

ENERGY AND PROTEIN PRODUCTION
FROM PULP MILL WASTES

Progress Report
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ABSTRACT

During the past quarter significant progress was made toward demonstrating the feasibility of producing protein and methane from pulp mill waste materials. The most significant result obtained in the project to date occurred when ozonated spent sulfite liquor, SSL, was demonstrated to be a suitable substrate for biosynthesis. Sustained production of methane was obtained by biological conversion of ozonated SSL. Total methane production approximated 2 volumes of gas per volume of ozonated SSL after approximately 3 days reaction time. A detailed study of the effect of pH on ozonation indicates that low pH's favor the breakdown of SSL into organic fragments that are more easily assimilated by microorganisms. In addition, approximately one-half as much ozone is required to effect maximum degradation at pH 4 as compared to pH 8. Even with this lower dosage of ozone the resulting product is more amenable to bioconversion. The results were confirmed by BOD measurements on ozonated products and the biosynthesis of methane. There is even a preliminary indication that ozonation upgrades the substrate to a level suitable for the biosynthesis of protein with *Torula* yeast.

BACKGROUND

The current quarter was initiated with all apparatus installed in an operating order. Cultures of the different microorganisms requested earlier had been received and were being maintained on slants and shake flasks. Exploratory work defining the useful range of process variables had been conducted and new experiments designed to investigate process feasibility under the most favorable conditions.

STAFFING

The project is fully staffed and operational. Included are two principal

co-investigators, one post-graduate research assistant and one Ph.D. graduate student. Three part-time undergraduate assistants have been added to assist in the numerous analytical and biological tests which must be conducted routinely in order to evaluate progress. This undergraduate research experience is proving valuable to the students education as well as an indispensable aid to the project. Dr. Jurgensen increased his contribution to 20% of his time allowing Dr. Patton to reduce his participation to 20%. Further exchange of responsibilities are planned during the next quarter to bring the annual manpower expended into agreement with the budget. Performance under this contract is in strict compliance with the terms and requirements specified by ERDA.

RESULTS AND DISCUSSION

Construction of the packed column reactor was completed during this quarter. Ozonations were conducted in continuous and batch modes of operation. Results generally paralleled those obtained in exploratory flask ozonations. Total ozonation time was reduced due to the more efficient gas-liquid contacting accomplished in the column reactor. A decision was made to ozonate SSL until ozone appeared in the effluent gas stream. The time required varies from 3 hours at pH 4 to about 6 hours at pH 8. Not only is the ozone consumption approximately one-half at the lower pH but the product is more easily assimilated by micro-organisms. Hence, experiments in this quarter have utilized material ozonated between pH 4 and pH 6.

Cultures of microorganisms useful for the production of methane, protein and desulfonylation were acclimated to the ozonated SSL substrate. The organisms were first cultured until vigorous growth was obtained using synthetic nutrients described in the literature. Synthetic nutrients were gradually reduced and substituted with 50% ozonated SSL and 50% water until they were completely eliminated.

At this point methane production averaged approximately two volumes of methane per volume of SSL fed with a residence time of approximately 3 days. For comparison purposes a fermentation was conducted in which the SSL is enriched with 25% of the initial starter growth nutrients. Methane production from this reference fermenter averages approximately 25% higher than the straight ozonated SSL system. All reactors are run in a continuous mode and the results obtained for a three week run are shown in Figure 1. The higher curve is the reference

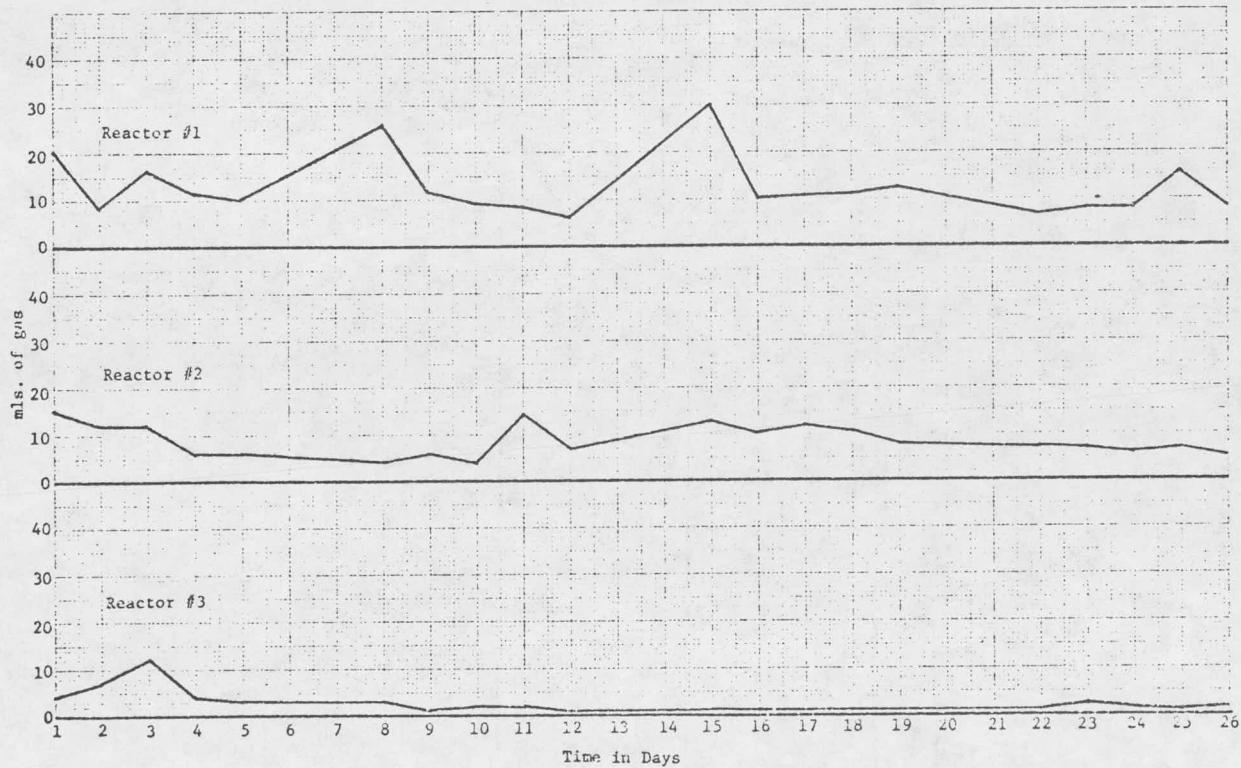


Figure 1. Gas production vs. time for three fermenters.

reactor, No. 1, containing synthetic growth nutrients. The second curves is of the first SSL fermenter and gas production from this fermenter is obtained with a residence time of about 3 hours. The third curve is a fermenter running on the effluent from the second SSL fermenter. The use of this second fermenter in

series is to assess the degree of conversion obtained in the first fermenter. By simple mathematics one can calculate gas production at a second and longer residence time than would be possible if only one fermenter were used. Very little methane is being produced in the third reactor indicating almost total conversion of available substrates to methane in the first 3 days in fermenter number two. The gas produced from fermenters one and two is high in methane content and easily supports combustion. Analysis of the produced gas using a gas chromatograph is shown in Figure 2. Fermenters one and two produce a gas

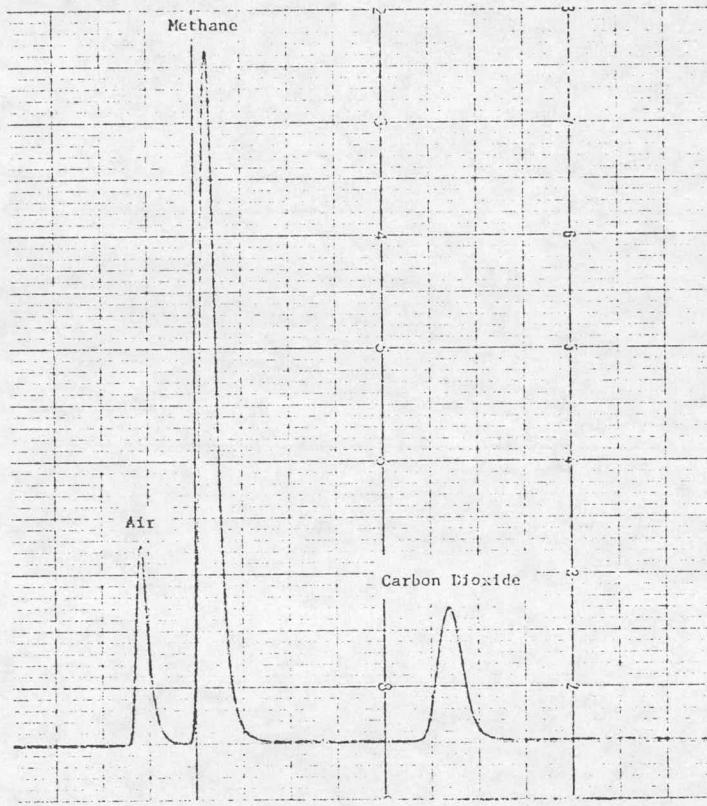


Figure 2. Gas chromatographic analysis of effluent gas from fermenter 2.

having a CH_4 to CO_2 ratio of 4:1. As yet, for some unexplained reason the gas produced in reactor number three contained some diluent other than CO_2 . The apparent methane to CO_2 ratio is still approximately 4:1, however, the concentration of

these two gases is only about 20%. The unknown diluent is not nitrogen, hydrogen, oxygen, or carbon monoxide. Additional effort is being expended to identifying the diluent and determining its origin.

Preliminary protein studies were encouraging. The pulp mill waste product, SSL, will not support the growth of yeast and has a comparatively low BOD. Samples of the ozonated SSL were adjusted to pH 4.5 and 0.1% potassium phosphate added to accommodate the metabolic requirements of yeast. Preliminary studies indicate that yeast will grow on this substrate. They appeared to increase in cell density during a period of one week. This is extremely encouraging and a major effort will be made during the next quarter to exploit this development.

SUMMARY AND CONCLUSIONS

Experiments in this quarter demonstrated the technical feasibility of producing protein and methane from pulp mill waste products. The conceptual process is operational in the laboratory and the progress to date will allow the program to proceed toward optimization of the critical process variables. The most significant contribution this quarter was the demonstration that ozonation will breakdown lignosulfonates into bioassimilable fractions. While the ozone treated SSL was only studied with respect to the production of methane, yeast and CO₂ there is a strong possibility that it would also be a useful substrate for a variety of other valuable fermentations.

PLANS FOR THE FUTURE

During the coming quarter, fermentation experiments will be performed to optimize the methane and yeast fermentation with the respect to ultimate yields and maximum production rates. Although the cultures currently being utilized have the required biological capabilities additional efforts will be directed toward

isolating other species or mutants which have improved characteristics. An effort will also be made to collaborate with other laboratories studying the process of methane biosynthesis in order to take advantage of any new techniques developed in related studies.