U. S. ARMY NATICK LABORATORIES
NATICK, MASSACHUSETTS

IN REPLY REFER TO:

SURVEY OF ALGAE STUDIES UNDER UNCONVENTIONAL FOOD RESEARCH

The work in the development of unconventional foods has been made necessary by the consideration of siege situations and development of closed ecological systems. The algae studies to date made on Chlorella pyrenoidosa and Chlorella 71105 indicate that algae fulfill several important requirements which include a source of high protein food, a means of converting urea to food, and a method of photosynthetic gas exchange for use in closed systems.

A typical analysis of Chlorella 71105 is as follows (dry weight basis):

Protein	55%
Fat	10%
Ash	5%
Crude Fiber	2%
Carbohydrate	28% by difference.

As harvested by centrifugation the moisture content of algae ranges between 67 and 73 percent, 70 percent moisture being a good mean value. Probably only one-third of the total carbohydrate is available; the remaining two-thirds include algenic acid and cellulose. Algae contain vitamins A, D, E, and K and are deficient in the B vitamins. The mineral content of the Chlorella 71105 grown on T-4 medium is as follows:

Prepared by Robert O. Matthern, May 1962.

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Element	% element of dry weight algae
N	9.81
P	1.30
K	1.09
Na	Trace
Mg	0.524
Fe	0.0314
S	0.935
Cu	0.01078
Cl	0.146
Ca	0.0103

From the above data it may be seen that algae can serve as an important source of protein, vitamins and minerals.

The various fields of research and development that are necessary before algae can be utilized as a satisfactory food supplement may be divided as follows:

1. Culture techniques;
2. Wholesomeness studies;
3. Processing for acceptability;
4. Food extension; and
5. New strains.

Culture techniques

Several culture procedures are available for culturing algae: open tank batch systems illuminated by fluorescent lights developed by Boeing and the Food and Container Institute; a continuous culture system using high intensity lights developed by Electric Boat; a continuous culture system using high intensity lights and mixing developed by the Food and Container Institute; and the culture of algae in complete

darkness by respiration or fermentation of carbonaceous material.

A 100-liter tank, batch process, has been continuously operated for 139 days with the supernatant from harvested algae recycled. The minerals consumed to develop plant tissue were replenished after each harvest. As no significant decrease in daily production occurred during the 139 days, it appears that viable cells do not produce any growth inhibiting metabolic products. Algae yields reached 35 gm per day using the T-4 medium, Table 1. A twenty percent increase in daily yield was experience when the sodium chloride was deleted from the T-4 medium. A further twenty percent increase in daily yield was attained by the addition of vigorous mixing.

The development of a steady state completely mixed system together with the application of a new concept for nutrient feeding has resulted in significant algae production. The use of this concept for nutrient feed to continuous systems has resulted in algae production as high as 36.5 gm dry weight per day from the 2.7-liter, high intensity, four-lamp system (52,000 foot candles) when carrying the concentration of algae at 8.98 gm per liter. At lower feed rates concentrations as high as 23.78 gm per liter have been attained. The nutrient feed is formulated considering the T-4 medium as containing the minimum concentration of nutrients necessary for growth plus 8.5 gm of cell tissue under high light intensity. To this additional nutrients are added in proportion to those contained in the algae tissue harvested. The steady state completely mixed four lamp culture chamber is shown in Figure 1.

Wholesomeness studies

With any new food, wholesomeness is a concern. With the Chlorella 71105 strain animal and human feeding experiments indicate no toxicity. A report that mice fed on algae were light-sensitive and actually died when placed under strong lights, needs more extensive study. The death of the mice may have been due to a vitamin deficiency rather than a sensitivity due to a toxicity developed from eating algae. Human feeding experiments conducted by Col. Marion E. McDowell at the U. S. Army Medical Research and Nutrition Laboratory indicate that up to 100 gm per day dry weight is acceptable. The best information available to date indicates that toxic species of algae are limited to a few species of the blue-greens.

S. S. Wilks at the U. S. Air Force School of Aviation Medicine has conclusively demonstrated that carbon monoxide is produced by illuminated cultures of the blue-green algae Anacystis nidulans when grown in the presence of oxygen. This also being true of higher green plants, it may be anticipated to occur with all algae species.

Some concern has been expressed by medical men on the effect on humans of the continual consumption of high concentrations of chlorophyll.

Processing for acceptability

The acceptability of algae as food will depend on three factors: chlorophyll removal, bitterness removal, and destruction of the cell wall. The chlorophyll may be and has been removed by methanol extraction, to a

degree by ethanol extraction, and by solarization of the algae. The removal of the bitter principle from algae may be accomplished to a degree by methanol extraction but is more completely done by ethanol extraction. Solarization of algae tends to decrease bitterness. The undesirable bitter aftertaste is most easily destroyed by preparing a roux of the algae in butter or oil. The destruction of the cell wall is essential for maximum utilization of the cellular protoplasm. Methanol extraction breaks the wall but does not destroy it. Studies, using a Morehouse colloidal mill to fracture the cell wall, have proved most successful.

A comparative taste test was performed on raw algae, completely bleached algae prepared by the methanol extraction and hydrogen peroxide process, and algae samples which had been treated for 1, 2, 4, 6, 18, and 24 hours in an ethanol Soxhlet extraction process. The ethanol extracted algae were found to be the least bitter, and the completely bleached algae were less bitter than the raw. The results from the test indicated that only one hour was necessary for optimum bitterness removal by the ethanol Soxhlet extraction process. It is thought that the bitter principle in algae is due to the long chain unsaturated fatty acids associated with the fats in algae.

Food extension

The development of algae food recipes has met with gratifying results. Algae food preparations include green noodles, algae date bars,

oatmeal cookies, algae fruit bars, three different algae soups, algae oatmeal cereal, and mashed potatoes with algae. The mashed potato algae mixture, containing 50 percent algae (dry weight), was most acceptable.

New Strains

Work is presently being carried on to isolate new algae strains which exhibit high growth rates. Of some 55 isolates found which grow at 35°C., at least 12 appear to have growth rates equal to or higher than Chlorella 71105.

Algae literature

Unfortunately, no one text, actually very few texts, contain information on the small unicellular strains of algae presently being considered as a food source for closed ecological systems. Information on the unicellular strains must be obtained from the literature contained in biological journals.

The following publications should serve as guides to work in algae culture:

1. Prescot. How to know the fresh-water algae. 1st edition, Dubuque, Iowa, Wm. C. Brown Co., 1954.

2. Dawson. How to know the seaweeds, 1st edition, Dubuque, Iowa, Wm. C. Brown Co., 1956.

3. Palmer, C. M. U. S. Department of Health, Education, and Welfare, Public Health Service, Publ. No. 657, 1959.

4. Ward, H. B., G. C. Whipple, and W. T. Edmondson. Fresh-water biology, 2nd edition, N.Y., John Wiley & Sons, 1959.

5. Burlew, J. S. Algal culture from laboratory to pilot plant, Carnegie Institution of Washington, Publ. 600, Washington, D.C., 1953.

The bibliographies in any of the above references will include the better biological journals.

School Projects

Many requests are received asking for information on algae for school science projects. Usually the student wishes to raise sufficient algae for animal feeding experiments or food for human consumption. The work and cost of producing photosynthetic cultured algae is enormous. Photosynthetically cultured algae costs over $150.00 per pound and requires equipment not usually available to students. Projects which measure the maximum and minimum limits of substrate concentrations or comparative growth rates would better serve science students and would be more within the limits of their capabilities and finances.

Table 1

T-4 Medium.

Source of Nutrient Element	Conc. of Stock Sol. gm/L	ml of Stock/L of Medium	gm/L of Medium
$(NH_2)_2CO$			0.4
KH_2PO_4			2.5
$MgSO_4 \cdot 7H_2O$			5.0
$CaCl_2 \cdot 2H_2O$			0.0294
NaCl			2.0
$FeCl_3 \cdot 6H_2O$	2.416	1	
Trace #1 Elements			
H_3BO_3	2.858	1	
$CuSO_4 \cdot 5H_2O$	0.079	1	
$MnCl_2 \cdot 4H_2O$	1.801	1	
$ZnSO_4 \cdot 7H_2O$	0.2200	1	
MoO_3	0.0190	1	
Trace #2 Solution containing		10	
$KCr(SO_4)_2 \cdot 12H_2O$	0.0098		
NH_4VO_3	0.0023		
$NaWO_4 \cdot 2H_2O$	0.00179		
$NiSO_4 \cdot 6H_2O$	0.00448		
$KTi(C_2O_4)_2 \cdot 2H_2O$	0.00740		
$Co(NO_3)_2 \cdot 6H_2O$	0.00495		
KOH	28.075		
EDTA	50		

Potassium hydroxide and acetic acid used for pH adjustment.

The main nutrient elements are in the same proportion as those recommended by Dean Burk to Electric Boat. However, a more complete trace nutrient formula than that recommended by Dean Burk has been used.

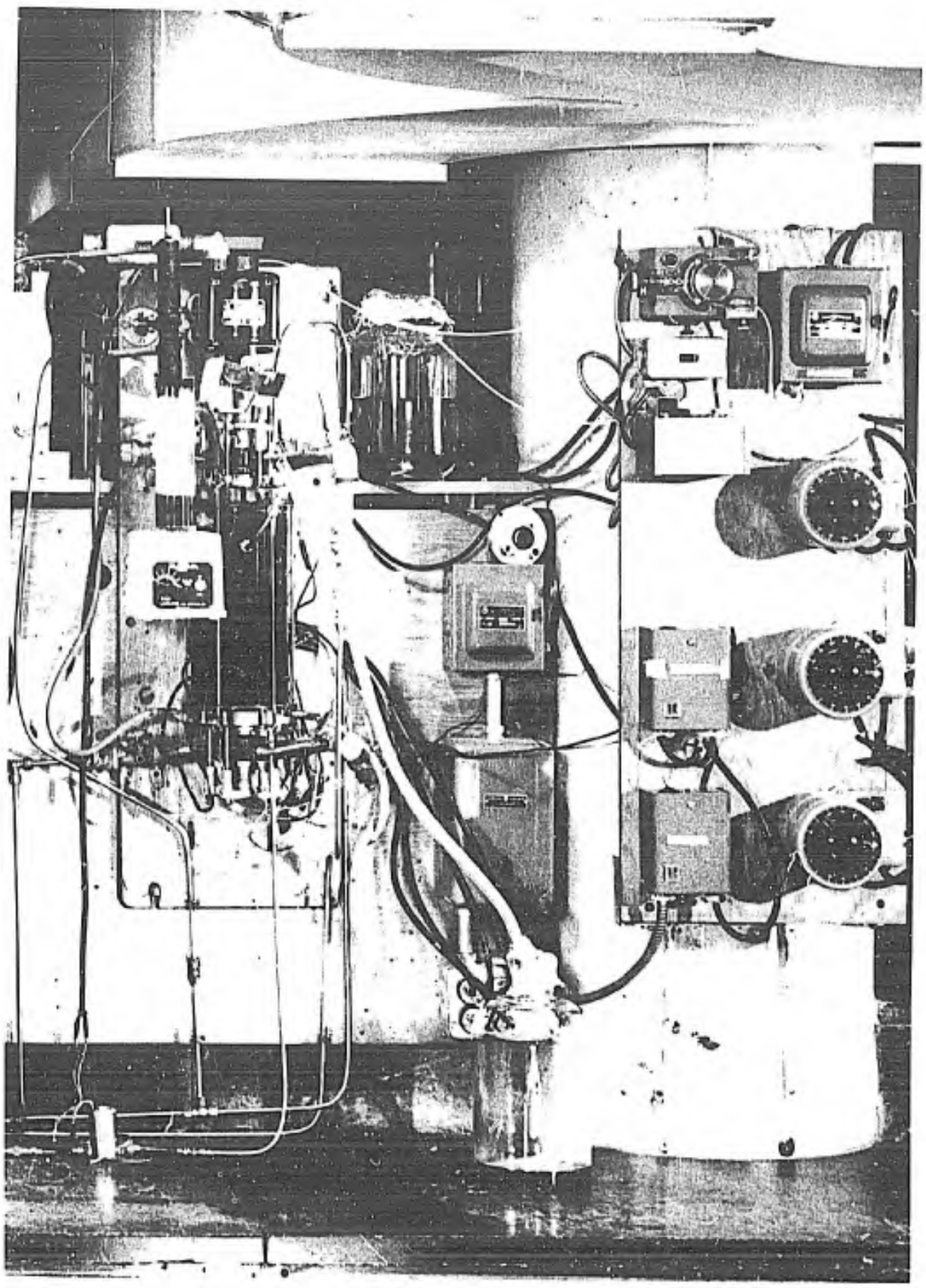
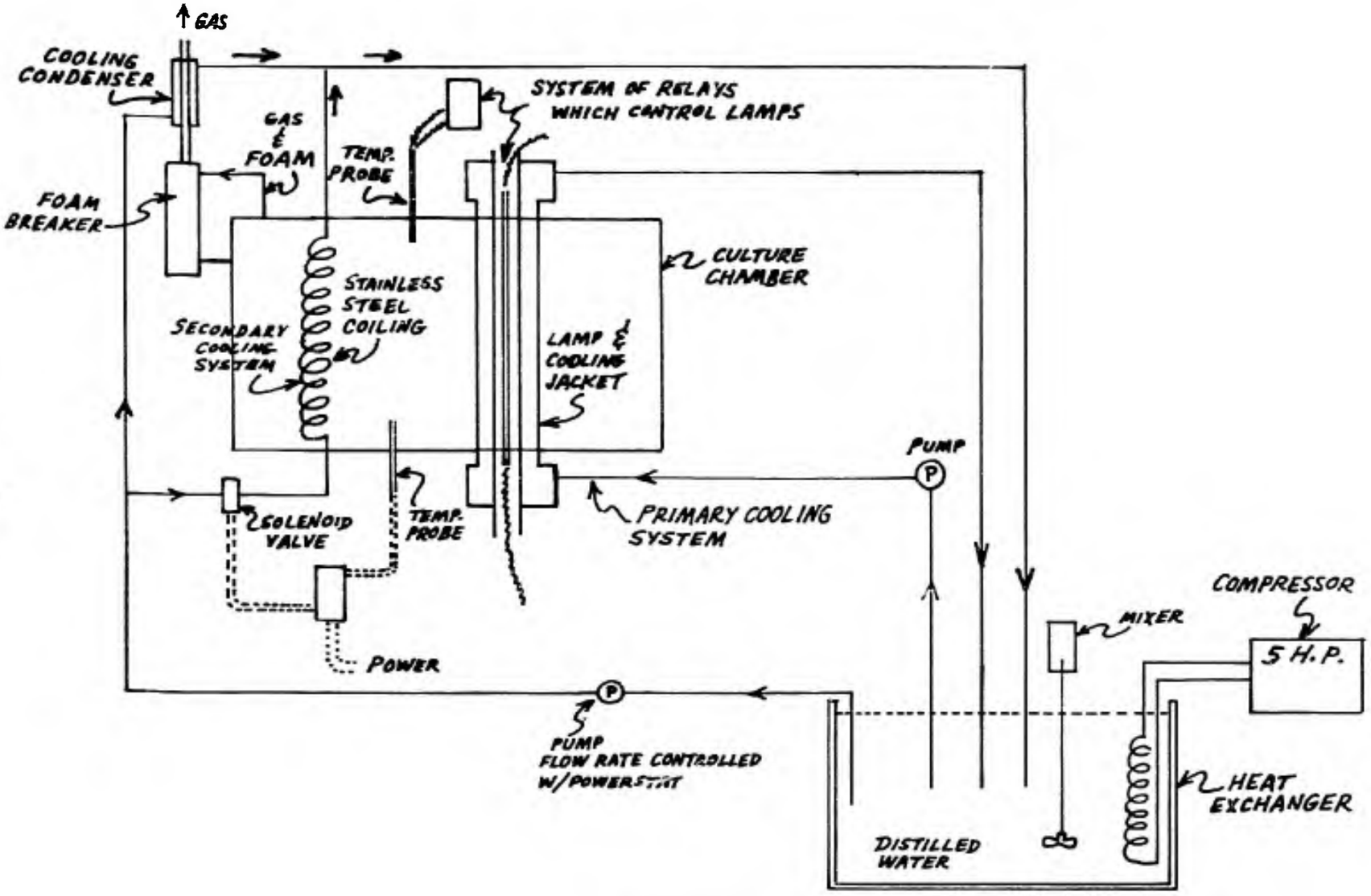


Figure 1. Steady State Continuous High Intensity Light Culture



SCHEMATIC OF COOLING

FIGURE 2

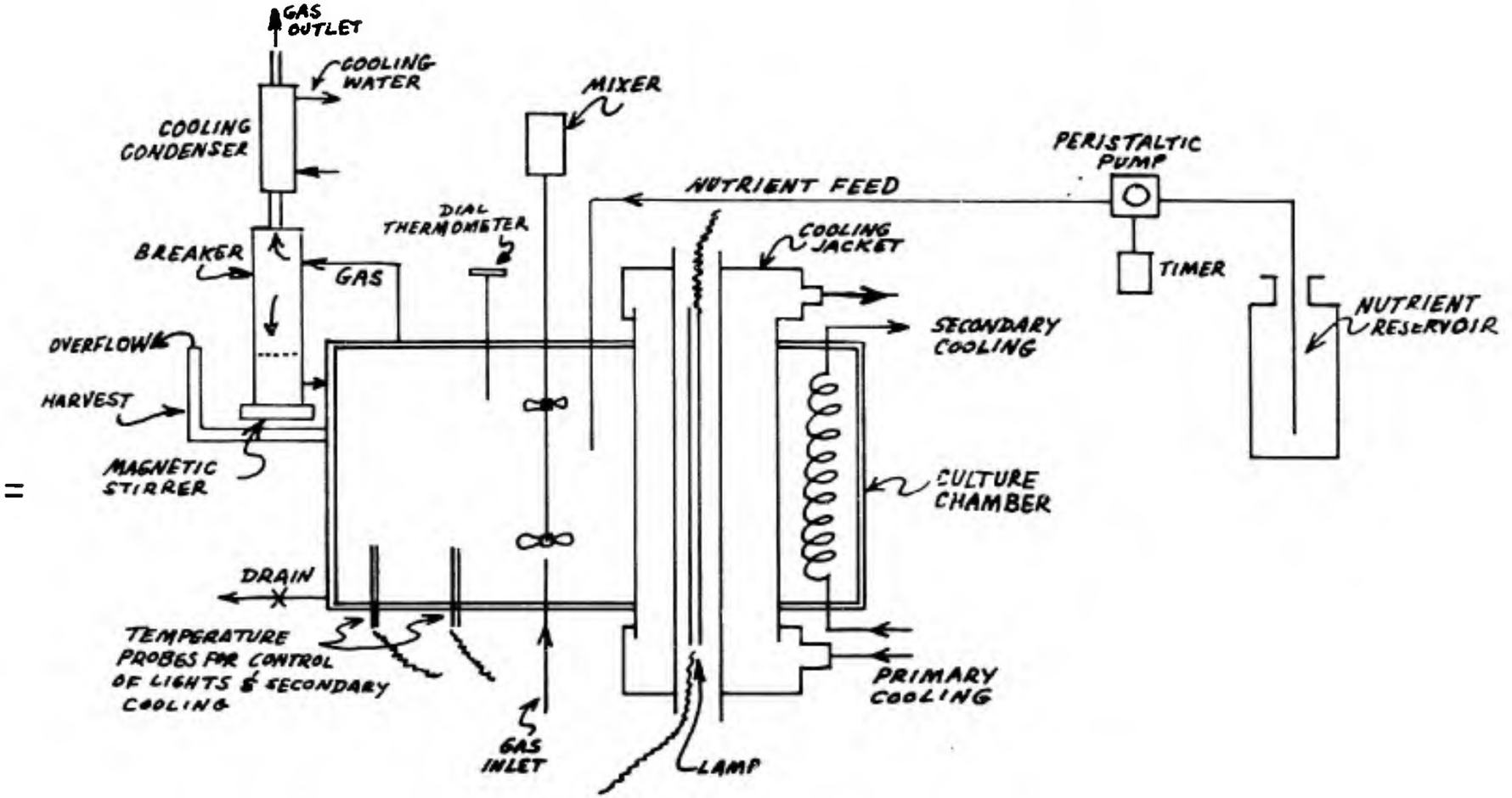


FIGURE 3
SCHEMATIC DIAGRAM OF
CULTURE CHAMBER