

and Chase have observed that adsorption of T_2 to bacterial membranes results in liberation of the phage DNA in a nonsedimentable form (5). In the present studies, emphasis is placed on the release of nonsedimentable nitrogen from the host-cell membranes, but release of nonsedimentable virus nitrogen is also shown. Representative experimental values are given in Table 1.

Electron micrographs taken at zero time and after incubation showed that adsorption of T_2 was accompanied by disintegration of the virus and the conversion of the membrane to a granular residue.

The chemical nature of the substances "solubilized" from the membranes and the application of the foregoing findings to studies with intact host cells are presently being investigated.

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The Effect of Pasteurization on the Stability of Phosphates Can Be Used as a Test for Heated Milk

Socrates A. Kaloyereas

Department of Agricultural Chemistry and Biochemistry, Louisiana State University, Baton Rouge

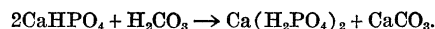
It has been known for a long time that heating of milk results in a lowering of its pH (1). However, neither the explanation given for its cause by Whittier and Benton (2), who attribute it to the lactic acid formed from lactose, nor the earlier idea expressed by Orla-jensen and Plattner (3), attributing it to the formation of lactic acid from casein, seems justified in light of the results reported here. It was found that heating of milk at $62^\circ C$ for 30 min, in order to pasteurize it, results in a decrease of its pH , whereas more prolonged heating does not have any further effect on the pH (Table 1).

These results indicate that a stable equilibrium is reached during the early period of heating and, as will be explained, the reaction that takes place stops when one of the reactants, namely, the carbonic acid, is unavailable. Titration curves of the rennet serums from raw and pasteurized milk show two characteristic differences at the points ending the titration of monocalcium phosphate and dicalcium phosphate. The dB/dpH values of milk serum changed by pasteurization from 0.0146 to 0.0159 at the range of the titration of monocalcium phosphate and from 0.0059 to 0.0052 at the range of the titration of dicalcium phosphate. These differences indicate an increase of monocalcium phosphate and a more or less proportionate

Table 1. Effect of heating on the pH of milk.

Sample	Raw (pH)	Pasteurized at $62^\circ C$	
		For 30 min (pH)	For 60 min (pH)
1	6.75	6.71	6.71
2	6.77	6.69	6.69
3	6.70	6.62	6.62
4	6.73	6.65	6.65
5	6.68	6.63	6.64
6	6.81	6.78	6.78
7	6.78	6.73	6.73
8	6.63	6.57	6.57
9	6.65	6.60	6.61
10	6.64	6.58	6.58

decrease of dicalcium phosphate in pasteurized milk serum. The new equilibrium established between monocalcium phosphate and dicalcium phosphate in the heated milk serums is a result of a reaction that, most probably, takes place between dicalcium phosphate and carbonic acid as follows:



Serum from heated milk exhibits a characteristic increase in stability, especially at pH 4.6, when heated at $70^\circ C$ as compared with the serum of raw milk. This increased stability is shown not only by the increased heating time required to bring about the precipitation of phosphates but also by the smaller amount of the precipitate formed. Furthermore, the increased turbidity found in the supernatant liquid of the precipitated heated-milk serum indicates the greater stability of the phosphates there and provides further evidence of the reaction that takes place during pasteurization. It was found that the greater cause of turbidity of the supernatant liquid is the presence of carbonates and that by eliminating them—according to the Curtman and Hart method (4)—the tur-

Table 2. Average values for moist precipitates turbidity of supernatant liquids and stability (time needed for flocculation) of serum at pH 4.6 when heated at $70^\circ C$ in a constant-temperature water bath.

Milk serum	Moist pre-precipitate	Stability (min)	Turbidity optical density	Decrease of turbidity after elimination of carbonates (%)
Raw	2.6	15–20	0.300	84
Pasteurized	1.0	20–25	.900	60
Powdered milk				
Pet	0.4	More than 30	.290	
Starlac	.1	No flocculation	.400	

bidity assumes very low values in both heated- and raw-milk serums (Table 2).

Additional evidence of the validity of this theory of the cause of the decrease in the pH of milk by pasteurization was obtained from experiments with artificial mixtures of monocalcium and dicalcium phosphates. A mixture of these two salts, at about the same concentration in which they occur in milk (5), was prepared, and its pH was adjusted to a value approaching that of milk by adding small amounts of carbonate solution. When this mixture was heated at the pasteurization temperature of milk, its pH decreased, and generally the mixture behaved like milk in this respect. These facts indicate definitely that the decrease in the pH of milk effected by the pasteurization treatment is due to the new equilibrium established between monocalcium phosphate and dicalcium phosphate through a reaction that takes place between dicalcium phosphate and carbonic acid.

The work of Muller and Knöffel (6) has shown that carbonic acid at higher temperatures reacts to a certain extent with secondary and tertiary calcium phosphates with the production of primary calcium phos-

phate. This can also be deduced from the result obtained by Windish and Dietrich quoted by Mellor (7).

We have found that the lowering of the pH of milk effected by heat treatment and the resulting increased stability of the phosphates of the serum can be used as a test for differentiating heated milk from raw milk in their mixtures. Details of this work, as well as other possibilities now being explored, for using the change of the stability of phosphates in other biological fluids, will be reported later elsewhere.

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Communications

Effect of X-irradiation on the Adrenal Cortical Steroid Excretion in Urine

Abundant indirect evidence (1-4) supports the prevalent idea that adrenal cortical activity is altered following the stress of x-irradiation. The following preliminary results of the determination of the adrenal cortical steroids in the urine of pigs after lethal x-irradiation give more direct evidence (5).

Two castrated, male Chester-White pigs each weighing approximately 22 lb were given 1000 and 750 r, respectively, of total body x-irradiation. The factors were 1000 kv, 3.0 ma, Pb parabolic, 35-in. target-skin distance, and output 8.9 and 9.25 r/min, respectively.

Table 1. Total neutral adrenal cortical steroids in urine of pigs given lethal total body x-irradiation.

Day	Pig No. 1, 1000 r, mg/24 hr	Pig No. 2, 750 r, mg/24 hr
- 2		1.1
- 1	0.9	0.7
0	.9	.5
1 +*	1.7	2.4
2 +	1.4	0.7
3 +	0.4	1.1
4 +	.3	1.8
5 +	Animal expired	0.8
6 +		1.0
7 +		Animal expired

* Irradiated on this day; 24-hr urine collection was begun immediately following irradiation.

Control and post-irradiation 24-hr urine collections were obtained. The urine was stored at 5°C without preservatives. The total neutral C₂₁ adrenal steroids were determined by the method of Burton, Keutmann, and Waterhouse (6) with the modification that they were quantitated by the method of Mader and Buck (7).

Table 1 shows the marked increase in the urinary excretion of adrenal cortical steroids in these animals after lethal total body x-irradiation. It is to be noted that the increase was most marked in the first 24-hr period following exposure of the animals to the radiation.

The data are conclusive only in that they supply direct evidence of acutely altered adrenal cortical activity following lethal total body x-irradiation.

F. T. BRAYER

S. R. GLASSER

B. J. DUFFY, JR.

Medical Division, Department of Radiation Biology,
University of Rochester School of Medicine and
Dentistry, Rochester, New York

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