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ELISA Absorbance Test Plate, EQA 3002 DK, Report 2023

Participants

From January 2023 to January 2024 an ELISA Absorbance Test Plate (grey filter) was circulated between 25 participating laboratories from nine countries with a total of 35 ELISA readers. DEKS has given each instrument a "Code" as described in Table 1. The table also includes details about the manufacturer, reader model and year of purchase.

Description of the Absorbance Test Plate

This plate with serial number 125929 is an absorbance test plate with associated certificate. The plate has a grey filter mounted in certain wells. The grey filter is only mounted in well C1, D4, E2, F5, G3, and H6.
Most of the other wells are blinded. Some smaller empty open-ings are, however, found in the corners of A1, A12, H1 and H12. They are used to check whether the mechanical movement of the plate is correct. The absorbance in these wells should be 0.000 +/- 0.015. C6 has a normal sized well and is without glass. This is used to check the "blind" reading. The readings are per-formed towards air.

Acceptance interval

Agilent Technologies, Inc., performed the latest calibration on 6th December 2022.
The certified target values of the plate are traceable to NIST SRM 1930 SET 1-095, 3-095, 50-095 95 020/NDGF/4XGQ. The calibration was performed at 70 ºF (21.1 °C) and 25 % humidity. The acceptance interval of the measured wavelength is written in the schemes as ±2 % of the reading ±0.02 OD.

Procedure

All participants have been asked to test the Absorbance Test Plate at their instrument. There are 8 adjustable wavelengths. If the tests are performed in a different wavelength other than the adjustable wavelengths, the result will be included in the closest of the wavelengths.
Two tests are performed as the plate is turned 180° horizontally between the readings so A1 is read as H1, etc. This is done to check the ELISA reader's mechanical movement of the plate. The absorbance values from the two tests are filled into an excel-reporting table and forwarded to DEKS.
After receiving the results DEKS is sending the recommended values and plate certificate to the respective users.

Outliers

Outliers are defined by an expert opinion. For two of the participants, all absorbance values from the second test are set as manual outliers because there was a suspicion that the instructions for the plate were not followed. The values were not included in the calculations.

Results and comments

Mean absorbance value and deviation (%) from the certified target value, for the various manufacturers according to the adjustable wavelength, has been calculated (see table 2-9).

There was generally, a good agreement between the different producers. All producers found that the mean absorbance values were within the acceptance intervals of the target value, with a few exceptions: For DYNEX Technologies, the mean absorbance values were outside the acceptance interval at wavelength 450 nm in well C1, G3, H6, F5 and D4. The same was in case for Biochrom at wavelength 630 nm in well F5. From those manufacturers, the mean absorbance is calculated only from one participant and generally associated with a large degree of uncertainty. For Agilent BioTek, a measurement deviated at wavelength 490 nm well G3, resulting in the mean absorbance value being outside the acceptance interval. The same case was at wavelength 630 nm well D4, without the mean absorbance value being outside the acceptance interval.

Thus, there were no systematic deviations that could be linked to the different manufacturers and only deviations related to the individual participants were seen. It was assessed that the manufacturers were in good agreement with the certified target values.

To document that the Absorbance Test Plate had not changed characteristics during handling in the 25 laboratories, the plate was tested by DEKS before and after the circulation. The reading was carried out on a VICTOR3 Reader, from Perkin Elmer, at the wavelengths 405, 450 and 492 nm.

The tests show that the plate has not changed before and after circulation (differences ≤±0.01) and the measured absorbances are within the acceptance interval of the certified target values.

Yours sincerely

Dår Kur, DEKS
Lisbeth Nielsen, DEKS

Table 1   Absorbance Test Plate (ELISA grey filter), program 3002 DK

Instrument overview 2023
Gray filter serial number: 125929

Code	Manufacturer	Reader model	Year of purchase
1	Agilent BioTek	ELx808	2004
2	Molecular Devices	SpectraMax 190	2018
3	ASYS Hitech	Expert Plus UV	2012
4	TECAN	Infinite F50	2020
5	Thermo Scientific	Multiskan FC	2013
6	Molecular Devices	SpectraMax i3	2014
7	BMG LABTECH	CLARIOstar PLUS	2020
8	Agilent BioTek	ELx800	2018
9	Molecular Devices	SpectraMax 190	2006
10	Agilent BioTek	ELx808	-
11	Agilent BioTek	ELx808	-
12	Agilent BioTek	ELx808	-
13	Agilent BioTek	ELx808	-
14	Holm & Halby	Bio Tek Synergy HT	2013
15	TECAN	Sunrise	2014
16	TECAN	Sunrise	1999
17	BioSan	HiPo MPP-96	2017
18	Agilent BioTek	ELx808	2016
19	Perkin Elmer	VICTOR3	2007
20	Biochrom	Asys Expert 96	2016
21	Agilent BioTek	Powerwave XS	1997
22	Agilent BioTek	800 TS	2020
23	Euroimmun	Analyzer I	2020
24	Agilent BioTek	Epoch 2	2016
25	TECAN	Sunrise	2021
26	TECAN	Infinite F50	-
27	TECAN	Sunrise	2009
28	TECAN	Sunrise	2016
29	Thermo Scientific	Multiskan FC	2016
30	Thermo Scientific	Multiskan FC	2022
31	DYNEX Technologies	DYNAREAD	2019
32	BioSan	HiPo MPP-96	-
33	TECAN	Sunrise	2023
34	Perkin Elmer	VICTOR3	2008
35	TECAN	Spark	2021

Table 2   **Absorbance Test Plate (ELISA grey filter), program 3002 DK**

Mean absorbances and deviations of manufacturers 2023
Gray filter serialnumber: 125929

405 nm

Well	N	A1 / A12 / H1 / H12 / C6 (T)	C1 T	C1 M	C1 D	E2 T	E2 M	E2 D	G3 T	G3 M	G3 D	H6 T	H6 M	H6 D	F5 T	F5 M	F5 D	D4 T	D4 M	D4 D
Agilent BioTek	10	0,000 +/- 0,015	0,146 (0,123-0,169)	0,144	-1,2	0,556 (0,525-0,587)	0,552	-0,7	1,167 (1,124-1,210)	1,165	-0,2	1,744 (1,689-1,799)	1,747	0,2	2,095 (2,033-2,157)	2,105	0,5	2,812 (2,736-2,888)	2,798	-0,5
TECAN	9			0,150	2,5		0,556	-0,1		1,169	0,2		1,753	0,5		2,109	0,6		2,802	-0,4
Thermo Scientific	3			0,152	3,8		0,552	-0,8		1,162	-0,4		1,741	-0,2		2,089	-0,3		2,772	-1,4
Molecular Devices	3			0,147	0,5		0,552	-0,7		1,163	-0,3		1,745	0,1		2,097	0,1		2,787	-0,9
BioSan	2			0,153	4,8		0,554	-0,4		1,159	-0,7		1,742	-0,1		2,091	-0,2		2,778	-1,2
Perkin Elmer	2			0,146	0,0		0,552	-0,7		1,163	-0,3		1,745	0,1		2,095	0,0		2,786	-0,9
Holm & Halby	1			0,149	2,1		0,550	-1,1		1,159	-0,7		1,738	-0,3		2,092	-0,2		2,773	-1,4
BMG LABTECH	1			0,152	3,8		0,558	0,3		1,168	0,1		1,746	0,1		2,091	-0,2		2,802	-0,4
Biochrom	1			0,146	-0,3		0,550	-1,1		1,160	-0,6		1,741	-0,2		2,073	-1,1		2,761	-1,8
DYNEX Technologies	1			0,157	7,5		0,554	-0,4		1,164	-0,3		1,745	0,1		2,104	0,4		2,803	-0,3
ASYS Hitech	1			0,144	-1,4		0,550	-1,1		1,161	-0,6		1,740	-0,3		2,082	-0,6		2,784	-1,0
All	**34**			**0,149**	**2,0**		**0,553**	**-0,6**		**1,163**	**-0,4**		**1,744**	**0,0**		**2,093**	**-0,1**		**2,786**	**-0,9**

N = Number of answers

T = Target value and acceptance interval

M = Mean

D = Deviation from target value (%)

Table 3   **Absorbance Test Plate (ELISA grey filter), program 3002 DK**

Mean absorbances and deviations of manufacturers 2023
Gray filter serialnumber: 125929

450 nm

Well		A1	A12	H1	H12	C6		C1			E2			G3			H6			F5			D4	
	N			T			T	M	D	T	M	D	T	M	D	T	M	D	T	M	D	T	M	D
Agilent BioTek	10							0,138	-0,6		0,512	-0,9		1,079	-0,3		1,618	0,1		1,866	0,2		2,481	0,2
TECAN	8							0,142	2,0		0,514	-0,5		1,081	-0,2		1,621	0,3		1,869	0,4		2,481	0,2
Thermo Scientific	3							0,143	2,8		0,510	-1,4		1,072	-1,0		1,607	-0,6		1,855	-0,4		2,464	-0,5
Molecular Devices	3							0,139	0,2		0,513	-0,8		1,080	-0,3		1,620	0,2		1,865	0,2		2,482	0,2
BioSan	2							0,144	3,3		0,512	-1,0		1,070	-1,2		1,609	-0,4		1,859	-0,2		2,470	-0,2
Perkin Elmer	2			0,000 +/- 0,015			0,139 (0,116- 0,162)	0,138	-0,5	0,517 (0,487- 0,547)	0,508	-1,7	1,083 (1,041- 1,125)	1,069	-1,3	1,616 (1,564- 1,668)	1,603	-0,8	1,862 (1,805- 1,919)	1,850	-0,6	2,476 (2,406- 2,546)	2,461	-0,6
Holm & Halby	1							0,142	2,2		0,512	-1,0		1,077	-0,6		1,615	-0,1		1,867	0,2		2,475	-0,1
BMG LABTECH	1							0,138	-1,1		0,509	-1,6		1,071	-1,2		1,607	-0,6		1,854	-0,4		2,471	-0,2
Biochrom	1							0,136	-2,5		0,510	-1,4		1,074	-0,8		1,609	-0,4		1,857	-0,3		2,470	-0,2
DYNEX Technologies	1							0,164*	18,0		0,539	4,3		1,191*	10,0		1,732*	7,2		1,966*	5,6		2,607*	5,3
ASYS Hitech	1							0,137	-1,8		0,511	-1,2		1,077	-0,6		1,613	-0,2		1,844	-1,0		2,443	-1,4
Euroimmun	1							0,142	1,8		0,516	-0,2		1,085	0,1		1,624	0,5		1,878	0,9		2,497	0,8
All	**34**							**0,140**	**2,0**		**0,514**	**-0,6**		**1,076**	**0,2**		**1,613**	**0,4**		**1,860**	**0,4**		**2,472**	**0,3**

N = Number of answers

T = Target value and acceptance interval

M = Mean

D = Deviation from target value (%)

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Table 4   **Absorbance Test Plate (ELISA grey filter), program 3002 DK**

Mean absorbances and deviations of manufacturers 2023
Gray filter serialnumber: 125929

490 nm

Well	N	A1 A12 H1 H12 C6 — T	C1 T	C1 M	C1 D	E2 T	E2 M	E2 D	G3 T	G3 M	G3 D	H6 T	H6 M	H6 D	F5 T	F5 M	F5 D	D4 T	D4 M	D4 D
Agilent BioTek	10			0,133	-1,0		0,512	-1,8		0,978*	-9,9		1,615	-0,3		1,822	-0,1		2,420	-0,3
TECAN	8			0,136	1,4		0,514	-1,4		1,080	-0,4		1,617	-0,2		1,826	0,1		2,425	-0,1
Thermo Scientific	2			0,141	4,9		0,514	-1,4		1,081	-0,4		1,620	-0,1		1,828	0,3		2,424	-0,2
Molecular Devices	2			0,133	-0,9		0,511	-1,9		1,077	-0,7		1,616	-0,3		1,820	-0,2		2,424	-0,2
BioSan	2	0,000 +/- 0,015	0,134 (0,111-0,157)	0,141	5,2	0,521 (0,491-0,551)	0,520	-0,3	1,085 (1,043-1,127)	1,082	-0,3	1,621 (1,569-1,673)	1,627	0,3	1,823 (1,767-1,879)	1,829	0,3	2,428 (2,359-2,497)	2,435	0,3
Perkin Elmer	2			0,135	0,4		0,514	-1,3		1,084	-0,1		1,625	0,2		1,826	0,2		2,432	0,2
Holm & Halby	1			0,137	1,9		0,512	-1,7		1,078	-0,7		1,616	-0,3		1,846	1,2		2,448	0,8
BMG LABTECH	1			0,131	-2,6		0,507	-2,8		1,072	-1,2		1,609	-0,7		1,813	-0,5		2,420	-0,4
Biochrom	1			0,132	-1,9		0,513	-1,5		1,081	-0,4		1,621	0,0		1,824	0,1		2,432	0,1
ASYS Hitech	1			0,131	-2,6		0,514	-1,3		1,084	-0,1		1,624	0,2		1,826	0,1		2,427	0,0
All	**30**			**0,135**	**0,5**		**0,513**	**-1,6**		**1,080**	**-1,4**		**1,619**	**-0,1**		**1,826**	**0,2**		**2,428**	**0,0**

N = Number of answers
T = Target value and acceptance interval
M = Mean
D = Deviation from target value (%)
* = One or more results are outside the acceptance interval

Table 5 **Absorbance Test Plate (ELISA grey filter), program 3002 DK**

Mean absorbances and deviations of manufacturers 2023
Gray filter serialnumber: 125929

550 nm

Well		A1 / A12 / H1 / H12 / C6	C1			E2			G3			H6			F5			D4		
	N	T	T	M	D	T	M	D	T	M	D	T	M	D	T	M	D	T	M	D
TECAN	4			0,129	-1,2		0,511	-0,9		1,075	-0,1		1,611	0,2		1,810	0,2		2,409	0,3
Agilent BioTek	4			0,131	0,7		0,508	-1,5		1,070	-0,6		1,603	-0,2		1,809	0,2		2,400	-0,1
Thermo Scientific	2			0,131	0,5		0,508	-1,5		1,071	-0,5		1,605	-0,1		1,808	0,2		2,405	0,1
Molecular Devices	2	0,000 +/- 0,015	0,130 (0,107-0,153)	0,130	-0,4	0,516 (0,486-0,546)	0,507	-1,7	1,076 (1,034-1,118)	1,068	-0,7	1,607 (1,555-1,659)	1,602	-0,3	1,805 (1,749-1,861)	1,803	-0,1	2,402 (2,334-2,470)	2,401	-0,1
Holm & Halby	1			0,133	1,9		0,508	-1,6		1,068	-0,7		1,601	-0,4		1,808	0,2		2,397	-0,2
BMG LABTECH	1			0,128	-1,5		0,503	-2,5		1,063	-1,3		1,593	-0,9		1,794	-0,6		2,382	-0,8
ASYS Hitech	1			0,131	0,4		0,517	0,2		1,089	1,2		1,633	1,6		1,823	1,0		2,426	1,0
All	**15**		**0,130**	**0,1**		**0,509**	**-1,4**		**1,072**	**-0,4**		**1,607**	**0,0**		**1,808**	**0,1**		**2,403**	**0,0**

N = Number of answers

T = Target value and acceptance interval

M = Mean

D = Deviation from target value (%)

Table 6   **Absorbance Test Plate (ELISA grey filter), program 3002 DK**

Mean absorbances and deviations of manufacturers 2023
Gray filter serialnumber: 125929

620 nm

Well	N	A1 A12 H1 H12 C6 (T)	C1 T	C1 M	C1 D	E2 T	E2 M	E2 D	G3 T	G3 M	G3 D	H6 T	H6 M	H6 D	F5 T	F5 M	F5 D	D4 T	D4 M	D4 D
TECAN	8			0,139	2,8		0,515	-1,3		1,083	-0,5		1,624	0,0		1,756	0,2		2,335	0,2
Thermo Scientific	3			0,140	3,6		0,513	-1,7		1,080	-0,7		1,618	-0,3		1,751	-0,1		2,327	-0,2
Molecular Devices	3			0,136	0,6		0,513	-1,8		1,080	-0,8		1,620	-0,3		1,748	-0,3		2,329	-0,1
Agilent BioTek	3			0,136	0,7		0,512	-2,0		1,079	-0,9		1,618	-0,4		1,752	-0,1		2,325	-0,3
BioSan	2		0,135	0,145	7,1	0,522	0,517	-1,0	1,088	1,080	-0,8	1,624	1,622	-0,1	1,753	1,752	-0,1	2,331	2,330	-0,1
Holm & Halby	1	0,000 +/- 0,015	(0,112-	0,139	3,0	(0,492-	0,513	-1,7	(1,046-	1,080	-0,7	(1,572-	1,618	-0,4	(1,698-	1,753	0,0	(2,264-	2,323	-0,4
Perkin Elmer	1		0,158)	0,139	2,6	0,552)	0,513	-1,7	1,130)	1,080	-0,8	1,676)	1,620	-0,3	1,808)	1,751	-0,1	2,398)	2,334	0,1
BMG LABTECH	1			0,134	-1,1		0,509	-2,6		1,073	-1,4		1,612	-0,8		1,742	-0,7		2,332	0,0
Biochrom	1			0,135	-0,4		0,513	-1,7		1,081	-0,7		1,619	-0,3		1,750	-0,2		2,329	-0,1
ASYS Hitech	1			0,135	-0,4		0,512	-1,9		1,079	-0,8		1,617	-0,4		1,744	-0,5		2,324	-0,3
All	**24**			**0,138**	**1,9**		**0,513**	**-1,7**		**1,079**	**-0,8**		**1,619**	**-0,3**		**1,750**	**-0,2**		**2,329**	**-0,1**

N = Number of answers

T = Target value and acceptance interval

M = Mean

D = Deviation from target value (%)

Table 7 **Absorbance Test Plate (ELISA grey filter), program 3002 DK**

Mean absorbances and deviations of manufacturers 2023
Gray filter serialnumber: 125929

630 nm

Well	A1	A12	H1	H12	C6	C1 T	C1 M	C1 D	E2 T	E2 M	E2 D	G3 T	G3 M	G3 D	H6 T	H6 M	H6 D	F5 T	F5 M	F5 D	D4 T	D4 M	D4 D
	N		T			T	M	D	T	M	D	T	M	D	T	M	D	T	M	D	T	M	D
Agilent BioTek	10						0,135	-0,4		0,507	-1,9		1,069	-0,8		1,601	-0,3		1,718	-0,3		2,280*	-0,9
TECAN	2						0,133	-2,0		0,508	-1,8		1,070	-0,7		1,605	-0,1		1,725	0,1		2,294	-0,3
Molecular Devices	2						0,135	-0,7		0,508	-1,8		1,069	-0,7		1,603	-0,3		1,720	-0,2		2,291	-0,5
Holm & Halby	1		0,000 +/- 0,015			0,136	0,140	2,6	0,517	0,508	-1,7	1,077	1,075	-0,2	1,607	1,611	0,2	1,724	1,740	0,9	2,301	2,305	0,2
BioSan	1					(0,113-	0,143	4,9	(0,487-	0,508	-1,8	(1,035-	1,061	-1,5	(1,555-	1,588	-1,2	(1,670-	1,683	-2,4	(2,235-	2,237	-2,8
BMG LABTECH	1					0,159)	0,134	-1,8	0,547)	0,504	-2,5	1,119)	1,064	-1,2	1,659)	1,597	-0,7	1,778)	1,718	-0,3	2,367)	2,305	0,2
Biochrom	1						0,138	1,1		0,503	-2,7		1,058	-1,8		1,585	-1,4		1,669*	-3,2		2,224	-3,4
DYNEX Technologies	1						0,148	8,5		0,513	-0,9		1,076	-0,1		1,610	0,2		1,733	0,5		2,306	0,2
All	**19**						**0,138**	**1,5**		**0,507**	**-1,9**		**1,068**	**-0,9**		**1,600**	**-0,4**		**1,720**	**-0,6**		**2,280**	**-0,9**

N = Number of answers

T = Target value and acceptance interval

M = Mean

D = Deviation from target value (%)

* = One or more results are outside the acceptance interval

Table 8 **Absorbance Test Plate (ELISA grey filter), program 3002 DK**

Mean absorbances and deviations of manufacturers 2023
Gray filter serialnumber: 125929

690 nm

Well	N	A1 / A12 / H1 / H12 / C6 T	C1 T	C1 M	C1 D	E2 T	E2 M	E2 D	G3 T	G3 M	G3 D	H6 T	H6 M	H6 D	F5 T	F5 M	F5 D	D4 T	D4 M	D4 D
TECAN	2			0,124	-1,6		0,433	-0,9		0,909	-0,1		1,370	1,2		1,434	1,0		1,897	0,5
Molecular Devices	2	0,000 +/- 0,015	0,126 (0,103-0,149)	0,126	0,0	0,437 (0,408-0,466)	0,433	-1,0	0,910 (0,872-0,948)	0,906	-0,4	1,354 (1,307-1,401)	1,355	0,1	1,420 (1,372-1,468)	1,420	0,0	1,887 (1,829-1,945)	1,890	0,1
Agilent BioTek	2			0,131	4,0		0,434	-0,8		0,906	-0,5		1,355	0,1		1,427	0,5		1,889	0,1
Holm & Halby	1			0,130	2,8		0,434	-0,7		0,908	-0,2		1,359	0,3		1,430	0,7		1,896	0,5
BMG LABTECH	1			0,124	-2,0		0,427	-2,4		0,896	-1,6		1,341	-1,0		1,411	-0,6		1,878	-0,5
Alle	**8**			**0,127**	**0,6**		**0,432**	**-1,2**		**0,905**	**-0,6**		**1,356**	**0,1**		**1,424**	**0,3**		**1,890**	**0,1**

N = Number of answers

T = Target value and acceptance interval

M = Mean

D = Deviation from target value (%)

Table 9 — Absorbance Test Plate (ELISA grey filter), program 3002 DK

Mean absorbances and deviations of manufacturers 2023
Gray filter serialnumber: 125929

750 nm

Well	N	A1 A12 H1 H12 C6 — T	C1 T	C1 M	C1 D	E2 T	E2 M	E2 D	G3 T	G3 M	G3 D	H6 T	H6 M	H6 D	F5 T	F5 M	F5 D	D4 T	D4 M	D4 D
Molecular Devices	2			0,132	0,0		0,385	-1,9		0,801	-0,9		1,196	-0,3		1,178	-0,1		1,566	-0,1
Agilent BioTek	2	0,000 +/- 0,015	0,132	0,137	3,8	0,392	0,387	-1,4	0,808	0,803	-0,7	1,200	1,198	-0,2	1,180	1,187	0,6	1,567	1,567	0,0
Holm & Halby	1		(0,109-	0,135	1,9	(0,364-	0,385	-1,8	(0,772-	0,801	-0,9	(1,156-	1,196	-0,3	(1,136-	1,185	0,4	(1,516-	1,568	0,1
BMG LABTECH	1		0,155)	0,130	-1,5	0,420)	0,382	-2,6	0,844)	0,798	-1,3	1,244)	1,191	-0,8	1,224)	1,174	-0,6	1,618)	1,560	-0,4
Alle	**6**			**0,133**	**1,0**		**0,385**	**-1,9**		**0,800**	**-0,9**		**1,195**	**-0,4**		**1,181**	**0,1**		**1,565**	**-0,1**

N = Number of answers

T = Target value and acceptance interval

M = Mean

D = Deviation from target value (%)