

	LUDOX CL-X	H2O
Replicate 1	0,072	0,042
Replicate 2	0,062	0,041
Replicate 3	0,072	0,039
Replicate 4	0,058	0,041
Arith. Mean	0,066	0,041
Corrected Abs600	0,025	
Reference OD600	0,063	
OD600/Abs600	2,495	

Enter Abs600 absorbance measurements into blue cells
Gold cells are calculated

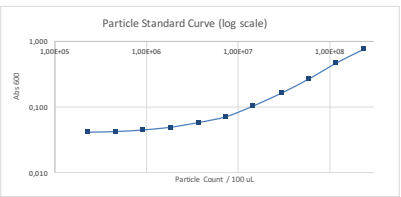
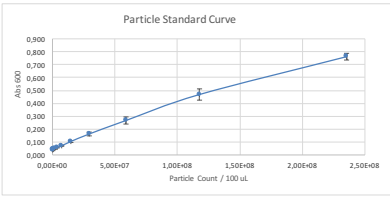
Corrected value is particle-only contribution

Reference value is for 100uL of LUDOX CL-X in a well of a standard 96-well flat-bottom black with clear bottom plate

Corrected value = scaling factor * measured value

Number of Particles	2,35E+08	1,18E+08	5,88E+07	2,94E+07	1,47E+07	7,35E+06	3,68E+06	1,84E+06	9,19E+05	4,60E+05	2,30E+05	0
Replicate 1	0,782	0,416	0,308	0,181	0,113	0,070	0,081	0,050	0,046	0,043	0,043	0,040
Replicate 2	0,784	0,523	0,258	0,166	0,108	0,077	0,062	0,053	0,045	0,042	0,042	0,038
Replicate 3	0,751	0,487	0,246	0,156	0,101	0,070	0,055	0,048	0,044	0,042	0,040	0,042
Replicate 4	0,730	0,468	0,257	0,147	0,095	0,068	0,054	0,046	0,044	0,042	0,040	0,039
Arith. Mean	0,762	0,469	0,267	0,163	0,104	0,071	0,058	0,049	0,045	0,042	0,041	0,040
Arith. Std. Dev.	0,026	0,044	0,028	0,015	0,008	0,004	0,004	0,003	0,001	0,000	0,002	0,002
Arith. Net Mean	0,722	0,429	0,228	0,123	0,085	0,032	0,018	0,010	0,005	0,003	0,002	

Enter Abs600 measurements into blue cells
Gold cells are calculated



Values should form a straight line on both linear and log scale
Slope should be 1:1
Common problems:
* Consistent pipetting error -> log graph is a straight line but not 1:1 slope
* Oversaturated detector -> low concentrations linear, but high concentrations saturate or fall

Particles / OD												
Number of Particles	2,35E+08	1,18E+08	5,88E+07	2,94E+07	1,47E+07	7,35E+06	3,68E+06	1,84E+06	9,19E+05	4,60E+05	2,30E+05	
Mean particles / Abs60	3,26E+08	2,74E+08	2,59E+08	2,40E+08	2,28E+08	2,33E+08	2,01E+08	1,93E+08	1,84E+08	1,84E+08	1,53E+08	
Mean of med-high levels:		2,67E+08										

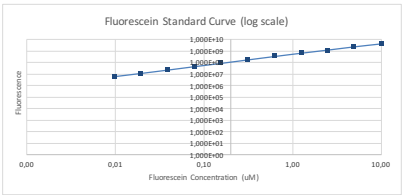
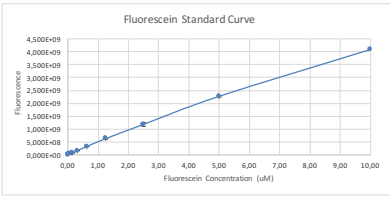
Final scaling level determined from medium-high points likely to be less impacted by saturation or pipetting error
If needed, you can shift which points are used, but it is likely better to correct instrument settings and protocol.

Cospheric Monodisperse Silica Microspheres 0.961um diameter	
Spheres/gram	1,20E+12
grams/mL	1,8
Spheres/0.55 mL	1,19E+12
Resuspend volume mL:	2,55
Dilution X:	100
Total volume mL:	255
Particles / mL:	4,71E+09
Well volume (mL)	0,1
Initial particles:	4,71E+08

Fluorescein uM	10,00	5	2,5	1,25	0,625	0,313	0,156	0,078	0,039	0,0195	0,0098	0
Replicate 1	4101774592	2316800512	1253407616	643101184	336733248	168875232	83255536	43427020	21543596	11019223	5679264	517818
Replicate 2	4117344256	2254552832	1080104576	651060736	342562976	179829136	91687536	48172432	25178436	13205588	7105064	575521
Replicate 3	4085643520	2257568256	1207511168	627929472	316352480	158051824	78904608	40389504	19943828	10159737	5104447	510238
Replicate 4	4053898240	2266416896	1199320448	606238656	314220832	158020480	78050824	40234440	20250010	10229300	5524524	705505
Arith. Mean	4,090E+09	2,274E+09	1,185E+09	6,321E+08	3,275E+08	1,662E+08	8,297E+07	4,306E+07	2,173E+07	1,115E+07	5,853E+06	5,773E+05
Arith. Std. Dev.	2,713E+07	2,908E+07	7,392E+07	1,972E+07	1,429E+07	1,043E+07	6,240E+06	3,714E+06	2,402E+06	1,423E+06	8,691E+05	9,632E+04
Arith. Net Mean	4,089E+09	2,273E+09	1,185E+09	6,315E+08	3,269E+08	1,656E+08	8,240E+07	4,248E+07	2,115E+07	1,098E+07	5,276E+06	

Enter fluorescence measurements into blue cells
Gold cells are calculated

Values measured are fluorescence from 100ul of X uM fluorescein solution



Values should form a straight line on both linear and log scale

Slope should be 1:1

Common problems:

* Consistent pipetting error -> log graph is a straight line but not 1:1 slope

* Oversaturated detector -> low concentrations linear, but high concentrations saturate or fall

Fluorescein/a.u.	10,00	5,00	2,50	1,25	0,63	0,31	0,16	0,08	0,04	0,02	0,01
uM Fluorescein/a.u.	2,45E-09	2,20E-09	2,11E-09	1,98E-09	1,91E-09	1,89E-09	1,90E-09	1,94E-09	1,85E-09	1,85E-09	1,85E-09
Mean uM Fluorescein / a.u.:		2,02E-09									
MEFL / a.u.:		1,22E+04									

Final scaling level determined from medium-high points likely to be less impacted by saturation or pipetting error
if needed, you can shift which points are used, but it is likely better to correct instrument settings and protocol.

Fluorescein uM -> MEFL calculation:

Initial Molarity	1,00E-05
Molecules / Mole	6,02E+23
Well volume (L):	1,00E-04
Initial Molecules:	6,02E+14
MEFL / uM	6,02E+12

Raw Plate Readings

If you followed the recommended plate layout:
Copy fluorescence and Abs600 measurements from your plate reader
They will automatically propagate into the correct locations in the spreadsheet

Fluorescence Raw Readings:

Hour 0:	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4
Colony 1, Replicate 1	3737780	8598575	57112852	13725994	4071682	46342088
Colony 1, Replicate 2	3984544	8774553	59249556	14935108	4273014	45478184
Colony 1, Replicate 3	4067602	8765117	59673932	14549522	4212216	51024520
Colony 1, Replicate 4	3982304	8741440	53220388	14190767	4283382	46393720
Colony 2, Replicate 1	3688771	8518456	54990812	13836162	3999170	44263916
Colony 2, Replicate 2	3823365	8592262	57224908	14567838	4252518	43430904
Colony 2, Replicate 3	3819795	8740283	61415148	14965353	4090535	43858552
Colony 2, Replicate 4	3915248	8947030	60375324	15114168	4165222	43775728

Hour 6:	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4
Colony 1, Replicate 1	4041034	51018068	130267544	130274232	5537580	203024864
Colony 1, Replicate 2	4557892	56405652	131888104	141961168	6074006	211913952
Colony 1, Replicate 3	4658624	55013656	128607064	134899552	5936775	205731552
Colony 1, Replicate 4	4579504	55967444	132479632	141232736	6375383	205116896
Colony 2, Replicate 1	4175749	51407056	119862612	144530512	5327578	132026200
Colony 2, Replicate 2	4519702	57412388	130411696	158798032	6214479	160329936
Colony 2, Replicate 3	4516921	56182004	124317932	150581840	5972472	154131776
Colony 2, Replicate 4	4430078	58913748	125426380	160833760	6133252	143455920

Assumed plate well pattern:

A1	A2	A3	A4	A5	A6
B1	B2	B3	B4	B5	B6
C1	C2	C3	C4	C5	C6
D1	D2	D3	D4	D5	D6
E1	E2	E3	E4	E5	E6
F1	F2	F3	F4	F5	F6
G1	G2	G3	G4	G5	G6
H1	H2	H3	H4	H5	H6

ments from your plate reader into blue cells
he correct locations in the Fluorescence Measurement Sheet

Abs600 Raw Readings:

Device 5	Device 6	LB + Chlor (blank)	Hour 0:	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4	Device 5	Device 6	LB + Chlor (blank)
16896500	6413294	2917991	Colony 1, Replicate 1	0,058	0,056	0,054	0,06	0,059	0,056	0,055	0,057	0,042
17706426	6728308	3131822	Colony 1, Replicate 2	0,057	0,057	0,053	0,056	0,059	0,055	0,056	0,057	0,041
18066842	6497363	3057722	Colony 1, Replicate 3	0,058	0,057	0,054	0,056	0,061	0,055	0,057	0,058	0,042
17679176	6415930	3218852	Colony 1, Replicate 4	0,059	0,058	0,054	0,057	0,059	0,055	0,057	0,059	0,042
22841736	9309888	3158208	Colony 2, Replicate 1	0,059	0,058	0,05	0,057	0,059	0,056	0,067	0,058	0,042
22625746	10623452	3323871	Colony 2, Replicate 2	0,061	0,058	0,054	0,057	0,06	0,057	0,057	0,057	0,042
23341112	10847352	3169005	Colony 2, Replicate 3	0,059	0,062	0,053	0,058	0,061	0,059	0,058	0,058	0,042
24292710	10809843	3137583	Colony 2, Replicate 4	0,059	0,057	0,052	0,057	0,061	0,057	0,058	0,056	0,041

Device 5	Device 6	LB + Chlor (blank)	Hour 6:	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4	Device 5	Device 6	LB + Chlor (blank)
24965424	30040686	2782015	Colony 1, Replicate 1	0,291	0,253	0,245	0,276	0,265	0,23	0,249	0,265	0,044
24694246	32007030	2927911	Colony 1, Replicate 2	0,274	0,239	0,237	0,261	0,267	0,225	0,248	0,287	0,043
25801366	31393318	2751600	Colony 1, Replicate 3	0,278	0,26	0,228	0,269	0,286	0,231	0,249	0,266	0,043
26079876	33555608	2929858	Colony 1, Replicate 4	0,279	0,255	0,246	0,257	0,264	0,228	0,257	0,262	0,043
75463344	63873800	2790836	Colony 2, Replicate 1	0,286	0,262	0,253	0,285	0,282	0,244	0,254	0,28	0,042
82481440	71929584	3196464	Colony 2, Replicate 2	0,27	0,26	0,262	0,271	0,283	0,246	0,247	0,272	0,043
82881072	70888216	2801255	Colony 2, Replicate 3	0,277	0,268	0,259	0,276	0,279	0,245	0,253	0,273	0,041
79441768	72088576	3070474	Colony 2, Replicate 4	0,273	0,257	0,265	0,27	0,278	0,238	0,238	0,268	0,045

A7	A8	A9
B7	B8	B9
C7	C8	C9
D7	D8	D9
E7	E8	E9
F7	F8	F9
G7	G8	G9
H7	H8	H9

Unit Scaling Factors: These are imported from the prior sheets
C0007 / Abcd00
uM Fluorescein / s.u.

2.55
2.02E-09

Enter fluorescence and Abcd00 measurements into blue cells on "Raw Plate Reader Measurements"
Gold cells are calculated from values on other sheets

Experimental Values:

uM Fluorescein / OD

Hour 0:

	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4	Device 5	Device 6
Colony 1, Replicate 1	0.041	0.128	3.832	0.481	0.055	2.538	0.870	0.188
Colony 1, Replicate 2	0.043	0.265	3.782	0.456	0.051	2.446	0.786	0.182
Colony 1, Replicate 3	0.051	0.338	3.835	0.664	0.049	2.984	0.809	0.174
Colony 1, Replicate 4	0.036	0.278	3.770	0.550	0.051	2.488	0.780	0.152
Colony 2, Replicate 1	0.025	0.271	3.339	0.576	0.040	2.374	0.837	0.111
Colony 2, Replicate 2	0.021	0.268	3.622	0.606	0.042	2.102	1.041	0.384
Colony 2, Replicate 3	0.024	0.229	3.362	0.558	0.039	1.928	1.046	0.308
Colony 2, Replicate 4	0.028	0.254	4.288	0.625	0.042	2.024	1.008	0.423

Hour 6:

	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4	Device 5	Device 6
Colony 1, Replicate 1	0.004	0.187	0.513	0.444	0.010	0.971	0.088	0.338
Colony 1, Replicate 2	0.006	0.221	0.538	0.518	0.011	0.929	0.086	0.094
Colony 1, Replicate 3	0.007	0.186	0.550	0.471	0.011	0.871	0.090	0.304
Colony 1, Replicate 4	0.006	0.203	0.516	0.523	0.012	0.884	0.087	0.111
Colony 2, Replicate 1	0.005	0.139	0.448	0.473	0.009	0.573	0.277	0.208
Colony 2, Replicate 2	0.005	0.202	0.470	0.552	0.010	0.624	0.314	0.241
Colony 2, Replicate 3	0.006	0.191	0.451	0.509	0.011	0.686	0.305	0.237
Colony 2, Replicate 4	0.005	0.211	0.493	0.567	0.011	0.588	0.320	0.267

Net Fluorescein a.u.

	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4	Device 5	Device 6
819780.00	5688084.00	54114861.00	10808003.00	1153691.00	43424897.00	18978509.00	34953033.00	
823722.00	5642721.00	56117794.00	11803286.00	1141192.00	42466232.00	18718604.00	33964640.00	
1000880.00	5707095.00	56632210.00	11491800.00	1154484.00	47966768.00	15009126.00	3438641.00	
781452.00	5512586.00	50062126.00	10971915.00	1044130.00	41174868.00	14460314.00	1197078.00	
536662.00	536206.00	51832604.00	1087784.00	840962.00	41105768.00	23682628.00	6311886.00	
499494.00	5238201.00	52801037.00	11243967.00	928647.00	40102021.00	19361872.00	7299581.00	
469796.00	5271276.00	50404452.00	11789284.00	2215262.00	4089921.00	20211227.00	243847.00	
777625.00	5089471.00	57227743.00	11792585.00	1027630.00	40638145.00	21115127.00	7672262.00	

	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4	Device 5	Device 6
1259010.00	48128051.00	1274805226.00	12748021720.00	2751566.00	200342846.00	22833409.00	27258671.00	
1622082.00	51477741.00	124860193.00	139031257.00	3146095.00	208860241.00	21761151.00	29076110.00	
1867026.00	51626266.00	123851844.00	131447812.00	3185175.00	203979952.00	23494766.00	28641714.00	
1649646.00	52027586.00	1276487743.00	138302878.00	3446525.00	202187038.00	23150014.00	26257520.00	
1384914.00	48666206.00	117071775.00	141789576.00	2738742.00	120235344.00	708715068.00	61602664.00	
1313216.00	54515524.00	127151522.00	155601568.00	3018015.00	1571139472.00	70504076.00	68711120.00	
1715666.00	53983149.00	1271518677.00	147789581.00	3171217.00	151138521.00	88079617.00	68889811.00	
1309624.00	53843174.00	1233030620.00	157761284.00	3060778.00	140385446.00	78371294.00	49018102.00	

Net Abs 600

	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4	Device 5	Device 6
0.016	0.014	0.012	0.018	0.017	0.014	0.011	0.015	
0.016	0.016	0.012	0.015	0.014	0.014	0.011	0.015	
0.016	0.015	0.012	0.014	0.019	0.013	0.015	0.016	
0.017	0.016	0.012	0.015	0.017	0.015	0.015	0.017	
0.017	0.016	0.008	0.015	0.017	0.014	0.016	0.016	
0.016	0.016	0.012	0.015	0.018	0.016	0.015	0.015	
0.017	0.016	0.011	0.016	0.019	0.017	0.016	0.016	
0.018	0.016	0.011	0.016	0.020	0.016	0.017	0.017	

	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4	Device 5	Device 6
0.047	0.206	0.201	0.232	0.221	0.186	0.205	0.221	
0.211	0.196	0.194	0.218	0.224	0.182	0.208	0.244	
0.212	0.217	0.148	0.226	0.241	0.188	0.206	0.221	
0.216	0.212	0.200	0.214	0.221	0.185	0.214	0.219	
0.244	0.209	0.211	0.245	0.248	0.202	0.212	0.208	
0.227	0.217	0.219	0.228	0.240	0.205	0.204	0.229	
0.236	0.222	0.218	0.235	0.238	0.204	0.212	0.232	
0.228	0.212	0.220	0.225	0.231	0.193	0.201	0.221	

Unit Scaling Factors: *These are imported from the prior sheets*

Particles / Abs600	2,47E+08
MEFL / a.u.	1,22E+04

Enter fluorescence and At
Gold cells are calculated f

Experimental Values:**MEFL / particle****Hour 0:**

	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4
Colony 1, Replicate 1	2,52E+03	2,00E+04	2,22E+05	2,96E+04	3,34E+03	1,53E+05
Colony 1, Replicate 2	2,62E+03	1,74E+04	2,30E+05	3,87E+04	3,12E+03	1,49E+05
Colony 1, Replicate 3	3,11E+03	1,87E+04	2,32E+05	4,04E+04	2,99E+03	1,82E+05
Colony 1, Replicate 4	2,21E+03	1,70E+04	2,05E+05	3,60E+04	3,08E+03	1,64E+05
Colony 2, Replicate 1	1,54E+03	1,65E+04	3,19E+05	3,50E+04	2,44E+03	1,45E+05
Colony 2, Replicate 2	1,29E+03	1,62E+04	2,21E+05	3,69E+04	2,54E+03	1,32E+05
Colony 2, Replicate 3	1,88E+03	1,37E+04	2,61E+05	3,63E+04	2,39E+03	1,18E+05
Colony 2, Replicate 4	2,13E+03	1,79E+04	2,56E+05	3,69E+04	2,53E+03	1,25E+05

Hour 6:

	Neg. Control	Pos. Control	Device 1	Device 2	Device 3	Device 4
Colony 1, Replicate 1	2,51E+02	1,14E+04	3,12E+04	2,71E+04	6,14E+02	5,30E+04
Colony 1, Replicate 2	3,47E+02	1,34E+04	3,27E+04	3,14E+04	6,91E+02	5,65E+04
Colony 1, Replicate 3	4,00E+02	1,19E+04	3,35E+04	2,88E+04	6,45E+02	5,32E+04
Colony 1, Replicate 4	3,44E+02	1,23E+04	3,14E+04	3,18E+04	7,68E+02	5,38E+04
Colony 2, Replicate 1	2,79E+02	1,09E+04	2,73E+04	2,87E+04	5,20E+02	3,15E+04
Colony 2, Replicate 2	2,87E+02	1,23E+04	2,86E+04	3,36E+04	6,19E+02	3,81E+04
Colony 2, Replicate 3	3,58E+02	1,16E+04	2,74E+04	3,10E+04	6,56E+02	3,65E+04
Colony 2, Replicate 4	2,94E+02	1,30E+04	2,74E+04	3,45E+04	6,47E+02	3,58E+04

cence and Abs600 measurements into blue cells on "Raw Plate Reader Measurements"
calculated from values on other sheets

Net Fluorescein a.u.

Device 5	Device 6
5,29E+04	1,15E+04
4,78E+04	1,11E+04
4,93E+04	1,06E+04
4,75E+04	9,26E+03
3,88E+04	1,89E+04
6,34E+04	2,40E+04
6,21E+04	2,36E+04
6,13E+04	2,52E+04

Neg. Control	Pos. Control	Device 1	Device 2
819789,00	5680584,00	54194861,00	10808003,00
852722,00	5642731,00	56117734,00	11803286,00
1009880,00	5707395,00	56616210,00	11491800,00
763452,00	5522588,00	50001536,00	10971915,00
530563,00	5360248,00	51832604,00	10677954,00
499494,00	5268391,00	53901037,00	11243967,00
650790,00	5571278,00	58246143,00	11796348,00
777665,00	5809447,00	57237741,00	11976585,00

Device 5	Device 6
5,33E+03	6,07E+03
5,23E+03	5,87E+03
5,51E+03	6,32E+03
5,33E+03	6,88E+03
1,69E+04	1,26E+04
1,91E+04	1,48E+04
1,86E+04	1,44E+04
1,95E+04	1,52E+04

Neg. Control	Pos. Control	Device 1	Device 2
1259019,00	48236053,00	127485529,00	127492217,00
1629981,00	53477741,00	128960193,00	139033257,00
1907024,00	52262056,00	125855464,00	132147952,00
1649646,00	53037586,00	129549774,00	138302878,00
1384913,00	48616220,00	117071776,00	141739676,00
1323238,00	54215924,00	127215232,00	155601568,00
1715666,00	53380749,00	121516677,00	147780585,00
1359604,00	55843274,00	122355906,00	157763286,00

Device 3	Device 4	Device 5	Device 6
1153691,00	43424097,00	13978509,00	3495303,00
1141192,00	42346362,00	14574604,00	3596486,00
1154494,00	47966798,00	15009120,00	3439641,00
1064530,00	43174868,00	14460324,00	3197078,00
840962,00	41105708,00	19683528,00	6151680,00
928647,00	40107033,00	19301875,00	7299581,00
921530,00	40689547,00	20172107,00	7678347,00
1027639,00	40638145,00	21155127,00	7672260,00

Net Abs 600

Neg. Control	Pos. Control
0,016	0,014
0,016	0,016
0,016	0,015
0,017	0,016
0,017	0,016
0,019	0,016
0,017	0,020
0,018	0,016

Device 3	Device 4	Device 5	Device 6
2755565,00	200242849,00	22183409,00	27258671,00
3146095,00	208986041,00	21766335,00	29079119,00
3185175,00	202979952,00	23049766,00	28641718,00
3445525,00	202187038,00	23150018,00	30625750,00
2536742,00	129235364,00	72672508,00	61082964,00
3018015,00	157133472,00	79284976,00	68733120,00
3171217,00	151330521,00	80079817,00	68086961,00
3062778,00	140385446,00	76371294,00	69018102,00

Neg. Control	Pos. Control
0,247	0,209
0,231	0,196
0,235	0,217
0,236	0,212
0,244	0,220
0,227	0,217
0,236	0,227
0,228	0,212

Device 1	Device 2	Device 3	Device 4	Device 5	Device 6
0,012	0,018	0,017	0,014	0,013	0,015
0,012	0,015	0,018	0,014	0,015	0,016
0,012	0,014	0,019	0,013	0,015	0,016
0,012	0,015	0,017	0,013	0,015	0,017
0,008	0,015	0,017	0,014	0,025	0,016
0,012	0,015	0,018	0,015	0,015	0,015
0,011	0,016	0,019	0,017	0,016	0,016
0,011	0,016	0,020	0,016	0,017	0,015

Device 1	Device 2	Device 3	Device 4	Device 5	Device 6
0,201	0,232	0,221	0,186	0,205	0,221
0,194	0,218	0,224	0,182	0,205	0,244
0,185	0,226	0,243	0,188	0,206	0,223
0,203	0,214	0,221	0,185	0,214	0,219
0,211	0,243	0,240	0,202	0,212	0,238
0,219	0,228	0,240	0,203	0,204	0,229
0,218	0,235	0,238	0,204	0,212	0,232
0,220	0,225	0,233	0,193	0,193	0,223