INFLUENCE OF CLAY ON SOIL PROCESSES

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Experiments were carried out, using filters or columns of sand fraction (40–70 mesh) and mixtures of the sand fraction and clay (bentonite, powdered and passed through 120-mesh sieve) treated with different forms of organic matter and inorganic fertilisers with a view to (*a*) making a broad analysis of the physical conditions, nitrogen and phosphorus status, the micro-organisms in these systems, and (*b*) obtaining an idea of the continuous metabolic changes over a period of 38 days as reflected in the effluents from the different filters.

The results showed that in the presence of bentonite clay (*i*) more moisture was retained; (*ii*) water stable aggregates of 2 mm size were formed; (*iii*) nitrification set in earlier and more nitrite and nitrate were formed; (*iv*) less nitrogen (total N) and more phosphorus were retained; (*v*) retention of water-soluble phosphorus by the sand was prevented to the extent of about 50 per cent; (*vi*) the numbers of bacteria, actinomycetes, and fungi were more and yeasts and algae were not found; (*vii*) the ciliate protozoan *Colpidium* sp. developed and the maximum numbers of this protozoan were found in the filter treated with straw, which had the highest percentage (90) of water stable aggregates of 2 mm size; and (*viii*) where nutrients were not a limiting factor, clay seemed to provide more favourable conditions for plant growth.

INTRODUCTION

The purpose of the present paper is briefly to describe and discuss the results obtained by a new approach to the study of soil processes on the basis of the evidence accumulating in this laboratory for over 50 years (Pillai *et al.* 1971) on the principles of sewage purification. The new aspect of this approach relates to the use of columns of sand fraction with and without bentonite clay and different forms of organic matter and inorganic fertilisers, and distilled water, raw sewage and refluxed sewage as irrigants, and to the examination of the effluents and filter materials for the various changes.

MATERIALS

In the experiments described here, 30 cm columns of sand (passed through 40-mesh sieve and retained on 70-mesh) and mixtures of the sand fraction and bentonite clay were used. The sand used was originally obtained from a river bed (the Arkavati river bed, about 16 km away from Bangalore).

The same sand fraction (40-70 mesh) was used in the second series of experiments but was mixed with bentonite clay. The bentonite clay was obtained through the courtesy of Dr. D. Gopal, Deputy Director, Board of Mineral Development and Consulting Geologist to the Government of Mysore. The bentonite was whitish grey in colour and was powdered and passed through 120-mesh sieve.

METHODS

The sand fraction and mixtures of the sand fraction and clay were taken in glass tubes of 45 cm length and 3.75 cm of inner diameter (similar to the chromatographic columns) (Fig. 1). At the base of each tube a layer of glass wool was placed in order to control the passage of the filter material. There were a set of 10 filters in each of the experiments, i.e. with the sand fraction as well as with the mixture of the sand fraction and clay. The weight of the sand fraction in each filter was 450 g and the weight of the sand fraction and clay in each filter was 461.3 g. The sand fraction was mixed with bentonite clay at 2.5 per cent level on the basis of the experiments with different amounts of bentonite clay and the reasonable rate of filtration observed with water and sewage (Table I). In this connection it may also be stated that Madhok (1937) employed 2.5 per cent bentonite with sand as "synthetic soil" for his experiments, and that Martin and Waksman (1940) and Conn and Conn (1941) also employed 2.5 per cent bentonite clay with sand in their experiments.

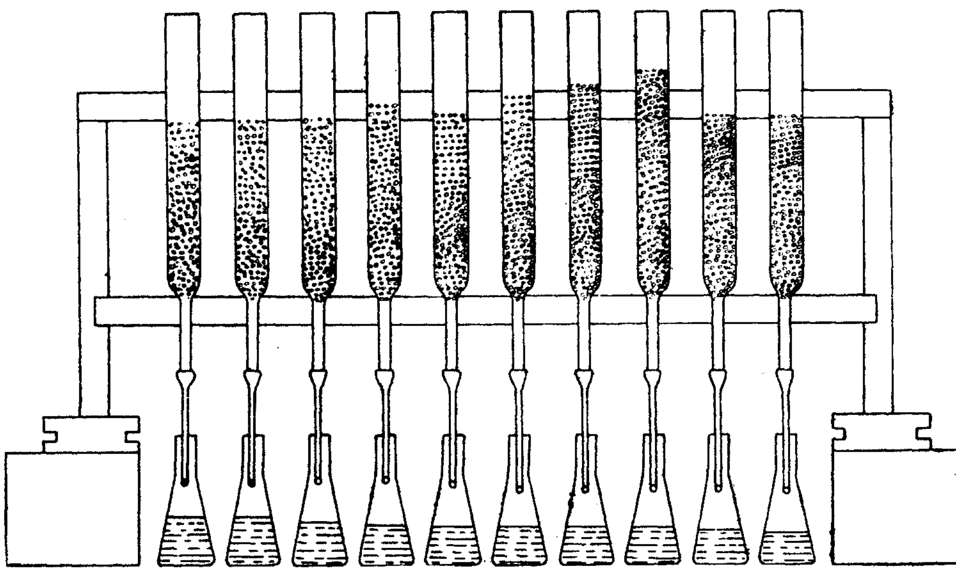

FIG. 1. Sketch of the experimental filters set up in the laboratory.

These two sets of filters received the following treatments : (1) distilled water only (control); (2) raw sewage (as obtained from the sewage works at this Institute); (3) refluxed sewage which had been refluxed by keeping it on a hot plate for 15 min and then cooling it under a tap; (4) sewage solids, prepared by evaporating samples of sewage on a water bath and drying the solids later in an oven (103°C) for 1 hr; (5) activated sludge solids obtained by drying the sludge originally brought from the activated sludge tank at this Institute; (6) dried cowdung; (7) dried farmyard manure; (8) rice straw ; (9) defatted groundnut cake; and (10) ammonium sulphate and superphosphate. The materials (4) to (10) had been powdered and passed through 120-mesh sieve.

TABLE I

Rate of filtration of water and sewage in filters containing sand fraction (40–70 mesh size) treated with different amounts of bentonite clay

Filter	Time (sec) taken for filtration of 250 ml	
	Water	Sewage
Sand fraction only	30	60
Sand fraction with bentonite :		
1.0 per cent	30	60
2.5 per cent	45	135
5.0 per cent	125	255

Note: The filters containing sand fraction mixed with 10, 15 and 20 per cent bentonite did not filter.

TABLE II

The amounts and nitrogen contents of different forms of organic matter and inorganic fertilisers added to the different experimental filters

Filter	Organic matter or fertiliser	Nitrogen content (mg)
1. Control (Sand or sand + clay without any organic matter or fertiliser)		
2. Raw sewage	9.5 l	442
3. Refluxed sewage	9.5 l	310
4. Sewage solids	15 g	450
5. Activated sludge	7 g	455
6. Cowdung	27 g	405
7. Farmyard manure	50 g	400
8. Straw powder*	62 g	372
9. Groundnut cake	5 g	400
10. Ammonium sulphate+ superphosphate	2.1 g + 0.6 g	431

*After the first week, the filter containing sand and straw powder was clogged and did not function, hence in the second series with sand and clay, only 30 g straw powder was added and this filter functioned throughout the experimental period of 38 days.

The amount of each of these materials applied was calculated on the basis of the estimate of the nitrogen in the raw sewage (liquid) used for irrigation (but this is not applicable to the refluxed sewage, as during refluxing every time nearly 30 per cent of the ammoniacal nitrogen was lost). The amounts of nitrogen in the different dry materials, which were prepared and added to the different filters, are given in

Table II. The amount of superphosphate added along with ammonium sulphate (filter No. 10) was based on the approximate amount of water-soluble phosphorus in the raw sewage used.

Each of the filter Nos. 1, 4, 5, 6, 7, 8, 9 and 10, in both the series of the experiments, received daily 250 ml distilled water, filter No. 2 received 250 ml raw sewage and filter No. 3 received 250 ml refluxed sewage. The filters were thus irrigated continuously for 38 days, but during the first 21 days, the water or sewage added daily was recycled 7 times, at the end of every half an hour, during the day. This recycling of the effluent was done in order to accelerate the oxidative changes in the filter or to "ripen" the filters.

Analysis of the effluents

After a period of 21 days, the recycling was stopped and thereafter daily only once water or sewage was used for the filtration. Throughout the experimental period of 38 days, every day the final effluents were analysed for turbidity, total solids, acidity, alkalinity, oxygen absorbed from acidified potassium permanganate in 4 hr (4–hr permanganate value), biochemical oxygen demand (BOD), ammoniacal, nitrite and nitrate nitrogen and water soluble phosphorus, using the methods described in Methods of Chemical Analysis as Applied to Sewage and Sewage Effluents (1956), Standard Methods for the Examination of Water, Sewage and Industrial Wastes (1960), and Manual of Methods for the Examination of water, Sewage and Industrial Wastes (1963). But only the results of analysis of ammoniacal nitrogen and water soluble phosphorus are given in this paper.

Examination of the filter materials

At the end of the experimental period (38 days), in each series, the filters were allowed to rest for 48 hr and then the materials from the filters were carefully taken out and spread on separate sheets of clean paper kept on the laboratory bench. The air dried materials (dried over a period of 48 hours) were examined for (i) moisture contents; (ii) water stable aggregates; (iii) amounts of total nitrogen and phosphorus; and (iv) total counts of bacteria, yeasts, actinomycetes, fungi, algae and protozoa.

Water stable aggregates were determined by the method developed in this laboratory (Kasi Viswanath and Pillai 1968*a*, 1972*a*, 1972*b*, 1973). The media used for culturing and counting the different micro-organisms by the dilution method were as follows : nutrient agar and soil extract agar (Lochhead and Chase 1943) for bacteria, yeast extract agar for yeasts, starch agar (Kuster and Williams 1964) for actinomycetes, Rose Bengal agar (Agnihothrudu 1957) for fungi. Algae were grown in a mixture of salts : sodium nitrate, 0.5 g; potassium dihydrogen phosphate, 0.5 g; magnesium sulphate, 0.5 g; calcium chloride, 0.5 g; ferric citrate, 0.005 g in 1000 ml distilled water.

Autoclaved sewage as a medium for soil protozoa

Raw sewage, when freely exposed and more especially when it is artificially aerated, supports a variety of protozoa which are derived from the soil. This observation has been utilized for the preparation of a liquid medium by autoclaving settled domestic sewage and using it for the cultivation of different ciliates, including Vorticellid protozoa (Pillai and Subrahmanyan 1945–46; Pillai and Mohan Rao 1952; Pillai 1955; Srinath *et al.* 1962; and Kasi Viswanath and Pillai 1968*b*). Autoclaved sewage does not seem to have been used elsewhere for cultivating protozoa.

One gram each of the filter materials was added to 300 ml autoclaved sewage taken in 500 ml conical flasks. Air was bubbled through the suspensions for 72 hr. After aeration, the suspension from each flask was taken in a measuring cylinder and was allowed to settle for 30 min. Samples of the sediment or deposit were examined under the microscope and the numbers of different protozoa were counted.

Plant growth

Three hundred grams each of the different filter materials were taken in beakers from both the sand and sand-clay series, and seeds of *Sorghum* (CSH : 1, a high yielding variety) were sown. The beakers were kept in the glass house at this Institute. The contents of the beakers were moistened with distilled water (once in two days) to the extent of one-third moisture-holding capacity of the filter materials. The seedlings in each beaker were thinned down to three and were grown for 38 days. After this period, tops or shoots of these plants were cut and dried in an oven (103°C) overnight. The dry weights of the shoots were recorded.

RESULTS

Nitrogen and phosphorus in the effluents

Ammoniacal nitrogen—The results given in Table III show that the filters irrigated with raw sewage and refluxed sewage gave effluents containing considerable amounts of ammonia, particularly during the first four weeks, in both the series of the experiments. In the presence of bentonite, the effluents from the filter irrigated with refluxed sewage contained more ammonia than the effluents from the filter irrigated with refluxed sewage in the sand series.

In both the series, most of the ammonium sulphate applied to the filters came out in the effluents by the end of the first week, but the ammonia content was considerably more in the effluents from the clay series than in the sand series.

In both the series, during the first 3 weeks the effluents from the filters treated with sewage solids and groundnut cake contained ammonia; comparatively, the effluents from the filter treated with groundnut cake contained less ammonia.

Nitrite nitrogen—The results (not given here) show that small amounts of nitrite were found in the effluents from the filters treated with ammonium sulphate (in the second and third weeks, in the clay series only), sewage solids (in the fourth week, in both the series), activated sludge (in the fourth week, in the sand series), and groundnut cake (in the fifth week, in the sand series).

In the sand series, with raw sewage and refluxed sewage as irrigants, nitrite was found in the effluents in the second week, but the amount of nitrite was very much more with raw sewage.

TABLE III

Ammonical nitrogen (N, mg/l) in the effluents from the different filters

(Weekly averages)

Treatment	First week		Second week		Third week		Fourth week		Fifth week		Sixth week	
	Sand	Sand+ clay	Sand	Sand+ clay	Sand	Sand+ clay	Sand	Sand+ clay	Sand	Sand+ clay	Sand	Sand+ clay
Control	1	nil	nil	nil	nil	nil	nil	nil	nil	nil	nil	nil
Raw sewage :												
Influent	40	75	43	67	40	73	46	76	37	66	43	57
Effluent	33	48	29	32	28	36	22	15	25	7	27	2
Refluxed sewage :												
Influent	28	42	31	37	32	57	28	65	28	54	34	35
Effluent	21	30	38	38	45	53	10	37	13	16	17	11
Sewage solids	21	19	47	26	24	39	15	19	3	8	nil	2
Activated sludge	23	56	21	23	9	10	nil	3	nil	1	nil	nil
Cowdung	15	29	1	7	nil	1	nil	nil	nil	nil	nil	nil
Farmyard manure	11	23	nil	3	nil	12	nil	8	nil	3	nil	nil
Straw*	40	39	clogged	1	clogged	3	clogged	3	clogged	3	clogged	nil
Groundnut cake	49	61	21	95	4	32	nil	22	nil	9	nil	nil
Ammonium sulphate+ superphosphate	163	256	3	14	1	2	nil	nil	nil	nil	nil	nil

*After the first week, the filter containing straw powder and sand was clogged and did not function.

In the clay series, the effluents from the filter irrigated with raw sewage contained nitrite in the first week whereas the effluents from the filter irrigated with refluxed sewage did not show nitrite until after the fourth week.

Nitrate nitrogen—The results (not given here) show that in the sand series, nitrate was found in the effluents from the filters treated with sewage solids (in the fifth week) and activated sludge (in the fifth and sixth weeks). Nitrate was also found in the effluents from the filters irrigated with raw sewage as well as refluxed sewage in the third week.

In the clay series, with raw sewage irrigation, the effluents contained nitrate in the second week and the amount of nitrate was much more than that in the sand series with raw sewage irrigation. With refluxed sewage irrigation, nitrate was not found in the effluents until after the third week and the amount of nitrate was much less than that found with raw sewage irrigation. The effluents from the filter irrigated with refluxed sewage in the sand series contained more nitrate than the effluents from the corresponding filter in the clay series.

Water-soluble phosphorus—The results given in Table IV indicate that the effluents from the sand filters treated with farmyard manure and sewage solids continuously showed appreciable amounts of phosphorus until the end of the experiment. In the clay series, however, the effluents from all the filters (excepting the control filter) contained considerably higher amounts of phosphorus. Practically all the water soluble phosphorus from the filter treated with superphosphate was leached out from the sand series in the first three weeks and from the clay series in four weeks.

Physical and chemical examination of the filter materials

Moisture content—The results (Fig. 2) regarding the moisture contents of the different materials dried over a period of 48 hr show that the materials from the filters containing bentonite clay uniformly held much more moisture than the materials from the sand series.

The results further show that, in the sand series, the filter treated with cowdung retained maximum moisture and the filter treated with activated sludge came next. The moisture content of the material from the filter containing the sand fraction and straw only is not taken into consideration here since the operation of this filter had to be stopped owing to its clogging. Even then it may be noted that the material from this filter held moisture to the maximum extent.

In the clay series, the filter treated with activated sludge retained the maximum moisture and the filter treated with straw came next. The filter treated with raw sewage retained the least amount of moisture.

Formation of water-stable aggregates—The results given in Table V indicate that all the forms of organic matter promoted the formation of water stable aggregates of larger diameter. Aggregates of larger size (2 mm) were formed only in the filters containing bentonite clay. The highest percentage (90) of water stable aggregates (2 mm) was found in the clay series with straw. The filter treated with cowdung in the clay series contained 26 per cent of water stable aggregates.

TABLE IV

Water soluble phosphorus (P, mg/l) in the effluents from the different filters
(Weekly averages)

Treatment	First week		Second week		Third week		Fourth week		Fifth week		Sixth week	
	Sand	Sand+clay	Sand	Sand+clay	Sand	Sand+clay	Sand	Sand+clay	Sand	Sand+clay	Sand	Sand+clay
Control	nil	1.6	nil	0.7	nil	0.2	nil	nil	nil	nil	nil	nil
Raw sewage :												
Influent	4.6	8.8	5.9	7.4	5.1	10.0	4.5	10.1	4.3	9.4	4.4	8.2
Effluent	1.9	4.6	3.9	5.4	3.1	6.0	2.8	5.0	2.4	5.4	2.8	4.7
Refluxed sewage :												
Influent	3.4	8.4	5.2	8.0	2.6	10.3	2.9	10.3	2.2	9.0	3.3	8.4
Effluent	1.0	4.2	3.3	5.3	3.5	5.9	2.9	5.2	1.9	5.9	1.0	4.2
Sewage solids	5.6	22.3	9.8	17.9	7.1	14.0	9.7	10.8	6.1	6.2	4.2	3.5
Activated sludge	6.3	7.9	2.5	3.9	0.9	2.2	0.4	1.8	nil	1.6	nil	0.9
Cowdung	4.9	23.2	1.8	12.2	0.3	8.3	nil	4.8	nil	3.9	nil	3.1
Farmyard manure	32.0	89.1	10.9	21.0	5.6	18.1	5.0	13.7	2.1	9.3	2.4	7.5
Straw*	20.7	12.7	clogged	7.5	clogged	7.8	clogged	5.9	clogged	4.5	clogged	3.9
Groundnut cake	4.4	3.7	0.7	2.0	0.1	1.1	nil	1.4	nil	0.8	nil	0.4
Ammonium sulphate+ superphosphate	11.6	19.0	0.4	2.4	nil	0.8	nil	0.1	nil	nil	nil	nil

*After the first week, the filter containing straw powder and sand was clogged and did not function.

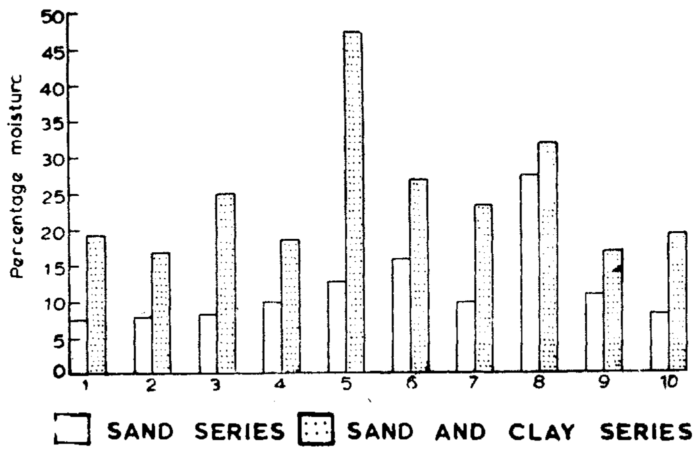

FIG. 2. Influence of clay on the retention of moisture in the air dry materials from different filters. 1, Control filter ; 2, Filter irrigated with raw sewage ; 3, Filter irrigated with refluxed sewage ; 4, Filter treated with sewage solids ; 5, Filter treated with activated sludge ; 6, Filter treated with cowdung ; 7, Filter treated with farmyard manure ; 8, Filter treated with straw ; 9, Filter treated with groundnut cake ; 10, Filter treated with ammonium sulphate and superphosphate.

TABLE V

Percentages of water stable aggregates in the different filter materials

Treatment	Diameter of the aggregates							
	0.25 mm		0.50 mm		1.00 mm		2.00 mm	
	Sand	Sand + clay	Sand	Sand + clay	Sand	Sand + clay	Sand	Sand + clay
Control	58	58	38	40	2	nil	nil	nil
Raw sewage	42	32	56	60	1	2	nil	2
Refluxed sewage	30	20	66	72	2	2	nil	2
Sewage solids	66	10	30	60	1	14	nil	13
Activated sludge	55	18	38	70	4	5	nil	5
Cowdung	24	14	64	50	10	6	nil	26
Farmyard manure	22	42	72	44	2	2	nil	2
Straw	24	2	70	2	4	2	nil	90
Groundnut cake	36	44	62	50	1	nil	nil	nil
Ammonium sulphate + superphosphate	32	46	66	41	1	nil	nil	nil

In the sand series, 2 mm water stable aggregates were not formed. In this series, the filter treated with cowdung gave maximum percentage (10) of 1 mm aggregates.

Total nitrogen—The results given in Table VI indicate that the materials from the clay series invariably retained less nitrogen. In this series as well as in the sand series the materials with cowdung and farmyard manure retained more nitrogen than the other materials.

TABLE VI

Percentages of total nitrogen and phosphorus in the different filter materials

Treatment	Nitrogen (N), mg/100 g		Phosphorus (P), mg/100 g	
	Sand	Sand+ clay	Sand	Sand+ clay
Control filter	4	2	nil	1
Raw sewage	21	8	3	10
Refluxed sewage	17	4	5	4
Sewage solids	31	17	8	13
Activated sludge	35	27	3	15
Cowdung	87	43	4	12
Farmyard manure	82	63	22	24
Straw powder	42	13	3	5
Groundnut cake	16	11	1	9
Ammonium sulphate + superphosphate	5	1	1	2

Total phosphorus— With regard to phosphorus (Table VI), all the filter materials in the clay series retained more phosphorus than those in the sand series. In the clay series, the filters treated with farmyard manure and activated sludge retained more phosphorus.

Micro-organisms in the filter materials

Bacteria, actinomycetes and fungi—The total counts of bacteria, actinomycetes and fungi were considerably more in the clay series than in the sand series (Table VII) because the clay retained more moisture. Madhok (1937) used "synthetic soil" as a medium for the study of certain microbiological processes and noted increased numbers of cellulose decomposing bacteria and decreased numbers of fungi after an interval of 10 and 20 days in the sand mixed with 2.5 per cent bentonite clay. Conn and Conn (1941) also observed increased numbers of "*Bact. radiobacter* and *Bact. globiforme*" inoculated into sand and bentonite mixtures and treated with basal nutrients, viz. 0.1 g glucose, 0.1 g $CaCO_3$, 0.3 g K_2SO_4 and 0.03 g $(NH_4)_2$ HPO_4, in all the cases (1 to 10 per cent bentonite clay with sand).

Yeasts and algae—The results in Table VII show that in the clay series yeasts and algae were suppressed. It may be relevant to refer to the observations of Alexander (1961) who reported that because of the scarcity of the yeasts in soil, it was not yet possible to correlate population size with environmental factors. Further,

TABLE VII

Total counts of bacteria, actinomycetes, fungi, yeasts and algae per gram (dry weight) of the filter material*

Treatment	Bacteria		Actinomycetes		Fungi		Yeasts	Algae
	Sand	Sand+clay	Sand	Sand+clay	Sand	Sand+clay	Sand	Sand
Control filter	3,06,100	33,85,100	3,95,500	2,15,000	1,165	10,000	14,140	19,36,000
Raw sewage	13,27,100	1,13,60,000	2,06,800	6,40,000	73	24,500	272	31,49,000
Refluxed sewage	3,45,400	95,60,000	89,950	4,40,000	405	16,600	1,750	56,01,000
Sewage solids	11,53,600	98,50,000	3,86,450	4,50,000	89	6,000	256	53,65,000
Activated sludge	11,95,000	18,20,000	1,72,400	10,80,000	322	1,500	264	87,48,000
Cowdung	13,63,500	72,39,400	2,36,050	1,41,600	1,663	859	2,034	3,04,30,000
Farmyard manure	7,96,150	35,06,100	2,80,000	2,37,900	749	455	nil	1,30,60,000
Straw powder	9,97,850	17,82,000	45,68,700	16,66,000	145	28,180	12,150	99,01,000
Groundnut cake	96,630	24,00,000	49,080	50,00,000	440	3,500	3,475	29,16,000
Ammonium sulphate+ superphosphate	4,24,250	93,50,000	28,990	10,50,000	11	3,500	247	46,18,000

*Yeasts and algae did not develop in the filter containing clay.

according to him, their relative infrequency suggests that the role of yeasts in soil transformations is negligible, but future work should establish the precise significance of these organisms. He also pointed out that, in a general sense, the algae could not be considered as contributing appreciably to the many biochemical transformations necessary for soil fertility and that under the stress of competition from the bacteria, fungi, and actinomycetes, particularly below the surface, a group poorly adapted to heterotrophy could make only a small impression upon the many biological reactions.

Protozoa—Ciliate protozoa developed in all the filters, but their numbers varied with the kind of organic matter (Table VIII). Closely associated with the formation of 2 mm water stable aggregates in the filters containing clay and straw were the largest numbers of the species of *Colpidium*.

TABLE VIII

Types and numbers of protozoa present in the different filters*
(Number of protozoa per gram of the material)

Treatment	*Amoeba* sp.		*Colpidium* sp.		*Vortiecella* sp.	
	Sand	Sand + clay	Sand	Sand + clay	Sand	Sand + clay
Control filter	nil	nil	200	20	nil	nil
Raw sewage	nil	nil	Few	20	500	nil
Refluxed sewage	500	nil	600	40	40	40
Sewage solids	14,000	nil	8,000	40	nil	20
Activated sludge	5,000	20	Few	20	60	20
Cowdung	10,000	40	nil	800	1,500	nil
Farmyard manure	Few	nil	Few	60	20,000	nil
Straw powder	1,000	nil	nil	1,200	nil	nil
Groundnut cake	4,000	nil	300	20	nil	nil
Ammonium sulphate + superphosphate	nil	nil	400	120	40	nil

*Types here indicate broadly the different species of the genera mentioned in the table. Besides these genera, the straw powder and groundnut cake contained small ciliates (unidentified), 800 and 400 per ml, respectively, in the sand series. Similarly, in the sand *plus* clay series, cowdung, farmyard manure, straw powder, groundnut cake and ammonium sulphate and superphosphate contained 80, 50, 600, 400 and 200 small ciliates, respectively. Species of *Stylonychia, Lionotus* and *Gonostomum* were found in the sand treated with farmyard manure (60/g), sand and clay treated with cowdung (20/g) and sand and clay treated with groundnut cake (300/g), respectively.

Plant growth on the filter materials

The results (Table IX) indicate (i) that the dry weights of plants grown on the filter materials containing clay, were more in the cases of the control filter and the filters treated with raw sewage, sewage solids, cowdung, straw and ammonium

TABLE IX

Dry weights of Sorghum plants grown on the filter materials for 38 days

Filter	Dry weight of plants (mg)	
	Sand	Sand+clay
Control	186	242
Raw sewage	186	400
Refluxed sewage	227	135
Sewage solids	206	256
Activated sludge	192	132
Cowdung	74	300
Farmyard manure	347	280
Straw powder	44	119
Groundnut cake	194	186
Ammonium sulphate+ superphosphate	121	155

sulphate and superphosphate than those grown on the corresponding filter materials from the sand series; (ii) that the dry weights of plants from the filters treated with refluxed sewage, activated sludge and farmyard manure were less than those obtained on the corresponding filters from the sand series; (iii) that the dry weights of plants grown on the filter materials treated with groundnut cake in both the series (with and without clay) were comparable; and (iv) that, in the presence of clay, raw sewage irrigation gave the maximum dry weight and in the sand series farmyard manure gave the maximum dry weight of the plants. The results, as a whole, indicate that the addition of bentonite clay to the sand fraction promoted the plant growth to a greater extent.

DISCUSSION

The main object of the experiments described here was to collect evidence on the general influence or the total effect of clay (bentonite) on the physical, chemical and microbiological changes in columns of the sand fraction treated with some of the common forms of organic matter and inorganic fertilisers used in agriculture.

In the past (Madhok 1937; Martin and Waksman 1940; and Conn and Conn 1941), such mixtures have been used, though not as columns as used in the present study, apparently realising that an admixture of sand and clay is quite suitable for experimental purposes. But these materials were used merely as experimental systems, which provide, unlike soils, the conditions for reproducibility of results. Madhok (1937) stated that one might conclude (from his results employing mixtures of sand and bentonite clay) that the sand-bentonite medium had distinct advantages over artificial media as well as over pure sand media for the study of certain soil microbiological processes, because it was more nearly similar to a natural soil yet it was simple in composition. Furthermore, it could be readily duplicated. The results obtained by the use of this medium could be interpreted readily in terms of soil processes.

As columns of these materials were employed, it was also possible to appreciate that along with the sand fraction only a small percentage (2.5) of bentonite clay could mix with it and allow a reasonable rate of filtration or percolation of water, whereas agricultural soils with good drainage generally contain a relatively high percentage of clay, e.g. Bangalore red soil contains 15 to 25 per cent clay. This, of course, points to a most important aspect of soil structure, which is, as Yoder (1936) put it, that nature through the integrated sum of her environmental factors acting through geological time on different parent rock materials, has produced a great variety of soil types, each of which is characterized by some sort of natural structure or aggregate distribution.

It is generally known that the clay in soil serves to retain the moisture, and the experiments here have strikingly brought out this property of clay. All the filter materials with clay, after air drying for 48 hr, contained invariably higher percentages of moisture than the materials from the sand series, the increases being 15 to 265 per cent, depending upon the type of organic matter.

The more important result of the present investigation is the observation on the formation of water stable aggregates of larger sizes, including 2 mm diameter, in the filters containing clay and organic matter like straw and cowdung. Strikingly, these filters, especially the filter with straw, contained incomparably more numbers of the species of the protozoan *Colpidium*. This filter with straw, clay and *Colpidium* sp. showed 2 mm water stable aggregates to the extent of 90 per cent, and this correlation seems to be of considerable fundamental importance to soil science and of practical significance to agriculture.

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REFERENCES

Agnihothrudu, V. (1957). Aspergilli from soils of South India. *Naturwissenchaften*. **44**, 65.

Alexander. M. (1961). Introduction to Soil Microbiology. John Wiley and Sons Inc., New York. pp. 77–97.

American Public Health Association (1960). Standard Methods for the Examination of Water, Sewage and Industrial Wastes (11th ed.).

Conn, H. J., and Conn, E. J. (1941). Synthetic soil as a bacteriological culture medium. *Soil Sci.*, **52**, 121–136.

Indian Council of Medical Research (1963). Manual of Methods for the Examination of Water, Sewage and Industrial Wastes Special Report Series No. 47.

Kasi Viswanath, G., and Pillai, S. C. (1968*a*). Influence of heat on soil structure. *Curr. Sci.*, **37**, 483–487.

———(1968*b*). Occurrence and activity of protozoa in soil, *J. scient. ind. Res.*, **27**, 187–195.

Kasi Viswanath, G. and Pillai, S. C. (1972*a*). Significance of heating agricultural soils. ***Indian J. agric. Sci.***, **42**, 75–80.

———(1972*b*). Influence of moisture on soil aggregation. *Curr. Sci.*, **41**, 547–553.

———(1973). Pretreatment of the soil in the determination of water stable aggregates, *Mysore J. agric. Sci.*, **7**, 550–556.

Kuster, E., and Williams, S. T. (1964). Selection of media for isolation of Streptomycetes. *Nature, Lond.*, **202**, 928–929.

Lochhead, A. G., and Chase, F. E. (1943). Qualitative studies on soil microorganisms : V. Nutritional requiremints of the predominant bacterial flora. *Soil Sci.*, **55**, 185–195.

Madhok, M. R. (1937). Synthetic soil as a medium for the study of certain microbiological processes. *Soil Sci.*, **44**, 319–322.

Martin, J. P., and Waksman, S. A. (1940). Influence of microorganisms on soil aggregation and erosion, *Soil Sci.*, **50**, 29–47.

Ministry of Housing and Local Government (1956). Methods of Chemical Analysis as Applied to Sewage and Sewage Effluents, H. M. S. Office, London.

Pillai, S. C. (1955). Investigations on Sewage Farming. Indian Council of Agricultural Research, New Delhi.

Pillai, S. C., and Ganguly, J. in association with Cama, H. R. *et al.* (1971). Fifty years of biochemistry at the Indian Institute of Science, Bangalore. *J. scient. ind. Res.*, **30**, 618–639.

Pillai, S. C., and Mohan Rao, G. J. (1952). Influence of filtering medium on the clarification of sewage in biological filters, *Sci. Cult.*, **18**, 289–290.

Pillai, S. C., and Subrahmanyan, V. (1945-46). Life cycles in the transformations of organic matter in sewage, soil and other biological media, *Sci. Cult.*, **11**, 592–596.

Srinath, E. G., Sastry, C. A., and Pillai, S. C. (1962). Removal of phosphorus from sewage by sand filters. *J. scient. ind. Res.* **21C**, 220–222.

Yoder, R. E. (1936). A direct method of aggregate analysis of soils and a study of the physical nature of erosion losses. *Agron. J.*, **28**, 337–351.