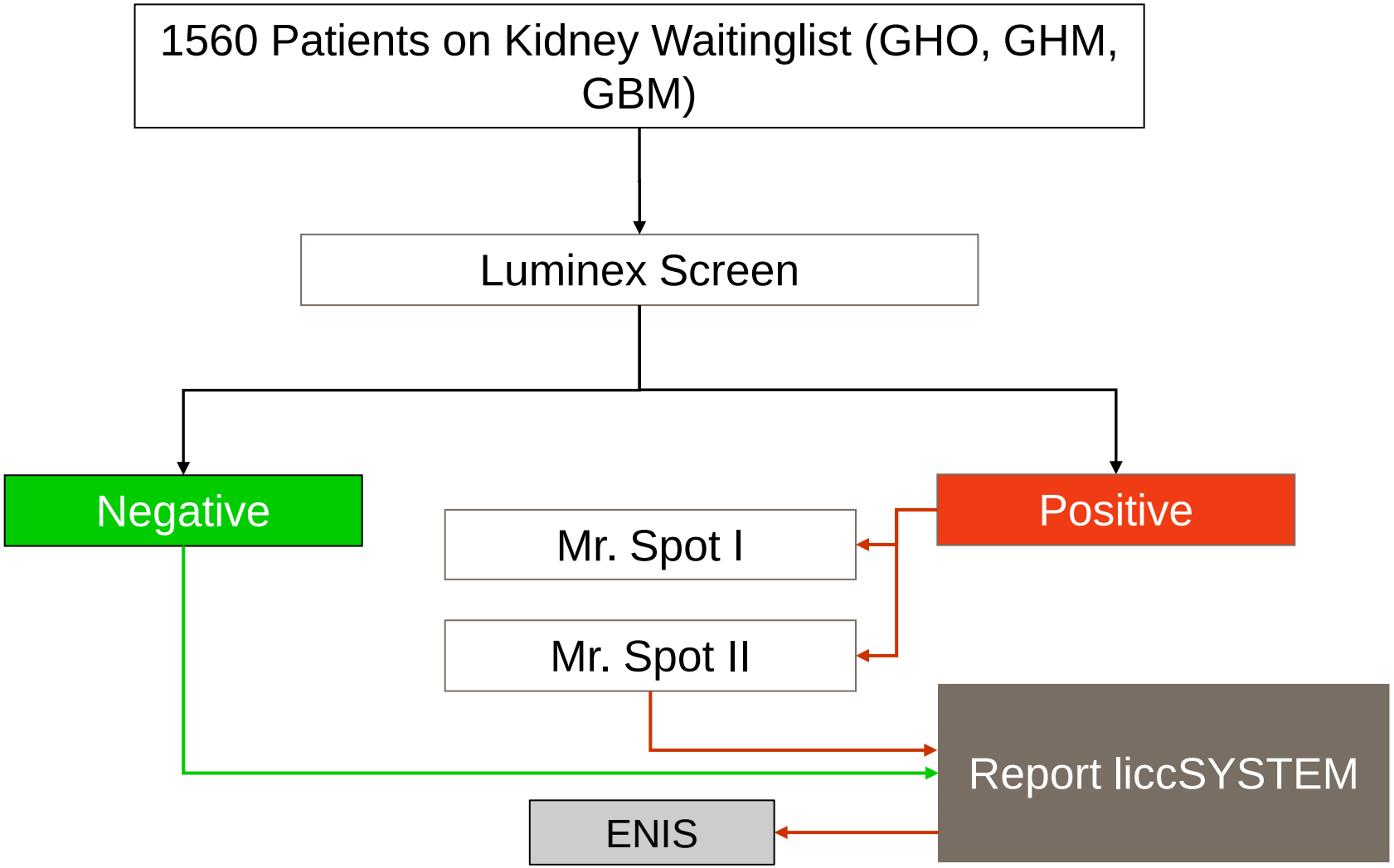
Experience with the ERY SPOT® SSO System

- Easy system to handle
- Easy automation and integration
- User friendly software
- High throughput
- Overall a good accordance of the results
- DQA1*03, *05, *06 are missing on the spot
- DPA1*02:01/DPB1*19:01 unspecific postiv

Quarterly Screening Kidney Recipients



Quarterly Screening Kidney Recipients



Thank you very much for your attention

Medizinische Hochschule
Hannover

Results HISTO SPOT® HLA AB compared to Luminex SA

HLA	Nr. of samples	MFI	Score 2	Score 3	Dropout
Class I	192	91,5%	91,9%	94,2%	0
Class II	87	91,4%	91,3%	92,8%	0