acid did. The gramicidins were active at 1:40,000 dilution and streptomycin at 1:10,000.

In vitro tests against *A. suis,* using the technic of Lamson and Brown (*5*), revealed that none of the antibiotics studied was active.

The acute toxicity of subtilin in mice, on intravenous injection of 1 per cent solution, was LD_{50} (60 ± 3 mg./kg.); on subcutaneous injection, the LD_{50} was 670 ± 30 mg./kg.; when given intragastrically, 5.0 grams/kg. killed. One per cent solution instilled into the rabbit's eye was nonirritating.

Gramicidin, 1 per cent in propylene glycol, given intravenously in mice had an LD_{50} of 1.5 mg./kg. This is slightly lower than reported by Robinson and Molitor (*9*). Gramicidin derivative was less toxic, LD_{60} being 4.7 mg./kg. Lethal doses of the gramicidins killed within one minute, which precluded the possibility of delayed hemolysis being responsible.

Summary. Subtilin, a new antibiotic obtained from *B. subtilis,* proved active *in vitro* against *L. plantarum, E. histolytica* and its associated bacterium 't', and *T. equiperdum* without causing immediate hemolysis of erythrocytes. Subtilin is tensioactive, and amounts required for antibiotic effect are within the range of surface tension activity. It was relatively nontoxic for four species of mammals, especially after intragastric administration. Gramicidin is more hemolytic and more toxic than subtilin.

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A Relation Between Size of the Divalent Cation and Solubility of Triple Acetate Salt of Sodium

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The importance of the size of the alkali metal cation to the formation of the triple acetate salt, $NaM^2UO_2(OAc)_9 \cdot 6H_2O$, was first demonstrated by Caley and Baker (*2*) when they proved that potassium, unlike lithium and sodium, formed only a double salt. In their paper, they listed the divalent ions which form triple acetate salts with sodium in an order of decreasing sensitivity toward sodium. Their list is reproduced in Table 1, together with the empirical ionic radii of Pauling (*5*) and the atomic radii of Goldschmidt (*3*). It can be seen that the solubility of the triple salt increases with the radii of the ions, whereas it bears no relation to the radii of the atoms.

TABLE 1

Radii of Divalent Cations Which Form Triple Acetate Salts Arranged in Order of Decreasing Sensitivity of Their Respective Reagents Toward Sodium

Cation	Radii in Angstrom Units		
	Ionic	Atomic	
Mg	0.65	1.62	
Ni	0.70	1.24	
Co	0.72	1.26	
Zn	0.74	1.37	
Fe	0.75	...	
Mn	0.80	1.36	
Cu	...	1.28	
Cd	0.97	1.52	
Hg	1.10	1.55	(liquid)

Caley and Baker did not assign a position to the ferrous acetate reagent because the difficulties involved in handling it made its exact position in the group uncertain. However, the value of the ionic radius of the ferrous ion establishes the position of reagent between those of zinc and manganese.[1]

TABLE 2

Radii of Alkaline Earth Metals Other Than Magnesium

Cation	Radii in Angstrom Units	
	Ionic	Atomic
Be	0.31	1.05
Ca	0.99	2.21
Sr	1.13	....
Ba	1.35	....

Likewise, if the assumption is correct that the solubility varies with the ionic radius, one should be able to assign a value to the radius of the cupric ion because the sensitivity of its reagent is known. Unfortunately, the limits are very wide, so additional information must be sought. The radius of the cuprous ion is known (0.96 A.—Pauling), as is the magnitude of the change in the radius resulting from the loss of an electron by the ferrous ion (0.15 A.). Although the conditions are not exactly the same for the ferrous ion and the cuprous ion, they are sufficiently similar to enable one to guess that the radius of the cupric ion is approximately 0.81 A.

From Table 2 one might predict that the radii of the divalent ions of the alkaline earth group do not

[1] In a private communication, Dr. Caley stated that this position is consistent with his observations.

fall into the range of those of the heavy metals and therefore are not likely to form triple acetate salts. One must admit, however, that other factors, such as the solubility of an individual acetate or the solubility of a double salt, together with the coordination number of the ion, may limit the possibility of the formation of a triple salt. It is interesting to note, therefore, that although a triple acetate has been reported for beryllium (*1*), the findings are open to question according to the work of another investigator (*4*). To date, no triple acetate has been reported for calcium, although very early work (*6*) reported two varieties of double salt, one of which might have been a triple salt. No triple salts of strontium or barium are known. Hence, it appears that the size of the ionic radius of a divalent ion not only affects the solubility of the triple acetate salt within the group listed in Table 1 but also provides a means of predicting whether or not any divalent cation is likely to form a triple acetate salt.

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The Mechanism of the Therapeutic Effect of Iodine on the Thyroid Gland

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It is now a well-established fact that in cases of toxic goiters the iodine produces an effect which, clinically, shows relief of the symptoms and, biochemically, decrease of the circulating thyroid hormone and an increase in the gland of total iodine, both free and organically bound. Histologically, this effect is manifested by the deposit of the colloid inside the follicles. These facts are generally interpreted as a blockage of the release of the secretion by iodine, but its mechanism is still not very well understood.

The theory of a mechanical blockage, supported by several authors (*5, 6*), can be hardly maintained in view of the modern concepts of enzymatic chemistry and histophysiology of the thyroid gland.

Salter and Lerman (*7*), as the result of a study of enzymatic synthesis carried out with proteases as catalysts, suggested that the therapeutic effect of iodine is due to the mass-law phenomenon, which acts by "forcing" the reaction in the direction of a synthesis and, in this way, inducing the colloid formation and storage.

In 1941 one of us (*1*) demonstrated that the colloid of rat thyroids, extracted from a single follicle, has a definite proteolytic activity, and established a correlation between this activity and the function of the thyroid gland. From these results, later confirmed by Dziemian (*3*), the conclusion was drawn that in the reabsorption of the colloid an enzymatic mechanism is involved which is responsible for the proteolysis of thyreoglobulin. It also was found that iodine, after a certain time, inhibits this proteolytic activity.

TABLE 1

PROTEOLYTIC ACTIVITY OF TOXIC GOITERS BEFORE AND AFTER TREATMENT *in Vitro* WITH IODINE

Blank Mg. of tyrosine and tryptophane set free	Iodinized extract Mg. of tyrosine and tryptophane set free	Per cent inhibition
0.116	0.031	73.3
0.164	0.035	77.1
0.094	0.042	55.3
0.225	0.105	53.4
0.092	0.011	88.5

Recently we found (*2*) in human thyreotoxicosis that the proteolytic activity of the total gland, as measured by the amount of tryptophane and tyrosine set free, is probably also decreased through the action of iodine administered in therapeutic doses. These results and those of Henrriott (*4*) on the inhibition of pepsin activity by iodization *in vitro,* led us to suppose that in the case of iodine treatment the clinical effect is due to an inhibition of the proteolytic enzyme system.

In order to test this assumption, glycerol extracts of human thyroid gland (toxic goiters) were iodinized *in vitro* with a final concentration (0.05 M) of iodine, and the proteolytic activity was determined by the amount of tyrosine and tryptophane set free after a 4-hour incubation at 37° C. with edestin as substrate. The details of this method were described in our previous paper (*2*). Here we wish only to add that the glycerol extracts, after iodinization, were dyalized for 24–48 hours at 3° C. Also, the blank (*i.e.* the same extract, but without iodine) was treated in the same way.

Certain of the results of this experiment are given in Table 1, from which the conclusion may be drawn that, under these conditions, there is 53.4 to 88.5 per cent inhibition of the proteolytic activity of the thyroid gland. It is interesting to point out that this