

ample, in an experiment on the corpuscles of beef blood, a value of 0.34 for w was obtained. The fraction of water in beef corpuscles is about 0.60. The discrepancy may possibly be explained by the presence of water not active as a solvent ("bound water") at the temperature of the freezing-point. Accordingly, the bound water content of the beef corpuscles in the above experiment is 26%.

Thus the equation, $\Delta = \Delta^1 - \frac{s u \Delta^1}{80 w}$, may be used in correcting for undercooling in the determination of the freezing-point in solutions of high solid content, the w in the equation for a given material being determined from freezing-point data on undercooling. Where large amounts of solid are present, it is important to make this correction. The undercooling data also provide a method for determining bound water in a rough manner; that is, the fraction of total water present minus the w of the above equation equals the fraction of the bound water.

NOTE: Dr. F. H. MacDougall has kindly developed an equation in which the specific heats of the various components are taken into account separately.

$$\Delta = \Delta^1 - \left[1 + \frac{(w_2 + w_3 c)}{w_1} \right] \frac{u}{80} \Delta^1.$$

In this equation w_1 equals the fraction of actual solution, w_2 the fraction of bound water, w_3 the fraction of solid material, and c the specific heat of the solids. The specific heats of the solution and of the bound water are assumed to be equal to unity.

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Action of Ultra-Violet Rays on New and Old Bacterial Cultures.

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Cultures of bacteria placed on agar are as a rule easily killed by exposure to ultra-violet light from a quartz mercury arc. The amount of radiation necessary for devitalization depends upon several factors. Perhaps the most intricate of these is the variation of resistance with age. This relation between resistance and age has not been determined in detail. The following study was aimed as a contribution to our knowledge in this respect.

In the following experiments a new Victor quartz mercury arc lamp was used without any filter. The current was 4.5 amperes at 65 volts and the distance from burner to plate was 24 inches.

An old culture was one that was grown and transplanted daily for one week and then sealed and left in the museum till needed; a period of from one to 6 weeks.

A new culture was one that was grown and transplanted daily from the old culture for a period of 5 weeks or so. *Coli communis* was used as the culture in most of our experiments, a control being run on each plate irradiated.

Series I. A small amount of culture was removed with a wire loop 2 mm. in diameter and this was mixed with 2 cc. water. Then one loopful of this suspension was spread on one-half of an agar plate and another loopful on the other half and these were marked "control" and "irradiated".

Series II. As much of the suspension as would adhere to the wire loop was removed from the agar after the exposure and planted in 5 cc. of beef broth. The same was done with the controls.

All cultures were irradiated for $\frac{3}{4}$ min. Four plates were used in each set of experiments. One irradiated immediately on transplanting from old or new culture, one each irradiated 1, 3 and 6 hours after transplanting with the results as shown on chart Series I and Series II.

In the above series where only 2 or 3 broth or agar plates showed growth at $\frac{3}{4}$ min. no growth was visible if a similar plate was irradiated for 1 min.

In Series I and II, 4 new and 4 old plates were irradiated together. The number which showed growth after exposure is shown in Column 3.

	Age of Culture	Number of Plates	
		Growth	No Growth
Series I	38 days	0	4
	1 "	4	0
	34 "	1	3
	1 "	4	0
	38 "	0	4
	1 "	3	1
	34 "	1	3
	1 "	3	1
Series II	38 "	1	3
	1 "	3	1
	34 "	2	2
	1 "	4	0
	38 "	3	1
	1 "	4	0
	34 "	1	3
	1 "	4	0

In the above two series 29 out of 32 new cultures resisted radiation whereas 9 out of 32 old cultures grew after the exposure.

In another series of experiments it was found that portions of old cultures which were grown for 1, 3, 6, 12, 18 and 24 hours and then irradiated were more resistant to ultra-violet than portions of the same cultures which were irradiated immediately after transplanting. Thus even after one hour some of the cultures showed still greater resistance. They gained resistance from the time of transplantation up to about 24 hours, but their resistance to ultra-violet was diminished with greater age as shown in Series I and II.

The "increased resistance" of the colonies might be due either to greater individual resistance or to the greater number of bacteria present some time after transplantation. We tried to differentiate between these factors by means of bacterial counts.

Consistant results of the counts were not obtained when the bacteria were planted on agar and therefore a peptone solution was used instead for these experiments.

One cc. peptone culture media with one wire loopful of *Coli communis* was diluted with 99 cc. sterile water to make a 1/100 solution. This was No. 1. Ten cc. of this No. 1 was diluted with 90 cc. water yielding a 1/1000 solution labeled No. 2. Ten cc. of No. 2 solution was added to 90 cc. water, giving a 1/10,000 dilution. This was solution No. 3. The diluting was continued in the same way until a 1/10,000,000 solution was obtained, *i. e.*, solution No. 6.

One-fourth cc. of each of the above solutions were spread on agar plates and irradiated. If more than $\frac{1}{4}$ cc. was used the solution would adhere to the sides of the plate by capillary attraction and the rays could not reach it. One cc. of each dilution was used and suspended in agar and plated for our control count.

We multiplied the number of colonies left after irradiation by 4 to get results based on one cc. of the suspended culture.

In Series V, VI, VII, VIII and IX the above was again repeated but with a greater difference in the ages of the various growths.

It is well known that older cultures have smaller numbers of bacteria than new cultures, and our experiments bring this out clearly. They also indicate, however, that the individual resistance of the bacteria to ultra-violet light is reduced. (This is interesting to note, as it is generally stated that old bacteria are more resistant to heat, boiling, antiseptics, etc.)

We found that the resisting powers toward the ultra-violet rays are quickly built up by reculturing, even as in some cases by being

		Counts of No. of Colonies		Calc. No. of Colonies per 1 cc.	
		Not Irr. (per 1 cc.)	Irr. (per $\frac{1}{4}$ cc.)	Not Irr.	Irr.
Series V	1 Day Old				
	Dilution I	—	100	10,500,000	400
	Dilution II	—	25	1,050,000	100
	Dilution III	—	6	105,000	24
	Dilution IV	—	0	10,500	0
	Dilution V	—	4	1,050	16
Series VII	Dilution VI	105	0	105	0
	37 Days Old				
	Dilution I	—	3	53,000	12
	Dilution II	—	0	5,300	0
	Dilution III	—	—	530	—
	Dilution IV	53	—	53	—
Series VIII	Dilution V	20	—	5.3	—
	Dilution VI	2	—	0.53	—
	60 Days Old				
	Dilution I	—	4	1,050,000	16
	Dilution II	—	4	105,000	16
	Dilution III	—	0	10,500	0
Series IX	Dilution IV	—	0	1,050	0
	Dilution V	105	0	105	0
	Dilution VI	17	1	10.5	4
	80 Days Old				
	Dilution I	—	0	3,000	0
	Dilution II	—	0	300	0
	Dilution III	—	0	30	0
	Dilution IV	3	—	3	—
	Dilution V	1	—	.3	—
	Dilution VI	1	—	.03	—

grown on new media for only one hour. Cultures which have been grown for about 24 hours have the greatest resistance to ultra-violet ray. When they are transplanted their resistance drops down and increases again with time so that after 12 hours it has come up to almost the same resistance as that of a 24-hour culture.

In connection with the above experiments, some other studies were made of which we wish to mention the results.

A suspension of *Oidium albicans* in Saboraud Agar was plated and exposed to 2 minutes of ultra-violet light at 12 inches. It was found that enough rays to kill the bacteria penetrated through the agar whose thickness averaged 0.66 mm. according to micrometer measurements. Bacteria below 0.66 mm. showed definite growth.

Staphylococcus, *Oidium albicans*, *Proteus vulgaris* and *Aspergillus fumigatus* were also used in some experiments which were carried out as Series I but consistent results of the killing time were not obtained in these preliminary tests.

Suspensions of *Oidium albicans* in agar were irradiated with X-rays, a coverglass was the only filter used (190 kv. at 60 cm. from the tube, 30 milliamperes) for 3 hours. The growth seemed re-

tarded but on transplanting it grew normally. *Staphylococcus aureus* and *Coli communis* were also irradiated with the X-rays for varying lengths of time up to 5 hours but no change was noted in the growth of the bacteria.

We are very much indebted to Dr. Arthur T. Henrici for his help in supplying us with the cultures and for giving us advice and suggestions.

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Comparative Study of the Quantity of Gas in the Bowel in
Simple and Closed-Loop Obstruction.*

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The results of previous studies have indicated that whereas the accumulation of gas as detected by X-ray examination is a reliable agent in the early diagnosis of simple obstruction, in strangulation obstructions Roentgen evidence is not of the same value in its early recognition.^{1, 2, 3} The results of the following study are based on direct measurements of gas in simple and closed-loop obstructions.

Method: Studies of 36 dogs form the basis of this report. Dogs with closed-loop rather than strangulation obstructions were used in this comparison because the dogs with strangulation obstructions only survive for short periods. In the closed-loop, as a rule there are no gross vascular changes until late in the course of the obstruction at which time closed-loop obstruction partakes of the features of strangulation obstruction. In the closed loop obstruction the oral source of gas has been eliminated and in establishing this type of obstruction every trace of gas was "milked out" of the bowel before the last bowel end was inverted. Thirty-one closed-loop obstructions of small bowel of varying lengths and at various sites were established by the usual standard methods. The dogs were then observed for various lengths of time and either killed when they appeared moribund or were reoperated upon before they reached this stage. At operation the closed-loops were removed and im-

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¹ PROC. SOC. EXP. BIOL. AND MED., 1930, **27**, 674.

² PROC. SOC. EXP. BIOL. AND MED., 1930, **27**, 952.

³ PROC. SOC. EXP. BIOL. AND MED., 1931, **28**, 343.