demonstrate that the hypophosphite preparations now on the market are woefully below standard and more care must be used in their manufacture than has been done heretofore.

INFLUENCE OF SIZE AND SHAPE OF BOTTLES UPON THE ASSAY OF PEPSIN.

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In a previous paper, entitled, "Some Observations upon the Assay of Digestive Ferments," appearing in the *Journal of Engineering and Industrial Chemistry,* (Vol. 3—No. 12—December, 1911), I gave a resume of tests applied by me and also called attention to some of the peculiarities of these delicate enzymes shown in their standardization.

Under the subject of Pepsin, I was the first to call the attention of chemists to the influence of the age of the egg upon the apparent digestive strength of a pepsin when using coagulated egg albumen as the proteid to be digested. I showed that eggs from 5 to 10 days of age gave a maximum digestive strength to a sample of pepsin. I also called attention to the important part played by the strength of acid used for digestion and showed that the pepsin dissolved in an acid solution of more or less than 0.3%, by weight, absolute hydrochloric, would not digest as much proteid as when dissolved in a menstruum of exactly this official strength (0.3%).

Another factor, overlooked by chemists in the past and very important from the standpoint of accuracy and uniformity of results in standardization by various chemists, is the size and shape of the bottle used in the digestion experiments. The United States Pharmacopœia states, in reference to the digestion bottle used with pepsin standardization, to digest in a wide-mouth bottle of 100 cc. capacity; but the question arises, do you get the same relative amount of digestion in a short round bottle of 100 cc. capacity that you do in a taller square bottle of the same volume? From the results of my experiments, my answer is "No"; different bottles give different results. This is illustrated by the following experiments:

Pepsin Sample No.	Style of Bottle.	Strength Tested for	Residue
Standard 1:3000	(No. 1) 6 oz. French square wide mouth, capacity 175 cubic centimeters. 5¼″ Tall.	1:3000	1 cc.
1:3000	(No. 2) 4 oz. French square wide mouth, capacity 120 cc. 4¾″ Tall.	1:3000	1 cc.
1:3000	(No. 3) 3 oz. French square wide mouth, capacity 90 cc. 4½″ Tall.	1:3000	1 cc.
1:3000	(No. 4) 3 oz. Round, Prescription wide mouth, 100 cc. capacity. 4″ Tall.	1:3000	1¼ cc.
1:3000	4″ Tall.	1:2750	1 cc.

In the above experiments the eggs were but five days old and were too fresh,

as shown by the residue remaining, 1 cubic centimeter. The experiment was repeated with eggs eight days old, with the following results:

Pepsin. Standard	Style of Bottle.	Strength Tested.	Residue
1:3000	No. 1	1:3000	0.5 cc.
1:3000	No. 2	1:3000	0.45 cc.
1:3000	No. 3	1:3000	0.5 cc.
1:3000	No. 4	1:3000	0.8 cc.
		1:2750	0.5 cc.

This was shown again in another test on eggs eight days old, as follows:

Pepsin. Standard	Style of Bottle.	Strength Tested.	Residue
1:3000	No. 1	1:3000	0.5 cc.
1:3000	No. 2	1:3000	0.5 cc.
1:3000	No. 3	1:3000	0.5 cc.
1:3000	No. 4	1:3000	0.8 cc.
		1:2750	0.5 cc.

Conclusions.—The experiments cited show that the style of bottle exerts a big influence upon the amount of proteid digested. Bottles No. 1, 2 and 3 are of the same style, i. e., wide mouth, French square of 6 oz., 4 oz. and 3 oz. capacity, and the results show that the digestion in these bottles seems to be the same in spite of the difference in relative capacity. With bottle No. 4, however, a strength of 250 units less is shown, and this bottle is a short, round, wide-mouth prescription bottle of 3 oz. capacity. My explanation is that in this style of bottle the contents do not receive the same agitation, due to the bottle being shorter in length and larger in diameter, even though its relative internal capacity is more than bottle No. 3; thus proving that the digestion conducted in a square long bottle leaves less residue than a short round bottle of larger internal volume.

Another point that I wish to bring up is that in the use of the 6 oz. French square, it is possible to add first the 10 grams of egg albumen, then all the acid, and finally the requisite amount of pepsin solution, and after securely inserting the stopper, to pound it upon a pad and completely disintegrate the albumen. With all the other sizes of bottles the directions of the Pharmacopœia to disintegrate the albumen first with a small quantity of the acid and a rubber-tipped glass rod and gradually wash rod with balance of acid, must be followed. As this might lead to an error, I recommend the use of the larger bottle and do not favor the introduction of any foreign substance at all, and the results show that the relative digestion is not changed by the size of this style of bottle.

In concluding, I wish to state that a personal equation as to agitation could not enter in my experiments, since the bottles were all tightly clamped into a revolving drum, which was immersed in a water bath, and when the drum was rotated, each and every bottle received the same amount of agitation and at the same time.