Reprinted from CHEMICAL ENGINEERING SYMPOSIUM SERIES, Water—1970, Volume 67, Number 107

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APPLICATION OF MICROBIOLOGY AND BIOENGINEERING PRINCIPLES TO BIOLOGICAL WASTE TREATMENT

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The scientific and engineering literatures relevant to aerobic sewage treatment were searched to identify factors limiting sewage treatment plant efficiency. A primary problem is the lack of knowledge concerning the identity of microbial species responsible for the biodegradation of sewage. Until these species are known, current capabilities to control environmental conditions in the treatment plant cannot be applied intelligently.

The following specific factors are among those which might limit biological activity in the sewage treatment milieu. Organic substrates are very dilute in sewage, approximately 1,000 times more dilute than in microbial growth media. Similarly, inorganic nutrients are present in suboptimal quantities. The average temperature in sewage treatment aeration basins is significantly below the optimum for intestinal organisms. Recent evidence strongly suggests that oxygen application rates may be as much as one order of magnitude below optimal. Surface area for microbial growth is probably not adequately available in aeration basins.

The need for automated chemical and biological assay equipment with feedback control over the sewage treatment process to apply limiting factor information is discussed.

Although sewage treatment is the largest industry in the world (on the basis of total volume processed), the biological principles upon which most treatment is based are poorly understood and incompletely applied. In an attempt to assess the situation, a literature review (*1*) was undertaken to determine factors in the aerobic treatment process which may limit the degree of biological treatment. The objective was to identify those limiting factors which might be controlled to improve performance in the sewage treatment plant. It was hoped that the study would help to improve biological purification effectiveness and obviate or defer the need for tertiary physicochemical treatment stages.

The most disturbing finding of the study is that knowledge concerning the identity of the microorganisms principally responsible for sewage treatment is in a primitive state. The majority of the microbial identification work in sewage treatment plants was done in the 1920's, 1930's, and early 1940's. Since that time very little productive effort has been expended on such work. Meanwhile, the nature of domestic sewage has changed with the introduction of detergents, garbage grinders, and new food preparation and consumption habits. Despite these substantial changes, the knowledge about the identity of microbial population—the functional part of the sewage treatment system—remains essentially that of 25 years ago, when it could only be described as poor.

Not only is there a paucity of information on the species responsible for sewage purification, but the few published studies are not generally in agreement as to the identity of these species. Moreover, there is disagreement regarding which genera and phyla(!) are most active. Some researchers claim that bacteria are the organisms affecting sewage treatment while others assign

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that role to protozoa. Organisms isolated from sewage have been identified in individual studies (*2* to *23*). However, only one systematic, qualitative study of the various microorganisms appearing in different components of a sewage treatment plant was found in the literature (*23*). This study was done in 1928. The literature search generated a list of 70 species of bacteria and protozoa identified in various studies. Only a very small number of these species were reported in more than one reference.

Modern biology strongly suggests that a wide variety of microorganisms participate in the metabolic processes by which sewage is degraded. The Embden-Meyerhof pathway and the Krebs cycle, as parts of life's evolutionary heritage, are common to very large numbers of species, including those active in sewage. However, the environmental conditions which are optimal for these processes vary widely in different species. A number of these conditions can be controlled economically in sewage treatment plants to inhibit or select for some species. This flexibility offers the prospect of improved biological treatment and makes identification of the active microorganisms of critical importance.

Another key finding of the study is that despite widespread belief to the contrary, relatively little microbial reproduction usually occurs in aeration tanks of activated sludge plants (*24*). The evidence strongly indicates that the sewage microorganisms remain in lag phase most of the time they are detained in the sewage treatment plant (*25*).

Primarily, the study sought specific reasons why metabolism, growth, and reproduction of sewage organisms fail to attain their inherent potential rates in aeration treatment.

SUBSTRATES

The primary function of the microorganisms, from the standpoint of sewage treatment, is the assimilation and degradation of the dissolved organic fraction of the sewage. Through the metabolic oxidation of this organic substrate, the microorganisms derive the required energy for metabolism and synthesis of new cells which subsequently metabolize additional substrate.

Substrate concentrations in domestic sewage are as much as 1,000 times less than substrate concentrations generally prescribed for bacterial culture media for the same groups of organisms (*26*). Thus, unless additional substrate is added to the sewage, or unless the substrate in the sewage is concentrated for the benefit of the microorganisms, optimal growth is unlikely. While the addition of substrate will increase the amount of substrate consumed, the effluent BOD will exceed that prior to the addition of the substrate. Concentration of available substrates already present in the sewage, on the other hand, would increase microbial activity and reduce total effluent BOD.

INORGANIC NUTRIENT CONCENTRATIONS

When reported analyses of inorganic constituents in sewage are compared to optimal nutrient requirements cited for various microorganisms, as presented in Table 1, it is revealed that sewage is deficient in almost every inorganic constituent generally required for optimal microbial growth. Those falling within this category include magnesium, sodium, calcium, iron, phosphate, and nitrogen. In some instances, the forms in which these nutrients are available to the microorganisms constitute a problem.

TABLE 1. REQUIRED CONCENTRATIONS OF VARIOUS INORGANIC IONS FOR OPTIMAL GROWTH OF *Escherichia coli* AND THE CONCENTRATIONS OF THESE IONS FOUND IN SEWAGE

	Concentration for Optimal Growth	Concentrations In Sewage (28)
	mg/l	mg/l
Sodium	2300	125
Potassium	3900, 100 (29)	10
Ammonium		20
Calcium	400	25
Magnesium	240, 10-20 (30), 6 (30)	5
Iron (Fe^{++})	2 (29), 0.025-0.1 (31), 2	less than 1
Bicarbonate		200
Sulfate		50
Chloride		50
Phosphate	300 (29)	5
Zinc	7	
Manganese	6	

Some of the nutrient concentrations reported in sewage are sufficiently close to the microbial requirements, or the microbial requirements are so low that addition of these nutrients to aid sewage treatment might be feasible.

Of additional interest is the possibility that carbon dioxide might normally be expelled from aeration tanks, by being entrained with the other exhaust gases, to the extent that this recently recognized nutrient becomes limiting.

TEMPERATURE

The temperature of sewage is almost always significantly below the optimal temperature for growth of the majority of species likely to be present. This undoubtedly is a major limiting factor for sewage treatment. Since elevating the temperature of the entire body of sewage is too costly, the only approaches to alleviating this problem would consist of concentrating the microorganisms and substrates into relatively small volumes for heated incubation periods, or, using the opposite approach, adjusting other environmental parameters to select for psychrophils.

OXYGEN

Early reports in the engineering literature largely discount the effect of increased oxygen availability above several tenths of a milligram per liter in mixed liquor. However, the scientific literature strongly supports other, and generally more recent, reports (*32, 33*) in the engineering literature that the rate of aeration applied in most aeration sewage treatment plants is limiting. Lack of sufficient oxygen inhibits BOD reduction and phosphate removal by microorganisms (*34*). Comparison between the scientific and engineering literatures indicates that normal activated sludge plants may be one to two orders of magnitude deficient in the provision of dissolved oxygen. It seems possible that high initial rates of aeration or oxygenation, followed by rates currently practical, may shorten treatment periods, thereby rendering such alteration in treatment economically feasible. Pilot-plant studies within the current state of the art could establish the facts in this case.

SURFACE AREA

Sewage treatment tanks and basins in the activated sludge or other aeration processes, in contrast to trickling filters, present little surface area to which microorganisms can attach. The scientific literature (*35* to *37*) indicates that available surface is highly important to microbial growth and reproduction. The organisms present in mixed liquor may suffer from lack of attachment surface. The effect of available surface on microbial growth is of particular importance in dilute media, such as sewage. Plant data collected from 90 operating years show that average substrate concentrations fell within the range in which the greatest benefit from added surface has been reported (*38*). The geometry of sewage aeration basins and the aeration process might possibly be altered to provide increased surface.

DIFFUSIBLE INTERMEDIATES

There is evidence that intermediate metabolites elaborated by microbial cultures serve to reduce the lag period imposed on new cells or subcultures. The diffusible intermediates are probably amino acids or biochemicals quickly produced from amino acids (*39, 40*). Most of these intermediates are left behind in the supernatant when concentrated sludge is returned from the settling basin to the aeration basin. The lag period in the aeration basin may thus continue until the microorganisms produce the required amounts of intermediates. Specific identification of such intermediates and measurements of their effectiveness would provide the data necessary to determine whether it would be useful or economical to provide required levels of the intermediates to the aeration basin. Such studies would also reveal the effect of sludge age upon the production of the intermediates.

OSMOTIC PRESSURE

Organisms accustomed to the intestinal tract, and many other microorganisms finding their way into sewage, undoubtedly undergo osmotic shock in their new, low-solute milieu. The extent of damage incurred by the microorganisms and the effect on the sewage treatment process have not been ascertained in the literature search. Although it is probable that low osmotic pressure prolongs the lag period in the aeration basin and selects against numerous species of microorganisms, it is doubtful that the osmotic pressure of sewage can be economically increased to offset these effects. However, it is possible that control of other environmental parameters in the sewage treatment process may indirectly reduce the effects of osmotic shock on microorganisms of interest.

NITRATE REDUCTION

As in the case of phosphate, the removal of nitrate from sewage effluents is desirable in combating eutrophication. While microorganisms present in sewage can reduce nitrates to atmospheric nitrogen, this process requires anaerobic or semiaerobic conditions and relatively long treatment periods. Although anaerobic conditions for phosphate stripping have been proposed, only the sludge would be subjected to anaerobiosis, whereas, in the case of nitrate reduction, the entire sewage flow must be deprived of oxygen. Thus the biological methods for BOD reduction and phosphate removal are incompatible with that of nitrate reduction. One or the other should be selected as a treatment goal, and the plant designed and operated accordingly.

BIORHYTHMS

Diurnal fluctuations in sewage quantity and quality have long been noted. More recently, variations in sewage treatment efficiency which seem dependent on the time of day, but independent of sewage quantity and quality, have been reported (*41*). The scientific literature reports extensively on biological clocks (*42* to *44*), mechanisms which, in varying degrees, control the metabolism and activity of almost all organisms. Metabolic rates may change markedly in accordance with these biorhythms. Whether or not the rhythms are endogenous to the organisms or are triggered by something in the external environment remains a mystery. In either case, however, the ability of microorganisms to assimilate and degrade sewage may vary considerably with the time of day. If this fact were established to have an appreciable effect on sewage treatment, alterations in the design of sewerage systems, holding tanks, and treatment plants might be made to emphasize utilization of the microorganisms during their most productive periods.

Another type of limiting factor in sewage treatment results from inadequacies in current determinative methods for the assay of critical treatment parameters and in the lack of means for the immediate application of the results. An example is the need for an assay for the determination of the amount of activated sludge to be returned to the aeration basin. Present control is based upon maintaining some prescribed suspended solids concentration in the mixed liquor. However, the parameter of interest is not that of suspended solids, but of living organisms. A rapid means for determining active biomass, as opposed to suspended solids, would probably enable significantly better control of the activated sludge process.

A case in which an improvement in assay technique has been made is the use of the dissolved oxygen electrode to replace the time-consuming wet chemistry method for measuring dissolved oxygen. However, the new assay method has not been utilized in treatment plants for the direct control of aeration. To overcome this type of limitation, it is necessary (1) to develop the critical set of assays required for sewage treatment control, and (2) to automate these assay methods into feedback systems with response times sufficiently short to achieve the desired control.

Microbiological data were extrapolated (*1*) from the literature to estimate the efficiency of microorganisms operating in the sewage treatment environment. The results indicate that microorganisms in a sewage treatment plant remove substrate at approximately 3 to 14% efficiency based on the rates reported for optimal culture conditions. These are gross estimates hazarded in order to gain some rough magnitude of the effect of the limiting factors studied. Undoubtedly, other important limiting factors escaped the attention of the authors. Perhaps some limiting factors are not yet reported in the literature. However, the conclusions and inferences made possible by this study strongly uphold the hypothesis that very substantial improvements in the efficiency of biological treatment of sewage may be possible through relatively modest efforts to determine and apply biological information.

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