THE CLEANABILITY OF MATERIALS IN CONTACT WITH DAIRY PRODUCTS

II. THE CLEANABILITY OF METHYL METHACRYLATES¹

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Plastics will probably be used extensively in the manufacture of dairy equipment of the future if they have the following qualifications: (a) meet the requirements of the Food Additive Amendment of the Food, Drug and Cosmetic Act; (b) are as readily cleanable as stainless steel having a 120 grit finish; and (c) have the desired physical properties, such as adequate strength and resistance to distortion.

Determining the cleanability of these plastics will be an important function of sanitarians. Hucker (3), Hucker, Emery and Winkler (4), Mallmann, Kahler and Butt (6), and Ridenour and Armbruster (8) used various techniques in their studies of the cleanability of plastics used for eating utensils. Although none of these data are applicable to the present problem, the radiological technique of Ridenour and Armbruster (8) are useful tools in evaluating the cleanability of the plastics that may be used in the manufacture of dairy equipment.

Hays, Burroughs and Johns (2) and, more recently, Masurovsky and Jordan (7) employed radioactive tracer techniques in studies of the cleanability of milk contact surfaces. Hays, Burroughs and Johns (2) also reported on the use of bacteriological techniques, as did Kaufmann (5). Laboratory "use-test" techniques should not be discarded entirely for the more glamorous radiological techniques.

This paper evaluates the relative cleanability of methyl methacrylates and stainless steel by both bacteriological and radiological techniques.

EXPERIMENTAL

As in the previous study (2), disks approximately two inches in diameter were used as the test specimen of each material. Since Food and Drug Administration approval is given to a specific material for use in contact with foods rather than blanket approval to all material of a general type, the trade

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names of the acrylics studied will be used in this paper. The materials tested were:

- 1. Plexiglas II, UVA.
- 2. Plexiglas V, Type 607.
- 3. 18-8 stainless steel, 120 grit finish.

The general experimental plan consisted of soiling these disks with dairy products which had been contaminated previously with Escherichia coli. The products used were homogenized milk, buttermilk, cream, and chocolate milk. On each disk, 0.4 ml. of one of the contaminated products was spread evenly on one side only and allowed to air dry. The soiled disks then were cleaned by scrubbing with a circular motion for about 15 seconds with a test tube brush in a cleaning solution at room temperature. Hand scrubbing approximates the methods used in the field to clean dairy equipment. The following cleaning solutions were used: (a) distilled water (b) 0.25 per cent solution of an alkaline cleaner, (c) 0.25 per cent solution of an anionic detergent, (d) 0.25 per cent solution of a nonionic detergent, and (e) 6.7 per cent solution of an acid cleaner.

Although the techniques used in assaying the soil removal were essentially the same as were used in the previous study (2), they will be reviewed briefly.

Radiological Techniques

Dairy products contaminated with a P^{32} labeled $E.\ coli$ suspension, prepared after the method of Ridenour and Armbruster (8) were used to soil the material to be evaluated by radiological techniques.

After the contaminated disks were air dried, the radioactivity of each was determined in an end-window "Sugarman" type proportional counter. The disks were marked so that they could be placed in the same position in the counter after cleaning. After the initial counts had been made, the disks were cleaned and then rinsed in tap water. After drying, the disks were examined and any residual radioactivity was recorded. All counts were corrected for background and decay.

Bacteriological Techniques

Dairy products contaminated with washed cells of

a 24 hour nutrient broth culture of *E. coli* were used to soil the disks which were to be evaluated by bacteriological techniques. The number of viable cells recovered from two air dried disks was determined by culturing portions of the sterile water in which the disks were soaked and swabbed. This procedure was repeated with each dairy product on each of the three materials. These recovery counts were used as the initial counts in determining the per cent removal of *E. coli* from other contaminated disks scrubbed in the various cleaning solutions.

After the contaminated disks had been scrubbed by hand in the cleaning solution, then rinsed in tap water, they were placed in either 25 ml. of sterile water or in 25 ml. of one of the following germicidal solutions; (a) a hypochlorite solution with 100 ppm available chlorine, (b) an iodophor solution with 25 ppm iodine concentration, and (c) a solution of a quaternary ammonium compound containing 200 ppm active ingredient prepared in water having a natural hardness of 200 to 250 ppm.

The germicidal solutions were inactivated after the disks had been exposed for one minute. After the cleaned disks had been in the sterile water or inactivated germicidal solutions for approximately five minutes, the liquid was cultured in 5 aliquots of 5 ml. each. The disks were also placed in petri dishes with the previously soiled side up. All plates were poured with Difco-brilliant green bile agar to which 0.5 per cent agar had been added so that the 5 ml. aliquots could be cultured in single plates.

RESULTS

The results of this study are given in the four accompanying tables. These tables show the per cent of *E. coli* removed from the surface of the test material as determined by both bacteriological techniques and the radioactive tracer method.

As noted in Tables 1 through 4, both residual coliform counts and residual radioactivity indicated that 99.80 per cent or more of the contaminating soil was removed by scrubbing the disks in any of the cleaning solutions used or even in distilled water.

All of the cleaned disks were sanitized within one minute upon exposure to the germicidal solutions.

The statistical analyses of the radiological data are given in Tables 5 through 8.

DISCUSSION

One P^{32} labeled $E.\ coli$ suspension and one non-labeled suspension were used in this study. Although refrigerated, the number of viable cells in the non-labeled suspension gradually diminished from 420 x 10^6 to 57.12×10^6 before the bacteriological phase of this study was completed. This in part accounts for the variation in initial counts noted in the tables. The initial counts never approached the number of organisms observed when $0.4 \, \text{ml.}$ of the suspension was cultured. This difference may have been due to organisms becoming non-viable during drying or the cells remaining on the disks after soaking and swabbing. The latter seems improbable as the disks were

TABLE 1-THE CLEANIBILITY OF SURFACES SOILED WITH HOMOGENIZED MILK

		Bacteriological	techniques			Radio	ogical techniques	
Materials	Type of cleaner	Initiala colony counts (x106)	Resi colony		Average per cent removal	Initial ^b radioactivity (x10 ³)	Residual ^b radioactivity	Average per cent removal
Plexiglas II,	Alkaline	. 0.26, 4.00	0,	0	100.00	4.67, 4.74	0.55, 0.22	99.99
UVA	Anionic		0,	3	99.99	4.51, 5.88	0.55, 0.44	99 99
	Nonionic		0,	199	99.99	5.46, 5.85	1.76, 6.54	99.93
	Acid		0,	24	99.99	5.04, 5.05	-0.28, 1.29	99.99
	Water		0,	2	99.99	4.79, 5.03	3.64, 1.82	99.94
Plexiglas V,	Alkaline	0.46, 2.25	0,	0	100.00	4.57, 5.29	-1.53, -3.60	100.05
Type 607	Anionic		0,	0 .	100.00	4.72, 5.90	-0.27, -0.72	100.01
	Nonionic		0,	9 .	100.00	4.81, 5.28	-0.37, 0.63	99.99
	Acid		0,	1	99.99	5.38, 5.41	1.39, -3.50	100.01
	Water		0,	103	99.99	4.85, 5.03	5.04, 2 .90	99.92
18-8 (120 grit)	Alkaline	0.27, 8.90	0,	0	100.00	4.36, 5.34	6.83, 4.15	99.88
	Anionic		0,	0	100.00	5.53, 6.20	7.48, -0.09	99.93
	Nonionic		0,	0	100.00	4.60, 5.58	9.56, 8.30	99.82
	Acid		0,	0	100.00	5.71, 5.80	4.08, 2.68	99 94
	Water		1,	94	99.99	5.38, 5.43	8.37, 9.33	99.83

^aInitial colony counts are the *E. coli* colony counts recovered from the air dried disks by culturing portions of the water in which the soiled disks were soaked and swabbed.

^bRadioactivity recorded as counts/minute corrected for background and decay.

always noted to be visibly clean at the time the initial counts were made. An exceptionally poor recovery count (initial count) was noted from the disks soiled with buttermilk. In one series, less than 10 coliforms per disk were recovered although a calcu-

lated 2.36×10^6 organisms had been placed on these disks. No doubt, the acidity of the buttermilk enhanced the lethality noticed during the drying of other dairy products.

Colonies were not observed in the cultures of 99

TABLE 2-THE CLEANABILITY OF SURFACES SOILED WITH CHOCOLATE MILK

	4	Bacteriological	techniques			Radio	logical techniques	
Materials	Type of cleaner	Initial ^a colony counts (x10 ⁶)	Resi colony		Average per cent removal	Initial ^b radioactivity (x10 ³)	Residual ^b radioactivity	Average per cent removal
Plexiglas II,	Alkaline	1.10, 10.50	0,	0	100.00	4.71, 6.04	-1.35, -0.54	100.02
UVA	Anionic		0,	0	100.00	4.75, 5.65	0.11, -0.45	100.00
	Nonionic		0,	45	99.99	4.53, 5.38	3.42, 4.15	99.92
	\mathbf{Acid}		0,	0	100.00	5.11, 5.23	2.47, 1.29	99.96
	Water		14,	2000	99.99	5.38, 5.56	2.49, 3.94	99.94
Plexiglas V,	Alkaline	1.65, 12.25	0,	0	100.00	4.49, 5.96	-2.07, 0.44	100.02
Type 607	Anionic		0,	0	100.00	4.73, 5.42	-1.17, 0.22	100.01
	Nonionic		0,	0	100.00	4.82, 5.06	2.14, 4.90	99.93
	Acid		0,	0	100.00	5.71, 5.92	-0.46, 0.86	100,00
	Water		0,	65	99.99	5.29, 5.67	0.52, -1.04	100.00
18-8 (120 grit)	Alkaline	1.17, 5.50	0,	0	100.00	4.78, 4.82	1.76, 1.21	99 97
	Anionic		0,	0	100.00	4.49, 5.57	2.09, 0.99	99.96
	Nonionic		0,	13	99.99	4.96, 5.87	7.42, 0.63	99.92
	\mathbf{Acid}		0,	0	100.00	4.94, 6.68	2.47, 0.75	99.96
	Water		16,	175	99.99	5.52, 5.59	3.84, 0,21	99.96

[&]quot;Initial colony counts are the E. coli colony counts recovered from the air dried disks by culturing portions of the water in which the soiled disks were soaked and swabbed.

TABLE 3-THE CLEANABILITY OF SURFACES SOILED WITH BUTTERMILK

		Bacteriological	techniques		Radio	logical techniques	
Materials	Type of cleaner	Initiala colony counts (x10 ⁶)	Residual colony counts	Average per cent removal	Initial ^b radioactivity (x10 ³)	Residual radioactivity	Average per cent removal
Plexiglas II,	Alkaline	(c), 0.0037	0, 1	99.99	2.55, 7.69	-0.70, 0.37	100,01
UVA	Anionic		0, 0	100.00	5.58, 7.71	-3.51, -3.60	100.05
	Nonionic		0, 0	100.00	1.17, 1.54	-1.19, 0.88	100.02
	Acid		0, 0	100.00	5.49, 5.62	1.82, 4.08	99.95
	Water		0, 0	100.00	5.77, 5.68	1.50, 1.50	99.97
Plexiglas V,	Alkaline	(c), 0.008	0, 0	100.00	0.96, 7.13	1.59, -0.47	99.92
Type 607	Anionic		0, 0	100.00	4.85, 6.05	-1.17, 0.00	100.01
	Nonionie		0, 0	100.00	1.83, 5.52	-1.14, -2.30	100,06
	Acid		0, 0	100.00	5.66, 6.02	-0.27, 1.10	99.99
	Water		0, 0	100.00	5.52, 5.92	1.61, 0.21	99.98
18-8 (120 grit)	Alkaline	(c), 0.0022	0, 0	100.00	4.77, 5.03	4.27, 4.88	99.91
	Anionic	, ,,	0, 0	100.00	5.98, 6.70	2.09, 5.17	99.93
	Nonionic		0, 0	100.00	2.99, 3.94	1.64, -0.89	99.98
	Acid		0, 0	100.00	6.42, 6.56	2.43, 1.76	99.96
	Water		0, 2	99.99	6.47, 6.79	6.44, 3.75	99.92

[&]quot;Initial colony counts are the E. coli colony counts recovered from the air dried disks by culturing portions of the water in which the soiled disks were soaked and swabbed.

^bRadioactivity recorded as counts/minute corrected for background and decay.

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^{&#}x27;Initial counts of some of the disks were less than 10 colonies per disk, although a calculated 2.36×10^6 organisms were in the buttermilk placed on these disks.

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TABLE 4-THE CLEANABILITY OF SURFACES SOILED WITH CREAM

		Bacteriological	techniques		Radio	logical techniques	
Materials	Type of cleaner	Initial ^a colony counts (x10 ⁶)	Residual colony coun	Average per cent ts removal	Initial ^b radioactivity (x10 ³)	Residual radioactivity	Average per cent removal
Plexiglas II,	Alkaline	0.325, 0.437	0, 0	100.00	4.89, 5.04	-1.33, 0.24	100.00
UVA	Anionic		0, 0	100,00	4.70, 4.92	-0.09, -1.98	100.02
	Nonionic		0, 0	100.00	4.82, 4.84	-1.93, -0.74	100.03
	Acid		0, 0	100.00	4.81, 5.09	3.31, 0.66	99,96
	Water	·	0. 11	99.99	5.23, 5.23	-2.04, 0.43	100 02
Plexiglas V,	Alkaline	0.250, 0.382	0, 0	100.00	4.88, 5.21	-0.47, -0.94	100.01
Type 607	Anionic		0, 1	99.99	4.93, 5.35	-4.05, -5.40	100.09
	Nonionic		0, 0	100.00	5.16, 5.19	-0.07, -0.15	100.00
	Acid		0, 0	100.00	5.03, 5.11	-2.45, 2.65	100.00
	Water	k.	0, 29	99.99	5.22, 5.33	6.01, 4.72	99.89
18-8 (120 grit)	Alkaline	0.242, 0.495	0, 0	100.00	4.93, 5.22	3.78, 3.90	99.92
	Anionic -	·	0, 0	100.00	5.39, 6.48	0.22, 0.88	99.99
	Nonionic		0, 0	100.00	5.32, 6.44	-0.22, 5.28	99,95
	Acid		0, 1	99.99	5.45, 5.75	1.65, 3.53	99.95
	Water		0, 0	100.00	5.65, 5.70	5.47, 9.76	99.86

^{*}Initial colony counts are the E. coli colony counts recovered from the air dried disks by culturing portions of the water in which the soiled disks were soaked and swabbed.

of the 120 disks after scrubbing in the cleaning solutions or distilled water. Of the 21 contaminated disks, twelve were scrubbed only in distilled water. The residual counts of these disks ranged from 2 to 2000 colonies (Table 2) with only three having a colony count in excess of 100 per disk (Tables 1 and 2). Only one of the disks that were scrubbed in a cleaning solution had a count in excess of 100. The residual count of this Plexiglas II UVA disk was 199. This disk had been contaminated with 45.2 x 10^6 coliforms in homogenized milk and cleaned with a nonionic detergent. (See Table 1).

The initial count of radioactivity ranged from 7,710 to an unexplained low of 960 per minute. In most cases, the counts per minute for the two disks used for each treatment were within 1000 of each other.

Residual radioactivity is measured by subtracting the experimentally determined background count from a reading of the sample being investigated. When the residual radioactivity is of a very low order, this reading would be very close to the background count. Depending upon how close to zero the residual radioactivity actually is, up to half of these counts could be negative, since both the background and radioactivity counts are subject to random variation. During the period in which the radiological data were collected, the background counts varied from 34 to 41 counts/minute. The standard deviation (1) of background count was about 2 counts/minute. Under these conditions 1 per cent of the

measurements would be expected to have negative counts as large as minus 6.

The residual radioactivity of 39 of the 120 disks was noted to range from zero to minus 5.40 counts/minute. These negative values were a natural consequence of the random distribution of counts of very low order, as mentioned above.

Radioactivity was observed on 21 of the 24 disks cleaned in distilled water. The residual activity of these disks ranged from 0.21 to 9.76 counts/minute. The latter count was obtained from a stainless steel disk which had been contaminated with cream. (See Tables 2 through 4).

TABLE 5-ANALYSIS OF VARIANCE (RADIOLOGICAL DATA)"

Source of variation Degree	of freedom	Mean square
Type of cleaner	4	124.81 ^b
Dairy product	3	70.21^{b}
Surface tested	2	523.08 ^b
Cleaner x dairy product	12	56.28 ^b
Cleaner x surface tested	8	21.01 N.S.
Dairy product x surface tested	6	43.61°
Cleaner x dairy product x surface	24	19.64 N. S.
Replication	60	15.97
	-	
Total	119	

^{*}After Snedecor (9).

^bRadioactivity recorded as counts/minute corrected for background and decay.

^bSignificant to .01 level

[°]Significant to .05 level

N. S. = Not Significant

Table 6-Effect of Surface Tested on Per Cent Contamination Removed

Surface	Average per cent contamination removed		
Plexiglas V, Type 607	100.00) (2		
Plexiglas II, UVA	$ \begin{array}{c c} 100.00 \\ 99.99 \end{array} $ (a)		
18-8 (120)	99.83		

L. S. D. (1% level) between two surface averages = 0.02%. (a) Averages enclosed in brackets are not significantly different.

Table 7-Effect of Cleaning Agent on Per Cent Contamination Removed

Cleaning agent	Average per cent contamination removed			
Anionic Detergent	100.00			
Alkaline Cleaner	99.98			
Acid Cleaner	99.97 × (a)			
Nonionic Detergent	99.96			
Water	99,94 ′			

L.S.D. (5% level) between two cleaning agents average = .02% (a) Averages enclosed in brackets are not significantly different.

TABLE 8-EFFECT OF DAIRY PRODUCT ON PER CENT CONTAMINATION REMOVED

Dairy product	Average per cent contamination removed
Buttermilk	99.98)
Cream	99.98 } (a
Chocolate Milk	99.98 } (a 99.97 }
Homogenized Milk	99.95

L. S. D. (5% level) between two product averages = 0.02% (a) Averages enclosed in brackets are not significantly different.

Residual radioactivity was noted on 60 of the 96 disks which had been scrubbed in the various cleaning solutions. These counts ranged from 0.07 to 9.56 per minute. The surface showing the highest residual radioactivity was a stainless steel disk soiled with homogenized milk and cleaned in a nonionic detergent (Table 1).

To complete both the bacteriological and radiological evaluation of the cleanability of these materials, the disks had to be soiled at least twice during each phase of the investigation. The phenomenon of soil build-up reported by Masurovsky and Jordan (7) was not observed.

SUMMARY

The relative cleanability of Plexiglas II, UVA; Plexiglas V, Type 607; and 18-8 stainless steel having a 120 grit finish has been evaluated by both bacteriological and radiological techniques.

Even with distilled water, 99.80 per cent or more of the contaminating soil was removed from the test materials.

All disks were sanitized within one minute by the hypochlorite, iodophor, and quaternary ammonium compound solutions at the concentration used.

It may be concluded from these data that Plexiglas II, UVA and Plexiglas V, Type 607 are as readily cleanable as is 18-8 stainless steel with a 120 grit finish.

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