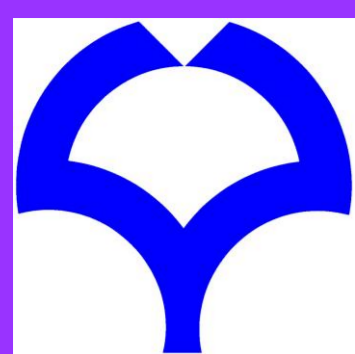


Investigation of Reprocessing of Flexible Endoscope Using dsDNA Measurement Methods



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Background

Cleanliness of flexible endoscopes can be examined by measuring proteins or ATP, and disinfection can be determined by bacterial culture. Both methods require intricate or costly procedures. However, it is now possible to perform easy, highly sensitive double stranded DNA measurements by fluorescent labelling.

Methods

We injected sterile distilled water into the suction channel of flexible endoscopes and measured protein (using the BCA method), ATP, double stranded DNA, and viable bacteria. Protein, Viable bacteria, dsDNA, ATP levels were calculated per unit surface area of the lumen. Comparisons were performed under three conditions: uncleaned (n=42); manual cleaning (n=21); and manual cleaning followed by reprocessing using the AER ESR-100 (Fuji Film Medical Systems, Tokyo, Japan), PAA disinfection, and alcohol flush (n=21).

Results

For uncleaned flexible endoscopes, average values for protein, double stranded DNA, ATP, and viable bacteria were 0.43 $\mu\text{g}/\text{cm}^2$, 4.66 ng/cm^2 , 24.5 RLU/ cm^2 , and 149463.3 CFU/ cm^2 , respectively. For manually cleaned flexible endoscopes, the values were 0.06 $\mu\text{g}/\text{cm}^2$, 2.07 ng/cm^2 , 10.3 RLU/ cm^2 , and 1.4 CFU/ cm^2 . For AER-reprocessed endoscopes, the values were 0.09 $\mu\text{g}/\text{cm}^2$, 0.40 ng/cm^2 , 16.3 RLU/ cm^2 , and 22.8 CFU/ cm^2 . Five of 21 flexible endoscopes were positive for bacteria after full processing.

Process	Protein			double stranded DNA			ATP			Viable Bacteria		
	After Bedside Cleaning	After Manual Cleaning	Full Processing	After Bedside Cleaning	After Manual Cleaning	Full Processing	After Bedside Cleaning	After Manual Cleaning	Full Processing	After Bedside Cleaning	After Manual Cleaning	Full Processing
Unit	$\mu\text{g}/\text{cm}^2$			ng/cm^2			RLU/ cm^2			CFU/ cm^2		
n	42	21	21	42	21	21	42	21	21	42	21	21
Mean	0.425	0.064	0.093	4.664	2.072	0.399	24.51	10.31	16.30	149463.34	1.42	22.83
SD	0.54	0.05	0.04	8.29	2.19	0.40	35.5	8.4	11.8	535610.5	4.0	104.6
Median	0.15	0.06	0.08	1.67	0.77	0.26	11.9	6.8	12.7	54.7	0.0	0.0
Range	0.02 - 2.15	0.00 - 0.16	0.04 - 0.19	0.00 - 38.56	0.18 - 7.06	0.00 - 1.46	2.2 - 229.7	1.2 - 28.4	4.0 - 42.4	0.0 - 3250591.0	0.0 - 13.4	0.0 - 479.1

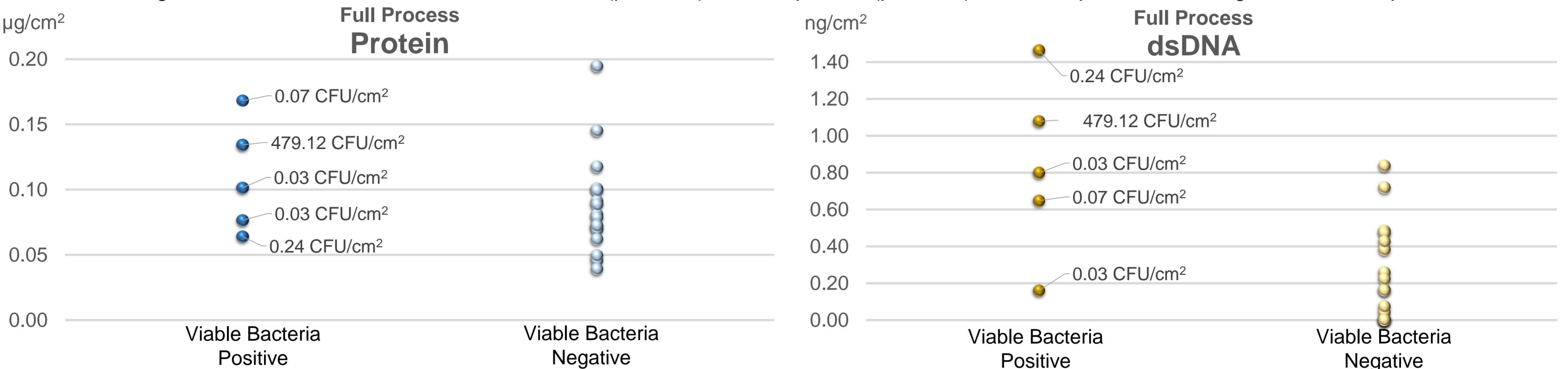
Spearman's rank correlation coefficients, based on all measurement results (n=84), were $\rho=0.293$ ($p < .005$) for protein and double stranded DNA, $\rho=0.458$ ($p < .0001$) for viable bacteria and protein, $\rho=0.558$ ($p < .0001$) for viable bacteria and double stranded DNA, i.e., double stranded DNA was more closely correlated than protein to the number of viable bacteria.

ρ	Protein	dsDNA	ATP	Viable Bacteria
Protein				
dsDNA	0.293***			
ATP	0.331****	0.079		
Viable Bacteria	0.458*****	0.558*****	0.103	

Prob > | ρ | * < .05, ** < .01, *** < .005, **** < .001, ***** < .0005, ***** < .0001

Furthermore, we compared protein and double stranded DNA measurements between disinfection endoscopes that tested positive for bacteria (n=5) and those that were negative (n=16).

There was a significant difference in double stranded DNA ($p < 0.05$), but not protein ($p > 0.05$), between positive and negative endoscopes.

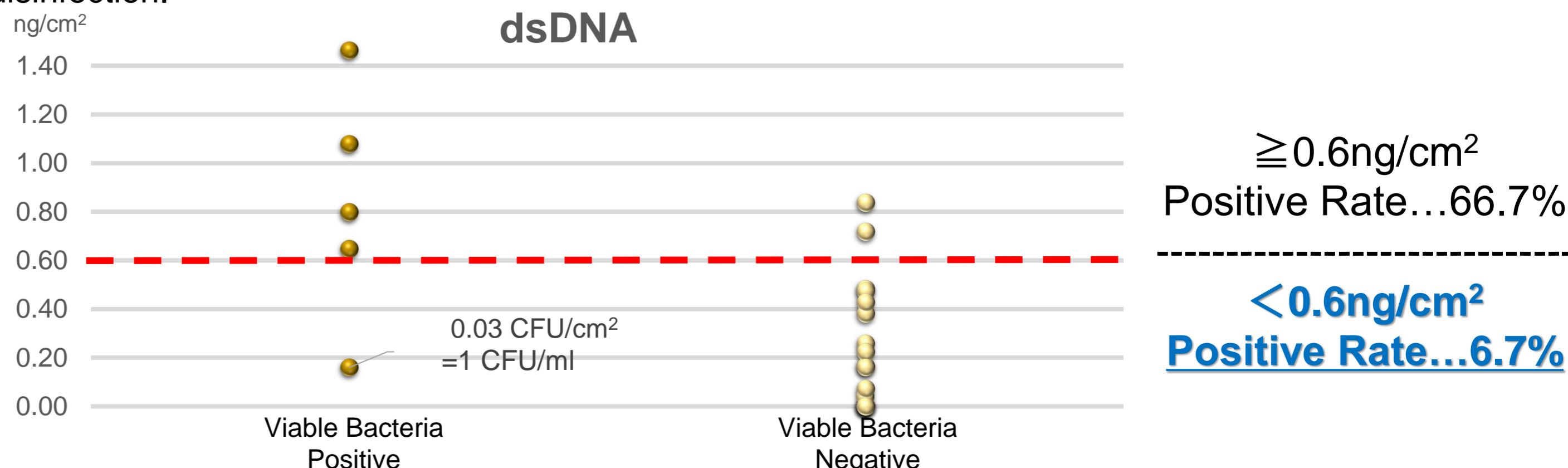


	Protein $\mu\text{g}/\text{cm}^2$		double stranded DNA ng/cm^2	
	Viable Bacteria Positive	Viable Bacteria Negative	Viable Bacteria Positive	Viable Bacteria Negative
n	5	16	5	16
Mean	0.109	0.088	0.829	0.264
SD	0.04	0.04	0.49	0.27
Median	0.10	0.08	0.80	0.19
Range	0.06 - 0.17	0.04 - 0.19	0.16 - 1.46	0.00 - 0.84
p value	> 0.05		< 0.05	

Dunn's test : p value ... Viable bacteria Positive vs Negative

The target value of double stranded DNA after cleaning and disinfection was set at 0.60 ng/cm^2 .

Six of 21 endoscopes had values ≥ 0.60 ng/cm^2 ; four of these six were positive for bacteria (positive rate, 66.7%). Moreover, 15 of 21 endoscopes had values < 0.60 ng/cm^2 , with one testing positive for bacteria (positive rate, 6.7%). However, the viable bacterial count was 0.03 CFU/ cm^2 (= 1 CFU/ml), within the permissible range for high-level disinfection.



Conclusion

Our data confirmed that flexible endoscopes subjected to complete reprocessing will have double stranded DNA levels ≤ 0.6 ng/cm^2 according to double stranded DNA test. Double stranded DNA measurement was better than any other intricate and costly procedure.