

mately one-half their daughters were affected (Table 1).

The 10 sire-families not showing any affected offspring by 10 weeks of age were discarded; the 3 families containing affected chicks were kept intact for about 5 months to secure data on the frequency of the character. Mortality from causes other than the lethal gene was very low, and no males were affected by the character.

Subsequent tests of 10 other sons of heterozygous sires revealed that 4 were carriers, thus making a total of 7 carriers among 23 cockerels tested. These results do not differ significantly from the expected number of 11.5 carriers ($P=.05-.10$).

As was pointed out previously, 2 daughters of sire K-1 were able to survive all attacks of the condition and became, to all outward appearances at least, perfectly normal adults. As a further test of the hypothesis of sex-linkage, these 2 females (presumably $xl/-$) were mated to a known carrier male. In such a mating one-half the pullet chicks should have been affected as before, and, in addition, one-half the cockerel chicks should have received a sex chromosome bearing xl from each of their parents, and should therefore express the character. Only a limited number of progeny (35) was obtained from these matings. Among these chicks, 40% of each sex died. Of the 8 males that died, 5 did so suddenly, at ages ranging from 11 to 17 days. At autopsy no abnormal changes were seen. The 3 other sons died at later ages, and these all showed symptoms identical with those which distinguish pullets affected with the xl syndrome. We interpret these facts as evidence that at least the sons which showed the syndrome were homozygous for the sex-linked gene xl .

Preliminary physiological tests have yielded no substantial information as to the mode of action of the lethal gene. The attacks almost invariably develop during, or just following, a 10- to 14-hour period of darkness and quiet. Attempts to revive comatose birds by intramuscular injection of adrenalin, or by intravenous injection of glucose, calcium gluconate, or parathormone, were all unsuccessful.

One other sex-linked gene with lethal properties is known for the fowl. The sex-linked gene n for naked (3) is lethal to about half the naked chicks during incubation, and to most of those that hatch, unless these are brooded at high temperatures. In the case of the lethal gene xl , chicks destined to show the lethal syndrome cannot be distinguished from their normal siblings. In this respect it is similar to the delayed lethal studied in the rat by Crew and Kon (1). The presence of the gene xl in the hemizygous state is apparently not inevitably lethal since about 3% of such birds escape death.

When added to the lists of mutations previously recognized in the fowl (2), the xl lethal raises the number of known sex-linked genes to 12, and that of lethal genes to 22.

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A Precision Method of Counting Radioactive Liquid Samples¹

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The lack of an accurately reproducible method of preparing radioactive samples for counting has often prevented the use of radioactive tracers in precise analytical work. Most of the common methods for handling solid samples will generally give reproducible results with carrier-free tracers. However, the end result of an analytical procedure is usually a sample containing a weighable amount of inert solid material, and in such cases self-scattering and self-absorption effects make the observed activity of the sample critically sensitive to small variations in the form and dimensions of the sample.

Good reviews of techniques currently in use for handling radioactive samples have recently been published (2, 3). While attempting to minimize self-scattering and self-absorption errors, the authors tested many of the common techniques, and in no case could results consistently precise to better than 1% be obtained with samples containing more than a milligram or so of solid material. Curve A in Fig. 1 shows a typical set of counting data obtained during the course of this work.

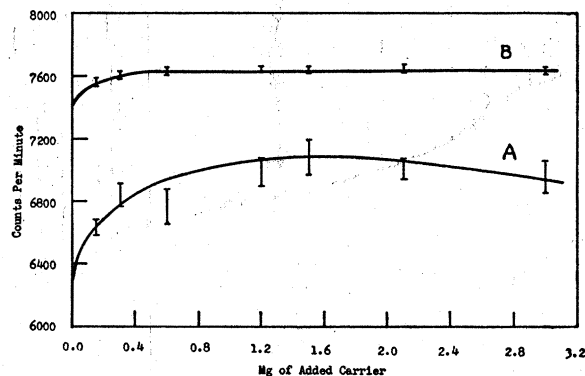


FIG. 1. Observed radiostrontium activity as a function of added inactive carrier. A, evaporated solid samples; B, lacquer-covered liquid samples.

The isotope used was Sr^{90} (25 y, $\beta=0.61$ mev) which decays to Y^{90} (65 h, $\beta=2.35$ mev). Yttrium sulfate in dilute sulfuric acid was used as carrier, and the samples were evaporated on 1-mil polystyrene film in order to minimize back-scattering effects. The samples were counted in the third shelf of the standard arrangement for use with an end-window Geiger tube. Each point in the figure represents an average of 3 counts on each

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of 3 independent samples, and the time for each measurement was sufficient to give a minimum of 10,000 counts (1% standard deviation). The lengths of the vertical lines in the figure represent approximately the observed standard deviations of the averaged values of the 3 samples taken for each point.

The initial rapid rise in counting rate as the weight of the sample increases is caused by self-scattering within the sample. The wide spread and poor precision of the values are the result of small changes in self-scattering and self-absorption which are due to sometimes minute variations in the dimensions and degree of "packing" of the solid material. The self-scattering effect reaches a maximum in this experiment at a sample weight of about 1.5 mg; above this weight, the increasing self-absorption of the sample causes the counting rate to decrease slowly. Results of this type are common, and it is clear that no very precise radiometric determinations can be made with techniques that give results similar to these.

The counting of liquid samples offers inherently better possibilities for precision counting, since self-scattering errors caused by irregularities in sample shape are absent. Various arrangements with commercially available dipping-type Geiger tubes were tried, but errors caused by instability of the tubes, and difficulties in decontamination of the tubes after each count, made this method impractical. It should be possible to do high-precision counting with dipping tubes and either stationary or flowing liquids, but such techniques await the development of better tubes and effective decontamination methods.

However, liquid counting was found to be possible with end-window Geiger tubes, and excellent precision was obtained with this method. The technique is as follows: One-ml liquid samples are placed in aluminum or copper cups 20 mm in diameter and 4 mm deep, and the liquid surfaces are covered with thin lacquer films. Most of the work was done using Zapon Clear-Lac 1233-D,³ diluted 5 times with Zapon Thinner 20, but several other commercial clear lacquers and thinners were equally useful. Eight drops of the diluted lacquer are carefully placed on the liquid surface and allowed to harden (about 30 min), and the cups are then mounted on cards and placed in the standard shelf arrangement for counting.

Curve *B* in Fig. 1 shows some results obtained with this method using Sr^{90} activity and yttrium sulfate carrier, as in curve *A*. The amount of activity used in the liquid samples was greater than that used in the solids, so that the absolute counting rates on the two curves are not to be compared. The number of samples and the counting time are the same as described for curve *A*, and the lengths of the lines again represent the experimental standard deviations of the mean values, about .3% in every case except for the carrier-free sample. The small rise in counting rate with the first addition of carrier is probably caused by the fact that the carrier-free yttrium activity adsorbs on the bottom of the

sample cup. A small amount of inert yttrium is sufficient to distribute the active yttrium through the solution, and further additions of carrier cause no change in the counting rate. Other isotopes give a small initial drop in counting rate with the first addition of carrier, because the active atoms adsorb preferentially on the surface of the lacquer film (Table 1). A comparison of

TABLE 1
EFFECT OF ADDED CARRIER ON COUNTING RATE
OF LIQUID SAMPLES

Isotope	β -energy, mev	Carrier	Conc., mg/ml	Av. cpm*	Std deviation, %
I^{131}	0.60	KI	0.00	$4,169 \pm 44$	1.1
			2.0	$2,016 \pm 10$	0.5
			100	$1,867 \pm 9$	0.5
UX	2.32	ZrOCl_2	0.00	$1,977 \pm 26$	1.3
			2.0	$1,663 \pm 12$	0.7
			70	$1,640 \pm 9$.6
$\text{Sr}^{90}\text{-Y}^{90}$	2.35	$\text{Y}(\text{NO}_3)_3$	0.00	$5,448 \pm 6$.1
			2.0	$5,525 \pm 25$.5
			20	$5,539 \pm 28$.4
			100	$5,548 \pm 16$	0.3

* Results represent averages of 3 counts on each of 3 independent samples.

curve *B* with curve *A* in Fig. 1 shows the much greater precision that can be obtained by counting a sample as a liquid rather than as a solid. The counting rate of the solid samples would be expected to decrease as the weight of the sample is increased above 10 mg because of self-absorption, but liquid samples containing as much as 100 mg of solid per ml have shown no decrease in observed activity when the β -energy is large (Table 1).

The method has been tested with a series of isotopes having β -energies ranging from 0.60 mev (I^{131}) to 3.07 mev (Pr^{144} in equilibrium with Ce^{144}). Isotopes with β -energies of about 1 mev or greater can be counted with an average precision of 0.5% or better. Lower β -energies result in slightly poorer precision, because of the greater effects of sample and film thicknesses on the counting rate. For β -energies greater than 1 mev, the counting rate is not sensitive to small variations in the densities of the solutions. In the cups used for the present work, a 1-ml sample represents an infinitely thick layer for a 1-mev particle. A sample this size fills the cup nearly to the top, however. Since it is difficult to prepare the cups with perfectly square edges on top, it is best to measure the volumes of the solution accurately in order to be sure that the surface area and counting geometry of the sample remain constant. The lacquer technique cannot be used with concentrated acids or bases, but dilute acids and bases are easily handled. If the cups react with the solution to be counted, they may be coated with lacquer before use. As already shown, carrier-free activities tend to adsorb on the bottom of the cup or on the lacquer film, and it is therefore advisable to have in the solution a few milligrams of carrier for the activity being counted. The length of time

³ Made by the Atlas Powder Company, Stamford, Conn.

a sample prepared according to this technique can be preserved depends upon the thickness of the lacquer film. Samples prepared during the course of the present work were kept up to 2 weeks before the films began to tear.

The authors made no attempt to use the liquid counting techniques for absolute β -counting. The absorption of the β -particles by the solution, and the back-scattering from the cup, tend to make the corrections that must be applied in order to find the true disintegration rate of a sample by this method very large. However, for most practical applications of radioactive tracers in chemical analysis, it is only necessary to know the ratio of the counting rate of the "unknown" sample to that of an empirical standard, and for this purpose the liquid counting technique is excellent. It is desirable to have the amounts of inactive materials in the "unknown" and standard approximately the same, but no accurate control is necessary. Because the solution absorbs part of the β -rays, the activity of a given sample counted as a liquid is in general about half that of the same sample counted as a solid, for β -energies greater than 1 mev. The relative loss in counting rate is of course greater for lower energy particles, but this is no serious handicap when the radioactive isotope can be obtained in reasonably high specific activity. The great gain in precision that can be obtained with liquid samples more than compensates for the loss in observed counting rate.

In order to obtain the maximum precision from the liquid counting method, the Geiger tube used must count reproducibly to within the limits of error desired for the experiment, and it should be stable during the period of the investigation. The authors tested a number of commercially available Geiger tubes and found that many were completely unsuited for use in precise analytical work. Some of the tubes could not reproduce the activity of a given sample to better than 5-10%. Most of these tubes showed a marked voltage hysteresis effect; that is, the counting rate for a given sample depended upon whether the operating voltage of the tube was approached from above or below, and upon the time the tube was maintained at constant voltage. Photosensitivity was observed with some tubes; this was a disturbing effect even with completely shielded tubes, because light that entered when the shield was opened to change samples caused a rise in counting rate which persisted for several minutes after the shield was closed. High humidity caused most of the tubes tested to count spuriously. This factor was controlled by keeping a good, dust-free drying agent in the counting shield at all times, and, on extremely moist days, by blowing a slow stream of dry air through the shield.

The tubes finally selected for use in the present work were models Nos. 100C and 200C, manufactured by the Amperex Electronic Corporation, Brooklyn, New York. These are end-window tubes with thin mica windows; they were used inside lead shields of standard design, which kept the background count at about 15 cpm. The tubes showed no voltage hysteresis and no photosensitivity; the humidity effect was controlled as above.

Four of these tubes showed equally good characteristics. A uniform sample counted 38 times over a 2-month interval in a 200C tube gave a standard deviation of the mean of .1%, so that the Geiger tube was not the limiting factor in determining the precision of the experimental results. This discussion illustrates the care which should be taken in selecting a tube for use in an analytical study. It should be pointed out that other counting instruments, such as methane-flow proportional counters and scintillation counters, are becoming increasingly well known. These instruments have much better counting characteristics and reproducibility than Geiger tubes and seem to offer a solution to this aspect of the problem involved in obtaining reproducible counting results.

The authors have applied the liquid counting method described in this paper in the development of a combined photometric-radiometric method for the determination of cerium (1). The over-all precision obtained in this work was about 1.4% standard deviation. By using any of the common methods for mounting solid samples, it would have been impossible to obtain better than 5-7% precision in the final results. Up until the present, radiometric methods of analysis have been severely limited in scope because of their poor precision. The use of the liquid counting technique, in conjunction with good counting equipment, should now permit the development of precise radiometric methods of analysis for many elements which are difficult to determine by other means, and should encourage the development of many other applications of radioactive tracers.

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Effects of Ultrasonic Waves and Nitrous Acid on the Production of Colloidal Sulfur

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Production of colloidal sulfur by mixing dilute solutions of sodium thiosulfate and HCl has been investigated by La Mer and co-workers (2-5). Colloidal sulfur begins to appear about 10 min after the addition of the acid and is recognized by the Tyndall scattering of light. La Mer and Yates (6) observed that the time of production of colloidal sulfur increased about 4 times when the distilled water used in this experiment is first irradiated by ultrasonic waves. They tentatively attributed it to the removal of the nuclei required for growth to colloidal dimensions, and they demonstrated that the effect