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WASTE REGENERATION

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AEROBIC WASTE DISPOSAL SYSTEMS

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INTRODUCTION

The degradation of matter by microorganisms has been viewed by man with mixed emotions, depending upon his interest in the preservation or destruction of certain materials. In either event, a considerable amount of effort has been expended attempting to recognize and understand microbiological processes. Perhaps one of man's more significant achievements has been the development and application of control methods designed to utilize many of the beneficial chemical processes performed by microorganisms.

For space exploration, microorganisms are being asked to provide a means for converting the anticipated waste material of man into reutilizable forms. Two conditions will be imposed on the system; (1) the process must be accomplished rapidly in a limited volume, and (2) the process must be more efficient than other contemplated waste recycling methods, all things considered.

The first condition appears relatively easy to satisfy when the extremes of environment under which microorganisms are known to florish in nature are considered. A controlled physical environment with all the essential substances necessary to the formation of cellular material in approximately the proper proportions will be provided. That a group of microorganisms can be found capable of recycling man's wastes into reusable by-products cannot be seriously doubted since such organisms are already available from our modern sewage treatment plants. A retraining period may be required to accustom the organisms to growth in concentrated wastes.

The second condition, that of relative efficiency when compared to other proposed biological, chemical and physical waste disposal processes, remains a matter of conjecture.

The aerobic microbiological process considered most logical for the processing of wastes accumulating from space explorations because of its compatibility with other components of a closed ecology and its minimum volume and weight requirements, is the activated sludge process. Briefly, the process consists of the establishment and maintenance of a mixed microbial population composed of bacteria, fungi, protozoans and metazoans. These organisms, suspended as flocculent particulates in the waste-sludge mixture, are capable of rapidly decomposing the organic material in the wastes, provided adequate oxygen is available. Part of the material oxidized is assimilated by the organisms for the synthesis of new cells, while the remaining portion is contained in the mixed liquor or liberated as gases, predominantly CO₂ and NH₃. Oxidation efficiencies in the range of 95-98 percent are obtainable, with the degree of oxidation being controlled by alteration of the detention time and/or aeration period.



Utilization of an aerobic microbiological system would offer several advantages; (1) the system would be self-perpetuating and regenerative so that no energy would be required to recover spent oxidants; (2) toxic and undesirable gases would not be formed; (3) odors would be rapidly eliminated; (4) the effluent from the system is rich in mineral substances, particularly nitrates, that could be used for plant nourishment; (5) the sludge sediment contains vitamins and amino acids which would be a valuable food supplement for man or other animals; (6) energy input and heat dissipation would be minimized since biological reactions proceed at relatively low temperatures, and (7) substances present would remain in biologically active forms.

The major disadvantages of a system of this nature include; (1) the requirement for constant monitoring by technical personnel to insure proper process performance; (2) the large fluid volume required to adequately process wastes, and (3) the large quantities of oxygen required for complete oxidation of the wastes.

As regards the requirement for constant monitoring, many of the control tests ordinarily performed on activated sludge processes could be eliminated or simplified so that technical maintenance would be minimized. The addition of automatic feeding, sampling and harvesting devices would further simplify the entire processing procedure. The large fluid volume requirement has been overcome through the careful selection of sludge cultures capable of oxidizing wastes several hundred times more concentrated than those processed by commercial sewage plants. By utilizing selected cultures, the total volume of the system could be greatly reduced. According to one estimate, the oxygen demand of an activated sludge process will vary from 0.5 to 1.3 grams of oxygen per gram of organic matter in the waste (72 to 186 gm. O₂ per person per day), depending upon the degree of oxidation desired. While it is true that corresponding volumes of CO₂ are liberated which would be available for plant propatation, space would be required to house the plant growth units.

A waste disposal system capable of providing the oxygen necessary for the decomposition of wastes from the carbon dioxide resulting from this degradation would be desirable. An independent system of this nature appears readily available by the addition of selected sewage algae to activated sludge cultures. Such a combination would offer additional advantages; (1) active participation of the algae in the breakdown of organic matter, (2) complete isolation of the system from other plant growth or animal systems, thereby eliminating contamination hazards, (3) utilization for space trips of any duration where waste storage presents a problem and, (4) enrichment of the sludge sediment as a food supplement.

MATERIALS AND METHODS

Cultures of microorganisms used in these studies were obtained from two sources. The concentrated activated sludge mixture was taken from the San Antonio, Texas municipal sewage plant as needed. The mixed sewage algae cultures, able to grow luxuriantly at temperatures up to 40°C, were kindly provided by Doctors W. J. Oswald and G. C. Galueke of the University of California at Berkeley. Algae cultures for experimentation were grown in an inorganic medium (Knop's) under increased CO₂ tension (approximately 3%), using artificial illumination (General Electric cool white fluorescent lamps).



Experiments were conducted in a rectangular plexiglas chamber (50.8 cm. wide, 91.4 cm. long, and 3 cm. thick), having a total volume of 13.95 liters (Fig. 1). The cell was supported in an upright position with the shorter axis horizontal to the floor. Openings at the top of the chamber contained a refrigerated water condenser to reduce water evaporation, a dial-type thermometer extending below the surface of the liquid, a port for feeding, and a movable mechanical scraper consisting of two windshield wipers mounted back-toback to dislodge algae adhering to the flat surfaces of the plexiglas walls. Openings at the bottom of the chamber consisted of a port on each side to accommodate a perforated Tygon tubing aerator, and a port for removing fluid samples. Gas flow was provided by two Dyna-Vac air pumps (capacity: 8 liters per minute per pump). The air was circulated from the pumps to the Tygon aerator, where it was dispersed as small bubbles through the sludge-algal mixture. Gas was removed from the cell via the condenser, passed through a foam trap, a ballast system, and returned to the pumps. The aeration period was 24 hours in all studies.

Illumination was provided by two banks of 48-inch Sylvania VHO Daylight Fluorescent bulbs located adjacent to both sides of the panel. Each bank consisted of 12 bulbs mounted parallel to each other with one-quarter inch spacing between bulbs. Total illumination at the outer surface of each side of the chamber was approximately 3,600 feet candles as measured by a Model 200 Photovolt Corporation light meter.

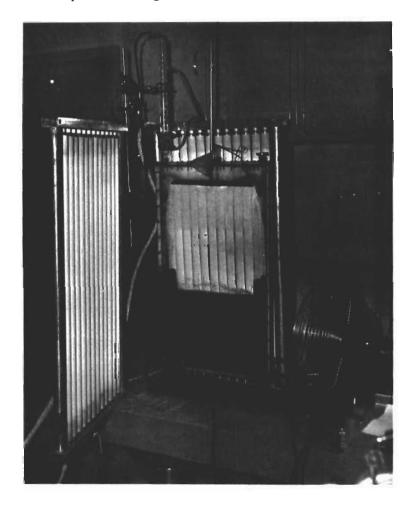


Figure I
Experimental Apparatus



When a run was initiated, fresh sludge and algal cultures were mixed in the reactor in an approximately 3:1 ratio to a total volume of 10 liters. The system was closed, and the culture allowed to equilibrate three days while base line studies were completed. All feces utilized in the studies were obtained from two volunteers on a normal diet, and was first lyophilized by means of a Virtis Freeze-Mobile (Model 10-140). The temperature of the mixed liquor in the reactor was maintained at 37°Ct^{20} C. Excessive foaming was controlled by small additions of Antifoam 60 (General Electric). At four-hour intervals, gas samples were removed for analysis, the temperature recorded, and the hydrogen ion concentration of the mixed liquor determined. The percent oxygen was measured by means of a Beckman F3 oxygen analyzer (sensitivity full scale at 25% 0_2 in 0_2), while 0_2 percentages were determined with a Beckman LB-1 analyzer (sensitivity full scale at 0_2 in 0_2). A Beckman Zeromatic pli meter was utilized for hydrogen ion measurements. Samples for total solids, volatile solids, ash content, and chemical oxygen demand (COD) were taken twice daily, and feed rates and harvesting data obtained. Chemical oxygen demand was determined by the modified procedure of Okey.

Harvesting was accomplished by removing a predetermined volume from the reactor and passing the mixed liquor through a Sharples Super Centrifuge for separation of the cells. The recovered cellular material was dried to a constant weight and stored.

For nutritional studies, the dried sludge-algal cells were ground to a fine powder (microscopic observation revealed few intact cells) and pressed (25,000 psi) into cylindrical pellets by means of a Carver hydraulic laboratory press. These pellets served as the sole source of food for five white mice for a period of 21 days. The weights of the mice were recorded twice weekly.

RESULTS

The data presented were obtained from an activated sludge-algal system which had been in operation for 21 days. During this period, a number of feed rates were examined, varying from 22.5 to 45.0 gm. lyophilized human feces (equivalent to 75-150 gm. wet weight). The feces were suspended in distilled water, homogenized by means of a Waring blendor for five minutes, and added to the reactor. No urine was employed in the studies. A detention period of three days was maintained throughout most of the experiment; however, occasionally, a two-day detention period was required to keep total solids between 5-6 gm/liter.

Figure 2 demonstrates the 0_2 -CO $_2$ percentages and the corresponding hydrogen ion concentration of the culture at four-hour intervals for two days. Chemical oxygen demand is superimposed on the pH graph. It will be noted that shortly after the addition of raw waste to the unit, the sludge microorganisms become active, and the CO $_2$ concentration begins to rise, reaching a maximum 4-8 hours later. Concurrent with the increase in CO $_2$ concentration, a decrease in O $_2$ concentration and pH occurs. Once the CO $_2$ concentration reaches its peak, there is a gradual diminution in its content as it is utilized by the algae. By the

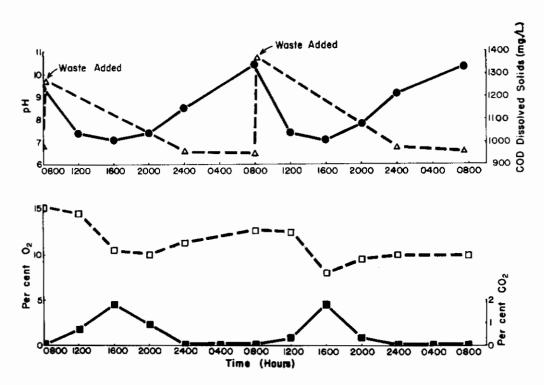


Figure 2 - Graph illustrating percent O₂, percent CO₂, pH, and chemical oxygen demand of an activated sludge-algal system for a 2-day period.

following morning, only minimal amounts can be detected. As the ${\rm CO}_2$ concentration diminishes, ${\rm O}_2$ concentration and pH begin to rise, reaching their peaks when ${\rm CO}_2$ is at its lowest level. It should be observed that the ${\rm O}_2$ concentration of the system is gradually depleted over a period of time, i.e., it never returns to its highest level of the previous day. This phenomenon was noticed in previous runs, and its significance will be discussed later.

The wide fluctuation in the hydrogen ion concentration (from 7 to 10.4) would indicate that the buffering capacity of the culture is limited; however, no deleterious effect on the microbial population was noted. Apparently, the microorganisms can withstand the adverse effects of an elevated pH for short periods of time.

As regards the presence of gases other than 0_2 and CO_2 , samples were removed at intervals and subjected to gas chromatographic analysis. Nitrogen accounted for the remainder of the gas sample; no CH_2 , H_2 or CO was detected. This finding was consistent even when the O_2 concentration was below 3 percent, indicating that O_2 at this low level was not limiting for the microorganisms. Although unpleasant odors were noticeable immediately after addition of waste to the culture, they were completely absent after a two-hour interval.

The chemical oxygen demand of the dissolved solids of the effluent is included in Figure 2 to illustrate its relationship with time to O_2 , CO_2 and hydrogen ion concentrations. Samples were removed for COD determinations to correspond to the disappearance of CO_2 . Recently, Chapman and Okey⁵ found that for activated sludge culture, the COD of the dissolved solids of the effluent was removed from the effluent within an hour after addition of the raw wastes.



They attributed the disappearance to the adsorption of the waste to the floculent sludge particulates. That a similar relationship exists for activated sludge-algal cultures is demonstrated in Figure 3. Samples were removed at 20-minute intervals and subjected to COD analysis. It may be seen that within a two-hour period, the COD of the effluent is almost completely removed.

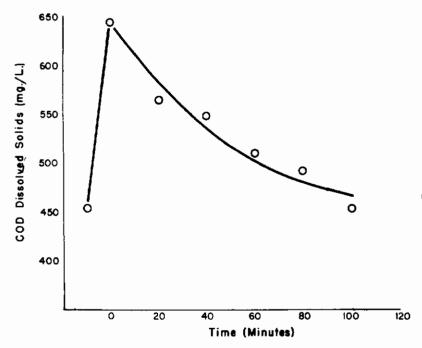


Figure 3

Graph illustrating rapid removal of dissolved solids from the effluent of an activated sludge-algal culture as measured by the chemical oxygen demand test.

Table 1
Chart depicting weights of mice during feeding experiment, using dried activated sludge-algal pellets as a sole source of food.

Mouse Numb e r	Weight (gm)						Net	
	6	12	MARC 15	н 1962 19	23	27	Loss	
	31.0	26.3	26.5	25.1	24.6	25.2	5.8	
2	34.0	30.2	30.5	27.8	27.5	26.6	7.4	
3	34.0	30.0	31.2	31.1	30.1	29.6	4.4	
4	29.1	25.2	26.2	24.0	24.4	23.1	6.0	
5	36.3	32.4	33.6	32.9	31.4	30.2	6.1	
Average loss							5.9	



Microscopic and cultural studies revealed the following general findings. The major algae genera isolated from the mixed liquor consisted of Chlorococcum, Chlamydomonas, Oscillatoria, and Chlorella. Chlorococcum was always greatly in the predominance with the latter three genera present in varying numbers, depending upon the time of observation. Chlamydomonas species were present in greatest numbers immediately before a feed. The rise in algae population could be followed visibly through the changing of the coloration of the mixed liquor from light brown to dark green in 24 hours. Bacteria isolated from the sludge included the normal intestinal organisms of man, plus a variety of Bacillus and Spirillum species. Paramecia and rotifers (2 major species of each) were the predominant animal forms noted, with occasional flagellated and amaeboid protozoans observed. Following the addition of fecal material, a sharp increase in the number of bacterial forms was seen. This increase was followed by a rise in the numbers of active paramecia and rotifers. Both the paramecia and the rotifers were actively feeding upon the bacteria; however, no engulfment of algal cells was noted. The number of bacterial cells was subsequently reduced so that by the time of the following feed, few were present. Concurrent with the reduction in bacterial population, encysted protozoan forms appeared. Rotifer population remained fairly constant throughout the cycle. Aggregation of the cells into clumps made quantitation of the individual types impossible.

Preliminary studies to determine the nutritional qualities of the dried, ground activated sludge-algal solids were conducted, using five adult, white female mice. Control mice were not included in the experiment since it was desired to determine only if the mice could survive utilizing the sludge-algal solids as a sole source of food. Results of the feeding experiment are shown in Table I. It will be seen that an initial weight loss of approximately 4 gm. per mouse occurred during the first week. Such a loss is not unusual when mice are transferred from one diet regimen to another. The inability of the animals to regain the original weight, plus evidence of slowly continuing weight loss, would indicate that the dried solids are deficient in some essential factor(s), or unpalatable to the mice in the form offered. No nutritional analyses of the solids was performed. The mice remained alert and active throughout the experiment. Termination of the test after 21 days was necessary because the supply of dried pellets was exhausted.

DISCUSSION

The practicability of utilizing an activated sludge-algal mixture for the decomposition of man's most concentrated excretory product over an extended period of time has been established. A 10-liter reactor maintained on a 3-day throughput rate has been shown to be capable of satisfactorily decomposing the average daily fecal output of man (100 gm. wet weight), with loadings up to 200 gm. for short periods of time. Assuming the combined total wastes/man/day will be in the order of 250 gm., then the total volume required for their decomposition will approximate 25 liters. This volume compares favorably with Pipes' estimate of 30 liters/man/day. Although the volume required for a system of this type appears somewhat larger than other proposed processes, it should be remembered that by producing most of the oxygen required for the decomposition of wastes directly in the reactor, other necessary O2 producing systems could be correspondingly reduced in size.



Under conditions of these experiments, results indicate that total solids cannot be allowed to accumulate above 5-6 gm/liter. When the solids content increases beyond these levels, excessive turbidity of the culture interferes with light penetration and subsequently reduces growth of the algae. To maintain total solids between 5-6 gm/liter, detention periods in the range of 2-4 days were necessary. While dissolved solids are adequately decomposed at detention periods of this duration, as evidenced by COD determinations, the extent of the breakdown of suspended solids was not measured. Determinations of this parameter will be included in future studies. The rapid removal of dissolved solids from the effluent would mean that excess dilution water could be recovered for reuse almost immediately.

The small daily loss in O_2 percentage of the system may be accounted for if it is assumed that the algal cells are actively participating in the degradation of organic compounds present in the wastes. Under these conditions, the algal organisms would require O_2 and would represent a drain on the total oxygen supply. The small loss in O_2 does not present a serious problem since O_2 apparently does not become limiting until extremely low concentrations are reached, provided adequate aeration is present. The system could be flushed with O_2 approximately every ten days to restore the desired O_2 level.

The limited feeding experiments conducted with mice indicate that while the dried sludge-algal solids are capable of sustaining life of the animals, the solids are unsatisfactory as a sole food source in their present form. There is no doubt, however, that the solids contain many essential nutritional substances which should prove useful for food supplementation.

SUMMARY

Man's exploration of space for extended time periods will necessitate the development of processing procedures for the recycling of accumulating wastes into edible forms. The advantages and disadvantages of utilizing an activated sludge process for the partial conversion of the wastes into useful by-products are considered. One of the major objections to the use of a system of this nature is the large volumes of $\mathbf{0}_2$ required for complete oxidation of the wastes. This requirement would impose a drain on the oxygen reserves of the closed ecology.

A microbiological waste disposal system, capable of satisfying its own oxygen demand from the evolved CO₂, would alleviate the depletion of oxygen reserves and offer the additional advantages of being completely separated from other components of the closed ecology. A system designed to accomplish this objective was devised by the addition of selected sewage algal strains to activated sludge cultures. The system, housed in a flat, plexiglas chamber and illuminated by means of fluorescent light banks, was closed to the atmosphere, and tested for its ability to decompose human fecal solids over a period of 21 days. Under the experimental conditions employed, it was demonstrated that while all the O₂ required for the oxidation of the wastes was not generated, the system was capable of producing significant quantities for oxidative purposes. Future studies will include the performance of the system following the addition of urine concentrates and other anticipated extraneous wastes.

Contrails

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