strain S, is specific as is the original filtrate. But with the second, obtained from the resistant strain R, Doctor Wollstein of the Rockefeller Institute has found a marked action on Shiga, on Flexner and on Hiss dysentery bacilli. In consequence of this observation, we have been able, by a method of successive passages through appropriate strains, to extend the lytic power to other species, as typhoid and paratyphoid bacilli, and have obtained by this somewhat different technique results similar to those recently published by Bordet and Ciuca.[1]

4. We have observed also that the modified *coli* of Bordet and Ciuca, *e.g.*, the *coli* which has resisted the lytic action, contains two types of organisms: a mucoid and fluorescent type, *coli* M 1, and a non-mucoid and translucent type, *coli* M 2. Both types, once isolated, keep their individuality after many passages in artificial media, but if the non-mucoid *coli* M 2 is submitted again to the lytic agent, we find amongst the organisms which resist a certain number of mucoid bacilli.

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A dietetic production of rickets in rats and its prevention by an inorganic salt.

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The occurrence of rickets in white rats maintained under laboratory conditions has been well known to pathologists since the first publication of Morpurgo in 1900; and the essential identity of the lesions with those of human rickets has been established by the work of Morpurgo himself, of Schmorl, of Weichselbaum, and especially by the detailed histological studies of Erdheim. One of us (A. M. P.) also has had opportunity to become familiar with the disease in rats, in the course of an investigation of the possible influence of the thymus upon the teeth and skeletal system. In none of these investigations, however, were the dietary conditions of the rats standardized and controlled.

[1] *C. R. Soc. belge Biol.*, T. 1921, lxxxiv, 278.

In continuing by means of feeding experiments upon rats the study of the mineral elements in nutrition which has engaged much of the attention of one of us for the past fifteen years, we have found a relatively simple diet which has led to the development of rickets in every one of the cases thus far examined; while complete protection was afforded by the addition of 0.4 per cent. of potassium phosphate (K_2HPO_4) to this diet, or (more strictly) by the introduction of this mount of potassium phosphate in place of a part of the calcium lactate which the rickets-producing diet contained.

In our experience healthy rats of families on normal diet when separated from their mothers at 28 to 30 days of age average about 40 grams in weight and a calcium content of about 0.3 gram or 0.7 per cent. If then placed upon good diet the calcium content of the body increases in greater ratio than the body weight so that the percentage of calcium in the body rises continuously until at about four to six months of age the adult percentage of calcium—about 1 to 1.25 per cent. of the body weight—is reached, after which the weight of the body and the weight of calcium which it contains continues parallel until growth is complete.

If, however, the healthy young rat after weaning is placed at the age of 28 to 30 days upon a diet consisting of patent flour and sodium chloride, growth in body weight is practically suspended and simultaneously the body practically ceases to increase its calcium content. In such case the rat usually lives about six weeks upon the experimental diet with hardly any change either in body weight or total body calcium. Such rats showed multiple fractures, marked deformity of the thorax, and osteoporosis.

In parallel experiments in which calcium lactate, with or without ferric citrate, was added to the diet of patent flour and sodium chloride (Diets 83 and 84), there was little or no growth in body weight but the body gained sufficient calcium so that its percentage of calcium after 4 to 6 weeks on this diet was about the same as in a normal rat of the same age, though the absolute amount of calcium contained in the body was much below that of a normal rat of the same age, as would be expected in view of the suspension of growth. These rats uniformly showed rickets.

When with all other conditions the same, the diet was modified

by the introduction of 0.4 per cent. of potassium phosphate, (Diet 85) rickets was prevented. Here again growth in body weight was practically suspended, but the addition of potassium phosphate to the diet resulted in an increased assimilation of calcium so that together with absence of bone lesions there was found a percentage of calcium in the body somewhat higher than the normal average for the age, though naturally in view of the marked stunting of the animals the absolute amount of calcium in the body was less than in a normal rate of the same age (which would have had at least twice the body weight).

Our pathological findings are summarized in Table I.

TABLE I.

SUMMARY OF FINDINGS AS REGARDS RICKETS.

Diet No.	Composition of Diet.	Total Number of Rats Examined.	
		A. Rachitic.	*B.* Non-rachitic.
83	Patent flour.....95% Calcium lactate..... 3% Sodium chloride..... 2%	2	0
84	Patent flour.....95% Calcium lactate..... 2.9% Sodium chloride..... 2.0% Ferric citrate..... 0.1%	13	0
85	Patent flour.....95% Calcium lactate..... 2.5% Sodium chloride..... 2.0% *Potassium phosphate*..... 0.4% Ferric citrate..... 0.1%	0	15[1]

In our observations as summarized in Table I, the diagnosis of rickets has been based upon the microscopic examination of ribs and femora, partially decalcified in Mueller's fluid; and in several instances, confirmed by X-ray examination. The histological criteria are: (1) the great increase in width of the zone of growing cartilage, and its irregular projection towards the diaphysis; (2) the failure of calcium deposition in the zone of preparatory calcification; (3) a pronounced increase in the osteoid margin, both in the region of the metaphysis, and along the shafts of the bones.

[1] 5 of these placed on special diet at age of 60 days; 1 at 81 days; all remaining animals started at four weeks of age.

It is seen from the Table that rachitic lesions developed in 15 rats maintained on Diets No. 83 or No. 84 for a period varying from one to two months after weaning. The rats surviving the longer period, naturally, showed more extreme changes, but the lesions were easily recognizable in the X-ray and in the gross changes at the chondro-costal junctions, after four weeks. Nine rats maintained for similar periods on Diet No. 85, which differs from Diet 84 only in the substitution of 0.4 per cent. K_2HPO_4 for an equivalent amount of calcium lactate, showed entirely normal bones, both grossly and microscopically; and six other rats, five of which were changed to Diet 85 at the age of 60 days and one at 81 days, also failed to develop rickets.

While our work was in progress there have appeared the first two of a series of papers by McCollum and his associates dealing with the experimental production of rickets in rats by means of a wide variety of deficient diets. In the first of these papers McCollum and his co-workers state (p. 340) that they "are not willing to hazard any statements in regard to the factors operating to produce rickets in the child or the experimental animal"; while in the second paper, published simultaneously, the same authors say (p. 344) that by the use of faulty diets "especially certain diets deficient in the socalled fat-soluble A or in both that substance and calcium, the cartilage and adjacent portions of the metaphysis of the long bones of the extremities could be rendered entirely free from calcium deposits and a condition identical with the rickets of human beings be obtained."

In view of the emphasis thus given by these investigators, as well as by others, to deficiency of calcium or of fat-soluble vitamine or both as causing rickets, it has seemed to us that we might aid in the attack upon this puzzling disease by placing on record at this time our observations upon the prevention of rickets when caused by the particular experimental diet here described, by the simple introduction of a single inorganic salt, and that not a salt of calcium, into the diet.

Without entering into detailed discussion of these results at this time we desire to emphasize the following facts in order to avoid misunderstanding. On the particular diet here used rickets uniformly appeared in the absence, and was uniformly prevented

by the presence, of the added potassium phosphate; but this does not imply that in these cases the cause of rickets was necessarily a deficiency either of potassium or of phosphorus. The quantitative relations of the inorganic ions rather than an absolute deficiency of any one of them, may have been the determining factor. Also it may well be that under certain dietary conditions, rickets may be caused by deficiencies or unbalanced quantitative relationships of organic as well as inorganic food factors. It would appear however to have been demonstrated that rickets may be caused or prevented without change in either the protein or vitamin components of the diet and therefore that neither of these can be regarded as a necessarily predominating factor. It is also of outstanding interest that the rats showing multiple fractures and marked deformity of thorax, which would probably be inincluded under the classification used by some writers as "presenting the gross picture of rickets" but whose bone lesions on microscopic examination were classified as those of osteoporosis, were those which had been subjected to even greater dietary deficiency than those showing typical rickets.

We are indebted to Dr. J. M. Steiner for his coöperation in the X-ray examinations and to Miss F. L. MacLeod for the quantitative determinations of calcium.

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Growth accessory substances in the nutrition of bacteria.

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In studying the growth characteristics of mucoid bacilli Thjotta observed that *B. influenzæ* will grow in blood-free broth containing the mucoid material from cultures of Friedlander's bacillus and other closely allied organisms. The growth accessory substance or substances which can replace blood and blood derivatives in the cultivation of Pfeiffer's bacillus Thjotta found in both the saline suspensions and watery extracts of the heat killed bacillary material. Furthermore, these bacterial emulsions and extracts can be boiled for ten minutes and filtered through Berkefeld