

Application Data Sheet

No. 1

UV
UV-Visible Spectroscopy

Quantitation of dsDNA Using the Micro-Volume BioSpec-nano Spectrophotometer and its Repeatability

The rapid quantitation of DNA is an important and critical function of the modern biological and biochemistry laboratory. The Shimadzu BioSpec-nano micro-volume spectrophotometer is an ideal instrument to meet this critical need. The BioSpec-nano can determine the concentration of DNA every fifteen seconds by placing a 1-2 uL drop of sample directly onto the sample target and pressing the "Start" button. The DNA quantitation value is displayed within seconds followed by automatic wiping of the sample target, readying the instrument for the next sample. Measurement, recording, and wiping all occur in seconds allowing for high operational performance and throughput, a perfect fit for the modern laboratory. In addition, the Shimadzu design provides for the accuracy and repeatability required in today's research settings.



Excellent linearity from low concentrations to high concentrations with high repeatability

Table 1 shows the experimental values of dsDNA samples obtained with the optical path length of 0.7mm. The measurement was performed ten times for each sample. The good match between the prepared concentration and the experimental values and the high repeatability for each concentration can be found from Table 1.

Table.1 Experimental values of dsDNA samples obtained with the optical path length of 0.7mm.

	Experimental value									
Prepared concentration	0.00	1.25	3.50	5.00	14.00	28.50	51.50	125.50	250.00	525.00
Trial1	-0.64	1.27	2.72	4.31	13.19	28.00	51.33	126.94	251.48	511.38
Trial2	-1.43	1.01	3.06	5.56	14.07	26.62	51.00	125.55	252.00	513.94
Trial3	-0.14	0.73	4.03	4.59	14.40	25.97	51.80	125.99	253.27	515.62
Trial4	-0.61	1.19	3.53	6.82	14.11	27.18	52.92	125.18	254.06	512.72
Trial5	0.25	1.22	3.41	5.51	13.18	26.68	50.25	126.80	257.38	518.72
Trial6	0.16	1.63	2.73	4.55	13.80	27.29	50.83	124.57	252.80	512.39
Trial7	0.25	1.57	2.90	5.05	15.21	26.72	50.27	125.96	252.81	514.52
Trial8	-0.77	1.01	3.16	3.88	13.22	26.44	50.72	126.08	251.55	513.06
Trial9	-0.57	1.37	3.15	5.46	14.00	27.03	52.33	126.26	252.16	516.39
Trial10	-1.05	1.46	3.22	5.12	15.16	27.95	50.66	125.99	254.54	514.55
Average	-0.46	1.25	3.19	5.09	14.03	26.99	51.21	125.93	253.21	514.33
Standard deviation	0.57	0.28	0.40	0.83	0.74	0.64	0.89	0.70	1.78	2.16

Good match

High repeatability

Figure 1 shows the correlation between the prepared concentration of dsDNA and the experimental values obtained with the optical path length of 0.7mm. The good correlation with $R^2=0.9997$ is obtained.

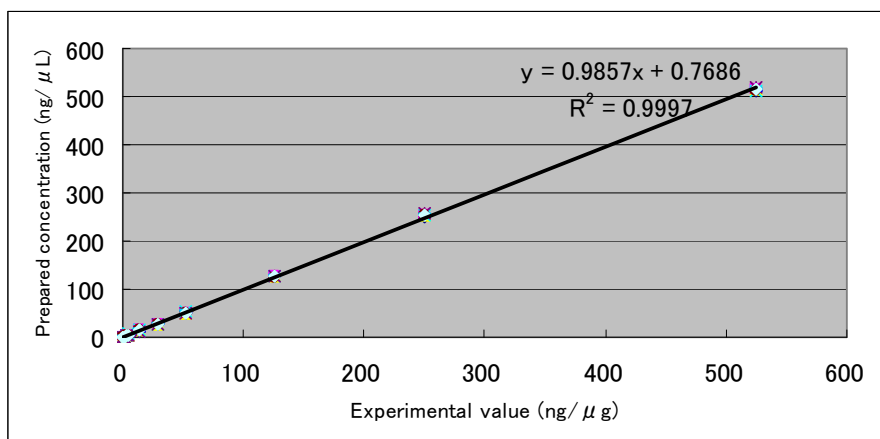


Figure 1 Correlation between the prepared concentration of dsDNA and the experimental values obtained with the optical path length of 0.7mm

Figure 2 is a graph of Figure 1 expanded in the range from 0 ng/μL to 30 ng/μL. The good correlation between the prepared concentration and the experimental values is shown even in the region of the low concentration.

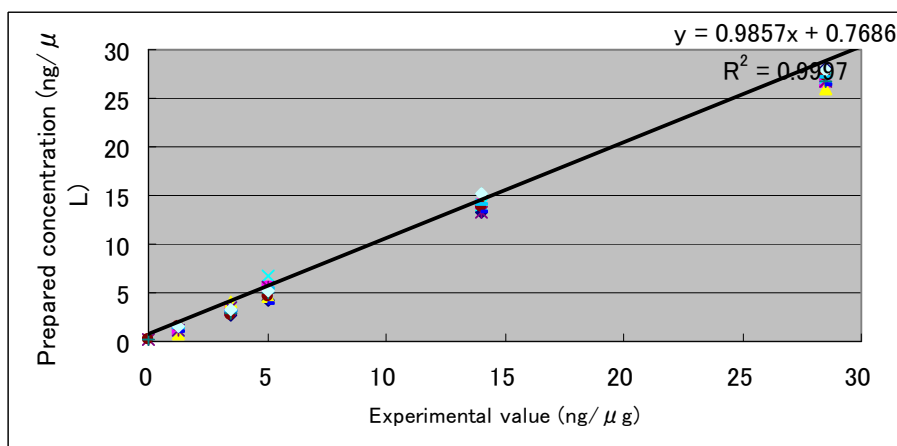


Figure 2 Expanded graph of Figure 1 in the range from 0 to 30 ng/μL

Figure 3 shows the experimental values of dsDNA sample of 1.25 ng/μL and the blank shown in Table 1 for both ten times measurements. The average and standard deviation of the experimental values for the concentration of 1.25 ng/μL are 1.25ng/μL and 0.28, respectively as shown in Table 1. Those for the blank are -0.46ng/μL and 0.57, respectively. The significant difference between the low concentration sample and the blank can be found from Figure.3.

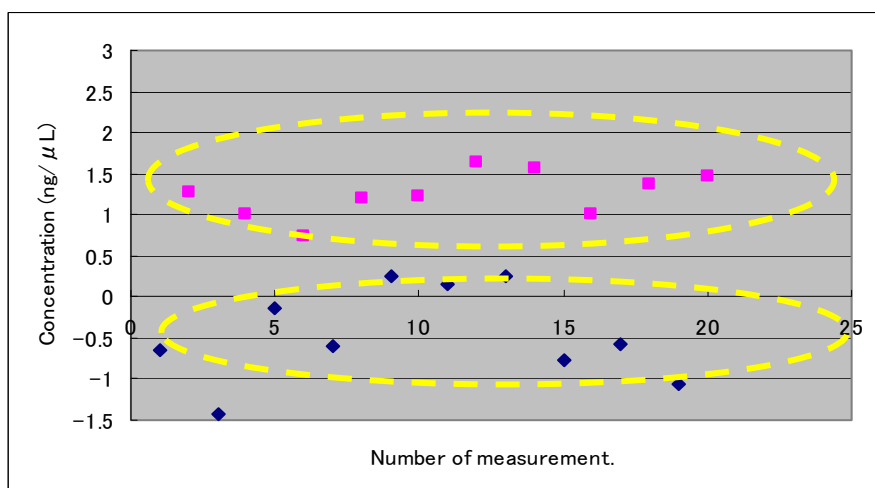


Figure 3 Experimental values of dsDNA sample (1.25ng/μL) and blank obtained with the optical path length of 0.7mm
Pink : dsDNA sample of 1.25ng/μL , Blue : blank

Using the equation for detection limits given in reference 1, a detection limit of 0.28 ng/μL could be calculated with an 85% confidence limit.

Reference 1

Peters, Hayes, Hieftje, "Chemical Separations and Measurements", W.B. Saunders company, Philadelphia, 1974, p. 29.