the bodies of bacteria and other organisms in the feces. The evidence further indicates that neither thiamine nor cocarboxylase are absorbed when physiological amounts are administered in a retention enema.

TABLE 1
FECAL THIAMINE AND COCARBOXYLASE

Stool weight gm	Thiamine content		Cocarboxylase* content	
	Per gm stool mcg	Total mcg	Per gm stool mcg	Total mcg
100	0.81	81	5.63	563
231	0.87	200	4.37	1,010
125	0.97	121	5.22	840
74	0.81	60	11.70	870
295	0.75	220	4.10	1,210
174	0.63	110	5.00	870
83	0.28	23	6.10	504
108	1.12	121	5.18	559
108†	0.53	57	...	...

* Expressed in terms of thiamine.
† An aliquot of the preceding stool; this aliquot was suspended in water and passed through a Seitz filter before analysis.

The analytical values for thiamine and cocarboxylase, obtained by chemical analysis,[6] in the human feces from a normal subject whose daily dietary intake consisted of 2,400 calories show that the feces contain more of these substances than any tissue of the body. The concentration of each substance is remarkably constant, and the ratio of free to phosphorylated thiamine is of the same order as that of tissues. Removal of organisms from a water suspension of feces by Seitz filtration results in a marked decrease in the thiamine present. This indicates that the thiamine exists largely in the bodies of fecal organisms.

TABLE 2
THE EFFECT OF A RETENTON ENEMA CONTAINING THIAMINE AND COCARBOXYLASE ON THEIR EXCRETION IN URINE AND FECES

	Fecal thiamine and cocarboxylase						
	24-hour urinary thiamine mcg	24-hour free thiamine mcg	Expected* free thiamine mcg	Recovery of expected free thiamine per cent.	Cocarboxylase† mcg	Expected* cocarboxylase mcg	Recovery of expected cocarboxylase per cent.
Before enema	116	121	...	...	559	...	...
	108	...	...	...	...	...	...
After enema	107	490	418	117	3,050	2,690	118

* Calculated on basis of stool weight times average concentration of thiamine and cocarboxylase in stools of same subject (Table 1); to this is added the amount of thiamine (0.25 mgm) and cocarboxylase (2.0 mgm) in the retention enema.
† Expressed in terms of thiamine.

Najjar and Holt[3] administered 50 mgm of thiamine rectally to two subjects on successive days. They observed rises in urinary thiamine excretion and from that concluded that the large intestine could absorb thiamine. It must be pointed out, however, that 50 mgm of thiamine is a huge dose compared with the amounts of thiamine normally present in human feces. Furthermore, most of the thiamine in feces is in the form of cocarboxylase, whereas they administered only thiamine. Cocarboxylase can not be absorbed as such and must first be split by dephosphorylating enzymes. It is dubious whether such enzymes exist in the large intestine.

Our experiment was devised to be more physiological. A retention enema, containing twice the amount of free and phosphorylated thiamine in the average 24-hour stool, was administered and retained for 24 hours. Not only was there no increase in the urinary thiamine excretion, but all the administered thiamine and cocarboxylase were recovered as such from the next 24-hour stool.

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DEAMINATION OF AMINO ACIDS BY THE HUMAN ORAL FLORA; ITS ROLE IN DENTAL CARIES IMMUNITY*

IT has been demonstrated in numerous studies that certain aciduric bacteria, particularly *L. acidophilus*, are missing from the mouths of patients who have no active caries.[1, 2, 3] It has also been shown that the saliva of these individuals does not rapidly convert glucose into acid.[4, 5] Viable *L. acidophilus* inoculated into the saliva or oral cavities of such individuals have disappeared from cultures made six to eight hours later.[6] The cause for the elimination of these bacteria and the inhibition of fermentation has been sought.

We found that human saliva inoculated into beef broth medium incubated for eight days and then filtered through a Seitz filter contained something that was bactericidal to *L. acidophilus*. When a small quantity of this filtrate was added to a saliva which would by itself degrade glucose rapidly, no acid was formed.

Experiments proved this inhibiting factor to be ammonia nitrogen. When a solution of an ammonium salt having a pH of 6.8 or above was added to saliva in the same concentrations of ammonia nitrogen as produced in salivary cultures, similar effects were obtained. Amounts as low as 0.5 mgm of ammonia nitrogen per ml were found efficient in inhibiting *L. acidophilus* growth and in preventing fermentation of

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glucose. Solutions of sodium and potassium salts having the same pH were non-inhibiting.

Grove and Grove[7, 8] called attention to the importance of ammonia in saliva several years ago, but the significance of their observations has not been grasped, perhaps because large quantities of ammonia nitrogen were not detected in the saliva of caries immunes. Small concentrations of ammonia formed more or less continuously on the vulnerable surfaces of the teeth may confer a natural protection against dental caries. The above authors as well as Stephan[9] have reported lowering caries incidence in many cases by using ammonia and urea mouth rinses.

The question arises as to what is present in the salivas of caries-free individuals that provides the ammonia. Early studies by us indicate that the source is not alone urea or mucin but the presence of various amino acids. Twenty-three amino acids alone and in various combinations have been checked for the possibility of ammonia production from them by the enzyme systems of the bacteria previously found in the salivary cultures.

Preliminary experiments have revealed that caries-immune individuals have enzyme systems capable of converting at least six amino acids into ammonia nitrogen. The six are arginin, alanine, aspartic acid, asparagin, glutamic acid, isoleucine and serine. Certain combinations of these amino acids proved much more effective. A wide variance of enzyme systems have been found in salivas from persons whose teeth are actively decaying. However, there seems to be in most instances an absence in the saliva of caries-active patients of a system that will convert glutamic acid to ammonia.

Microbiologic assays are now underway to determine the amino acid content of caries-immune and caries-active salivas. Our study to date suggests that caries immunity is based on the production of minute but continuous amounts of ammonia in the bacterial plaque resident on the tooth surface. The pabulum from which the ammonia is derived is apparently a small group of amino acids. These are present in the mouth as a result of the type of diet and body metabolism.

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THE PREPARATION OF HIGHLY PURIFIED PR8 INFLUENZA VIRUS FROM INFECTED MOUSE LUNGS[1]

ONE of the most convincing lines of evidence in establishing that tobacco mosaic virus activity is a specific property of a characteristic high molecular weight nucleoprotein was the demonstration that preparations of the virus protein from different species of plants, some of which were widely removed from the tobacco family, possessed essentially the same chemical, physical and biological properties.[2] This approach has not heretofore been used for viruses affecting animals because essentially pure preparations of virus from different animal hosts have not been available. Recently, highly purified preparations of PR8 influenza virus have been obtained from the allantoic fluid of infected chick embryos.[3,4] These preparations contain particles about 100 mμ in diameter possessing characteristic chemical, physical and biological properties.[3-9] It appeared, therefore, that if one could isolate a comparable product from another host, an approach similar to that used for tobacco mosaic virus could be made and thus provide important data on the nature of influenza virus produced in different hosts. The possibilities of comparison were enhanced by the fact that influenza virus appears to possess at least two major forms of biological activity, namely, virus activity and red cell agglutinating capacity.

An attempt was made therefore to obtain purified preparations of PR8 influenza virus from suspensions of infected mouse lungs. When methods of centrifugation alone were employed, such as had been used previously by other workers,[10,11] it was discovered that preparations were obtained which had low chick-cell agglutinating (CCA) activities and which obvi-

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