



Nitrogen utilization in the activated sludge process  
by Curtis Kenneth Townsend

A thesis submitted to the Graduate Faculty in partial fulfillment of the requirements for the degree of  
MASTER OF SCIENCE in Civil Engineering  
Montana State University  
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**Abstract:**

Under man's influence the process of eutrophication can be greatly accelerated by the concentration of nutrients in wastewater discharges. Biological growth can be controlled in a body of water by limiting the amount of any element required for growth of microorganisms. Nitrogen is required for the formation of amino and nucleic acids which are essential parts of living cells.

This research involved construction of a pilot plant and a study of nitrogen removal by the activated sludge process with cell retention time as a variable. In the conventional activated sludge process nitrogen can be removed by wasting cells that contain nitrogen.

One way to optimize nitrogen removal is by maximum cell wasting. However, this is limited by the necessity to keep the food to microorganism ratio within the proper range for adequate sludge settling.

A second approach involves wasting cells with a maximum concentration of nitrogen. The results of this experiment indicate insignificant changes in the nitrogen concentration of the activated sludge with cell retention times from 1.72 to 10.15 days.

The results of this research indicate that fixation may add significantly to the nitrogen content in the effluents from activated sludge treatment. In processes with no cell wasting (extended aeration), the nitrogen concentration of the effluent may exceed that of the influent.

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Signature *Carl O. Lawrence*  
Date *June 12, 1972*

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Bozeman, Montana

August 1972

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## ABSTRACT

Under man's influence the process of eutrophication can be greatly accelerated by the concentration of nutrients in wastewater discharges. Biological growth can be controlled in a body of water by limiting the amount of any element required for growth of microorganisms. Nitrogen is required for the formation of amino and nucleic acids which are essential parts of living cells.

This research involved construction of a pilot plant and a study of nitrogen removal by the activated sludge process with cell retention time as a variable. In the conventional activated sludge process nitrogen can be removed by wasting cells that contain nitrogen.

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## CHAPTER I

### INTRODUCTION

#### ROLE OF NUTRIENTS

Eutrophication is a process which involves an increase in the biologic productivity of a body of water as a result of nutrient enrichment. This occurs naturally, but under man's influence excessive amounts of nutrients can enter an aquatic ecosystem through wastewaters resulting in an acceleration of the eutrophication process.

Until recently, the degree of wastewater treatment was usually measured relative to the oxygen requirement and suspended solids of the effluent from treatment processes. The effect of the effluent on the oxygen balance of the receiving waters was of particular interest. Biological treatment facilities were often capable of removing up to 95% of this oxygen demand as measured by the five day biological oxygen demand (BOD<sub>5</sub>) test. This in turn represents a portion of the amount of oxygen required to convert any biologically oxidizable carbon in the wastewaters to CO<sub>2</sub>. Even when this BOD<sub>5</sub> was reduced to 95% of its original value, there were many cases where considerable growth of plants and microorganisms was evident below wastewater discharge points in the stream. This growth was in excess of what could be supported by the remaining 5% of the carbon available.

When sufficient carbon, hydrogen, nitrogen, oxygen, phosphorous and other required elements are present in a body of water; bacteria, algae and other flora will proliferate. If these elements are introduced to a

stream, river or lake, growth of plants and microorganisms will proceed to the extent that one of the elements becomes limiting. This phenomenon was first noted by Liebig (1), who demonstrated that growth is limited by the nutrient which is available in the least abundance relative to the nutritional requirements.

Excessive algal growths can occur in carbon rich receiving waters if triggered by a small amount of nutrients remaining in wastewater effluents even after the best secondary treatment. In recent years research in waste treatment has shifted toward optimizing processes for the removal of nutrients as well as carbon from wastewaters. The work of this thesis involves the study of nitrogen in the activated sludge waste treatment process with the aim of finding a means for optimizing nitrogen removal.

#### ROLE OF NITROGEN

Molecular nitrogen ( $N_2$ ) comprises 78.084% by weight of the atmosphere, but it is chemically inert and cannot be used by most organisms. These organisms must obtain nitrogen from the environment in some combined form such as nitrate or ammonia. Nitrogen in these combined states seldom exceeds a few parts per million (ppm) in surface water and soil, and its concentration often becomes the limiting factor in the growth of living organisms (2). Infrequently, it may reach levels greater than 100 ppm in ground water where it is not subject to biological uptake (3).

Nitrogen is a requirement for the formation of amino and nucleic acids, which are essential parts of living cells. Therefore, the amount of microbial growth in any body of water can be controlled to the extent that the useable nitrogen content can be controlled.

Grundy (4) has stated that 1,035 to 4,210 million lbs of nitrogen enter the aquatic ecosystem in the United States annually through natural processes. Contributions to useable nitrogen from natural sources include decomposition of organic materials from benthal deposits in lakes and rivers, decomposition of organic materials in the soil and subsequent runoff to streams, rivers and lakes, dissolution of salts containing nitrogen, and fixation of molecular nitrogen. He estimated the additional nitrogen entering this same ecosystem annually under man's influence to be 3,990 million lbs. This 3,990 million lbs is approximately equal to the maximum amount (4,210 million lbs) which enters the aquatic ecosystem through natural causes. Were nitrogen always the limiting nutrient, the rate of eutrophication of the entire aquatic ecosystem would double. If excess nitrogen is provided to a receiving water, then algae and bacteria and other aquatic plants will continue to grow until some other nutrient such as phosphorous or the substrate, carbon, becomes limiting.

## OBJECTIVES

### Design and Construction of Pilot Plant

The first part of the research entailed the design and construction of an activated sludge pilot plant. This included a continuous-flow, stirred-tank reactor, a gravity clarifier and the necessary feed, waste and recycle pumps with all controls and other appurtenances required for operation.

### Nitrogen Removal

The second part of this research involved measuring the nitrogen utilization by activated sludge organisms when varying the cell retention time i.e. sludge age while all other parameters were held constant. This experiment was carried out in a 6-liter pilot plant under constant conditions of feed, temperature and other variables.

The original intent in this part of the research was to measure the nitrogen utilization by activated sludge organisms with temperature as a variable, but the temperature control in the environmental cabinet was unreliable. Hence, it was expedient to investigate the nitrogen utilization by activated sludge as a function of the cell residence time.

## CHAPTER II

### ACTIVATED SLUDGE

#### PROCESS FOR WASTEWATER TREATMENT

Activated sludge is a process for removal of substrate carbon from wastewater by a community of microorganisms. Wastewater is introduced to an aeration tank containing an acclimated biological culture, called activated sludge. The culture utilizes the soluble and colloidal organic and inorganic carbon in the waste for respiration and growth. Competition between organisms, temperature, composition of the waste, cell retention time, and many other parameters tend to favor the dominance of certain species. In wastewater treatment the culture is continuously inoculated with new organisms from the influent waste, but this is not true in the pilot plant experiments herein reported because extensive effort was expended to keep the influent feed free from microorganisms.

The activated sludge process depends on the retention of the microorganisms in the aeration tank for a period longer than the hydraulic residence time. From the aeration tank the effluent flows to a settling basin where the microorganisms are separated from most of the remaining liquid by gravity. The settled microorganisms are then recycled back to the aeration tank. In most wastewater treatment plants a portion of the recycle microorganisms are wasted to maintain a steady population of activated sludge in the aeration tank. The amount of microorganisms

wasted plus those carried out in the effluent from the settling basin must equal the growth rate to maintain a constant mass population.

The success of the activated sludge process is primarily dependent on the settleability of the microorganisms. The settleability of the organisms has been shown (5) to be dependent on their growth rate. The growth rate can be controlled by the concentration of feed and organisms. This is expressed as the food to microorganism ratio and is generally expressed as the total lbs of BOD<sub>5</sub> entering the reactor per day divided by the total mass of microorganisms in the reactor which is sometimes termed as loading rate:

$$\text{Loading rate} = \frac{\text{lbs BOD}_5}{\text{Day}} \cdot \frac{1}{\text{lbs MLSS}}$$

where

BOD<sub>5</sub> = The five day BOD of the waste

MLSS = Mixed liquor suspended solids which represents the dry weight of unfilterable solids and is nearly the same as dry weight of organisms

The microorganisms generally exhibit satisfactory settling characteristics if the food to microorganism ratio is between 0.2 and 0.5/day (6). The concentration of the influent and effluent was measured as total organic carbon in this experiment. BOD<sub>5</sub> was not measured since this test is time consuming and is not accurate.

## ORGANISMS

Diaz et al (7) isolated over 300 bacterial strains by plating samples of activated sludge on sewage agar. Gram negative bacteria of the genera Zoogloea and Comamonas were predominant. Other workers including Allen (8), Jasewicz and Porges (9), and Rogovskoya and Lazareva (10) also studied activated sludge but were unable to find Zoogloea. Anderson and McCoy (11) showed that the dominant bacterial species of activated sludge was Pseudomonas.

Many of the bacteria isolated by Diaz et al (7) were tested for their ability to stabilize sterilized raw sewage. None of the isolates produced an effluent of equal quality to that of the entire activated sludge; however, Zoogloea did form flocs. They also speculated that the bacteria had a great advantage over protozoan in utilizing soluble substrate because the bacteria have a much larger surface to volume ratio.

Diaz et al (7) also found that many of the isolates contained poly-beta-hydroxy-butyric acid (PHB). From this they speculated that this might be the means by which the organic matter of sewage is rapidly removed during the early stages of activated sludge treatment and is then subsequently metabolized. PHB has been shown to act as a reserve material in many species (12, 13, 14, 15). This early removal could be the conversion of organics to stored PHB which is metabolized during subsequent aeration.

A study of 150 bacterial strains isolated from raw sewage indicated

significant differences from activated sludge (7). Coliforms, which constitute nearly a quarter of the bacteria in sewage isolates, were rarely encountered in sludge.

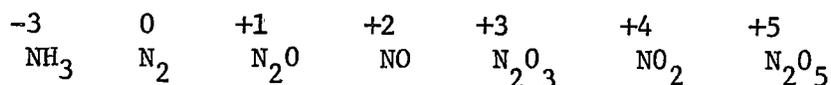
Prakasam et al (16) demonstrated that the plating medium has a significant effect on the species isolated from activated sludge. They speculate that it is impossible to cultivate many of the bacteria from sludge on any known media. Thus, while there have been many studies on the composition of activated sludge, there are still many bacterial strains that have yet to be isolated. They attained their highest productivity using activated sludge extract agar.

## CHAPTER III

### NITROGEN METABOLISM

#### VALENCE STATES OF NITROGEN

Inorganic nitrogen can exist in the following seven states of valence (17):



The principal biological processes involving nitrogen are shown in Figure 1 (18). The individual processes are discussed in the sections of this chapter on "NITRIFICATION," "DENITRIFICATION," "CELL SYNTHESIS," and "FIXATION."

Fewson and Nicolas (19) have proposed the pathway shown in Figure 2 as the most likely in the assimilation and dissimilation of nitrogen. Nitrite ( $\text{NO}_2^-$ ), nitric oxide ( $\text{NO}$ ), and hydroxylamine ( $\text{NH}_2\text{OH}$ ) are well established intermediates in the pathway. However, the evidence that nitroxyl ( $\text{HNO}$ ) and nitrous oxide ( $\text{N}_2\text{O}$ ), are intermediates is inconclusive (18).

Nitrogen in the -3 valence state is incorporated into organic compounds for cell synthesis. Some bacteria and algae are capable of reducing the higher valence states of nitrogen to the -3 state for this utilization, but most higher forms of animal life are not capable of this reduction and require their nitrogen supply in the  $\text{NH}_3$  state

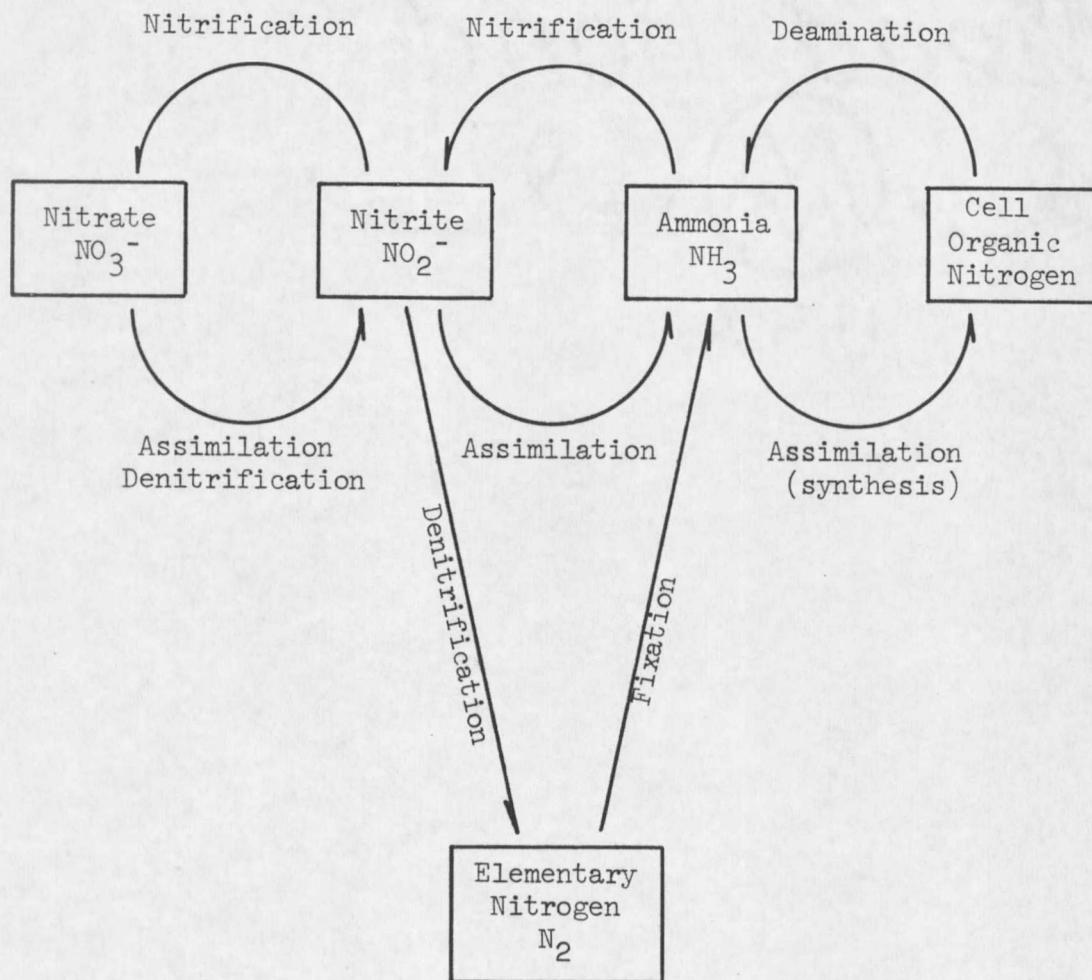


Figure 1. PRINCIPAL BIOLOGICAL PROCESSES INVOLVING NITROGEN (18)



























































































































































































