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NITROGEN TRANSFORMATIONS IN THE DECOMPOSITION OF SUGAR-CANE TRASH, WITH SPECIAL BEARING UPON PUERTO RICO SOIL PROBLEMS.*-**

JUAN AMÉDÉE BONNET, Research Chemist, Puerto Rico Insular Experiment Station.

INTRODUCTION

Sugar cane is the most important crop of Puerto Rico. As the area for sugar cane cultivation is limited, it has been necessary to increase the yields of tonnage and sucrose content in the sugar cane with the lowest possible cost. The 1910 production of the Island was 349,840 short tons of sugar. In that year, the Puerto Rico Sugar Growers' Association established an Experiment Station at Río Piedras, Puerto Rico, which in 1914 passed under government control. In its twenty years of active work attention has been given to various phases of tropical agriculture. Experimentation based on modern principles of soil science management has been applied to the cultivation of the sugar cane.

From 1911 to 1920, inclusive, the average production of the island was 414,426 short tons of sugar. During the last ten years the total production in short tons of sugar has been as follows:

1921	489, 817	1926	606, 464
1922	408, 325	1927	630, 202
1923	379, 171	1928	751, 332
1924	447, 587	1929	586, 760 *
1925	660, 532	1930	872, 326

The area under sugar-cane cultivation from 1915 to 1924 was between 203,105 to 256,431 acres. During the last years the area under cultivation has been, more or less, 242,000 acres. The 1930

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^{*} Decrease due to hurricane of September 13, 1928.

crop showed an increase of 93.51 per cent over the 1924 crop. The application of soil science principles have been successful in saving many a sugar-cane grower from dreadful bankruptcy. New sugarcane varieties, scientific applications of fertilizers, modern methods of field cultivation, and the proper use of machinery, irrigation, and drainage have been among the factors which contributed to the increase in the sugar-cane production of Puerto Rico to its present status.

THE ORGANIC MATTER PROBLEM

In the 1927–1928 report of the Insular Experiment Station of Puerto Rico, the present director, R. Fernández García says: (8)

"It is our belief, as has been pointed out in previous reports, that the apparent degeneration of cane varieties as shown by their susceptibility to diseases and pests and their ever-decreasing yields is not a degeneration of the varieties but is due to unfavorable changes in the soil conditions brought about by our present agricultural practices.

"Our inability to return to the soil, under present practices, the organic matter that is lost by tillage must necessarily not only lower the organic matter content of the soil but its quality and rate of decomposition. This continued loss of organic matter results in very unfavorable conditions for the plant and new varieties have to be imported or bred that will tolerate such conditions."

The organic matter of a soil is supplied by a great mass of substances of plant, animal, and microbiological origin. It is dynamic in nature and has a complex, variable composition. Its decomposition in the soil is mainly biological and greatly improves the chemical and physical properties of the soil.

Organic matter decomposition supplies the soil solution with carbon dioxide. The carbonated water renders soil nutrients soluble which can thus be taken by the higher plants. Organic matter improves the physical properties of the soil, such as, moisture-holding capacity, buffer effect, mineral absorption, temperature, etc., and introduces certain rare elements usually not brought in the organic fertilizers that may have some stimulating action on plant growth.

The additions of inorganic fertilizers to the soil supplies nutrients for the needs of the sugar-cane plant, but does not replenish the losses of organic matter. It requires a great outlay of money to supply organic matter to the soils with applications of organic commercial fertilizers. It has been economically advisable in many occasions to use green manure crops as a source of organic matter. It has been found also advisable to incorporate to the soil the straw which is left as waste in the wheat and sugar-cane fields.

The nitrogen added to the soil in the form of organic fertilizers

or organic residues is not present in an available form. It is this property which makes them frequently very valuable, especially, in wet districts. The nitrogen is not readily washed out and is gradually made available to the plant in the course of time. The addition of organic matter furnishes certain organic compounds which are used for structural and energy purposes by the non-symbiotic nitrogen fixing organisms. Leguminous plants serve as a host for the symbiotic nitrogen fixing organisms.

The physical condition of the heavy clay soils which are so common in the sugar-cane fields are greatly improved by the addition of undecomposed organic matter. The carbon dioxide produced in the decomposition processes makes the heavy soils more porous. It is more advisable to apply organic matter after it has been partly decomposed, to sandy soils. Under these conditions the coarser particles will be cemented by the colloidal "humus".

There is sufficient evidence to justify the belief that another important use of the organic matter is to supply during its decomposition certain compounds partially synthesized for the use of the plant. Knudson (10) showed in Cornell that corn absorbs and assimilates glucose, fructose, saccharose and maltose. It is not illogical to expect similar results from the use of amino acids and other products of protein decomposition.

The conservation of soil organic matter must be studied in relation to the plant, the soil and its microbiological population.

Some workers have associated the phenomenon of organic matter decomposition in soils with that of "denitrification". When such a nitrate reduction process goes to completion, nitrogen is lost in a gaseous form, either, as atmospheric nitrogen or as oxides of nitrogen. This concept made many farmers regard the addition of organic matter to their soils as an undesirable process.

Denitrifying organisms are active largely under anaerobic condi-It is favored by excess of water and organic matter. tions. When a solution of certain nitrate salts is inoculated with denitrifying organisms losses of nitrogen are observed. Such a phenomenon occurs in the soil in an entirely different way. When a soil is not too moist there could be partial reduction of nitrate salts to nitrite and ammonium salts without the reduction going further to gaseous nitrogen. As soon as the soil conditions favor the activity of the nitrifying organisms the ammonium salts will be reoxidized to nitrite and nitrate salts. The conclusion may be reached that the phenomenon of deni trification is of no economic significance in well aerated, not too moist soils, in the presence of moderate amounts of organic matter.

Under normal field conditions, usually, the nitrogen is transformed to less available organic forms by the microorganisms concerned with the decomposition processes of organic matter which need available nitrogen for building their cell protoplasm. In course of time the nitrogen is made again available. In the process of cell building the microorganisms compete with the plant for the available nitrogen. The carbon-nitrogen ratio of the organic material under decomposition is of special importance in this connection. It has been shown that there exists a more or less constant ratio between the nitrogen and carbon content of the soil, whatever the ratio was between these elements in the organic matter originally added to the soil. This ratio varies from 1:8 to 1:12, i. e., for every part of nitrogen there exists in the soil eight to twelve parts of carbon; the average ratio is about 1:10.

There is no appreciable effect upon the soil nitrate if the nitrogen of the organic matter added is about 1.8 per cent. (18.) A higher nitrogen content allows a rapid accumulation of ammonia and nitrate in the soil. Such is the case when dried blood, cotton seed meal, urea of other organic material with a high nitrogen content, and therefore, with a narrow nitrogen-carbon ratio is added to a normal soil. When the nitrogen-carbon ratio of the soil is wider than normal, plant growth suffers and nitrogen starvation will be observed as long as the excess of carbon lasts, since the microorganisms using the carbon as a source of energy will assimilate every trace of nitrogen that would otherwise be utilized by the higher plants.

As straw and sugar-cane trash have nitrogen-carbon ratios as wide as 1:80 it would not be advisable to incorporate such materials to a nitrogen starving soil unless nitrogen is supplied. If the sugarcane trash is allowed to decompose a few weeks previous to planting, nitrogen starvation of the plant is not so noticeable.

Soil microorganisms may obtain their nitrogen from protein and their degradation products or simple inorganic nitrogenous compounds, including the ammonium salts and nitrates. Some organisms, especially the heterotrophic bacteria, prefer and many even require complex proteins, albumoses or peptones as a source of nitrogen (and energy), while other microorganisms, especially the fungi and autotrophic bacteria, will thrive just as well and sometimes even better upon simple compounds of nitrogen. The minerals, chiefly phosphates and potassium salts, but also iron, magnesium, sulphur, calcium and traces of other elements, are utilized by all microorganisms either in the form of simple inorganic compounds or are obtained from complex organic substances in the process of their decomposition. The minerals may often be obtained from insoluble inorganic materials, especially if the organism produces acids which tend to make them soluble.

Temperature and moisture conditions in the tropics accelerate organic matter decomposition because they favor the activities of a group of soil microorganisms (heterotrophic) capable of breaking organic substances to use the carbon and free energy for their metabolic needs. The action of such microorganisms may be direct or through the agencies of enzymes. The protein nature of the microorganism protoplasm furnishes sources of proteins further decomposed by fungi and bacteria through polypeptides, amino acids and ammonium salts. The oxidation of the ammonium through the nitrite and nitrate stages furnishes energy for the activities of the nitrifying organisms. The sugars liberated in organic matter decomposition furnish energy for the non-symbiotic nitrogen fixers.

As organic matter is essential for microbiological life it is necessary to supply it to the soil, at intervals. If this is not done, a great part of the soil organic matter will be wasted as carbon dioxide into the atmosphere.

SUGAR CANE TRASH

The great bulk of the organic matter in the sugar cane fields is furnished by the leaves, leaf sheaths and tops of the sugar cane plant, which, in the form of trash are left in the field. Most of the green tops are used as fodder for farm animals. Dead roots contribute a small part. Maxwell (13) reports in 1898 an average of 33 per cent of total trash per ton of cane per acre for the Hawaiian crop. The weight of roots was considered to be very small, namely, about 4 per cent. (14.)

The bulkiness of the trash interferes with the crop cultivation. The most rapid method of getting rid of it is by burning. Sometimes this is done as a means to cut down field expenses. The trash is burnt either before or after cutting the crop. Claim is made that fire does not injure the following crop in its germination power and that injurious insects are destroyed.

Burning off destroys the borer parasite, but not always the borer. (13.) The borer is usually sheltered in the rotten cane left on the land which protects the insect from the fire effects. In 1916, Wolcott (30) reported that burning off increases borer infection 100 per cent.

Studies have been made on the effect of fire on tonnage and sucrose yield when the harvest is delayed for a few days. López Domínguez (11) in Puerto Rico experimenting with Cristalina and P.R.-209 canes found that when subjected to fire while standing in the fields, or if stored, the canes lost about 2 per cent of their original weight per day. The loss in weight caused by delayed milling was greater in the burned canes than in the unburned ones. The burned cane suffered inversion both when cut and when left standing in the field, but the standing cane suffered greater losses in sucrose than the cut cane. At the Ewa Plantation, (20) Hawaii, H-109 harvested five, ten, fifteen days after burning, lost respectively; 14.72 per cent, 29.7 per cent and 20.29 per cent, of the original weight. Burned cane cut at once and burned cane allowed to stand until milled differed little in sugar losses. Alineastre (3) says:

"Cane burning before cutting permits loss of sap and decomposition by microorganisms. Losses occur after 24 hours which offset the decreased harvesting cost."

O'Brien (16) also condemns this practice.

In Puerto Rico, a few plantations burn the trash when harvesting the ratoons. Most manage to handle the trash during the cultivation of the ratoons; but when the land is to be prepared for a new crop the field is cleaned by means of fire. In the irrigated fields of a certain large plantation the trash is burned. In their non-irrigated fields the usual custom for fall-planting is to leave the trash on the ground while preparing the field, raking it to one side and spreading it over after the planting is made. With spring planting, they always burn the trash. When there is too much straw while the ratoons are under cultivation it is the usual custom to bury part of the straw in the rows.

In the low lying lands of some plantations located at the western section of Puerto Rico, large amounts of trash are moved about by the floods and deposited in heaps. It is expensive to handle the piles of mud and trash. So; in these fields, trash is usually burned.

The following comments on sugar cane trash are taken from the report (12) presented by the Puerto Rico delegates on the 1927 International Meeting of Cane Technologists held at Havana:

Taggard, Louisiana:

"As trash buried at a great depth does not decompose, beneficial effects such as: addition of organic matter, improvement of soil physical conditions and addition of inorganic nutrients to the soil, are not obtained. For this reason in Louisiana the trash is slightly covered with soil."

Moir, Hawaii:

"The practice in Hawaii is to burn the trash. So far no evil effects havebeen observed in the soil." Agee, Hawaii:

"The roots that remain in the soil are sufficient to maintain the organic matter equilibrium in the soil."

Menéndez Ramos, Cuba:

"The most beneficial effect from the trash is the preservation of soil moisture and checking of weeds growth. The mulch effect is of more value than the fertilizer nutrients added to the soil."

It was also reported:

"In Cuba, the yields per acre were higher in certain fields where the trash was burned. This was attributed to the fact that no cultivation was given to the fields where the trash was left; while those where the trash was burned were carefully cultivated. Borer infection was higher where the trash was burned."

Storey, South Africa:

"Experiments were made with Adco and trash, to convert the latter into humus. The trash must be left moist. Trash was converted into humus in three months during the summer."

López Domínguez and del Valle (12) summarize those statements as follows:

"The results in Cuba indicate that it would have been better to cultivate the soil and line the trash. As to the non-evil effects observed in Hawaii from barning the trash, it must be considered that the Hawaiian soils are very porous, and that cane in Hawaii has been an important commercial crop for the last twenty-five years only. In Louisiana, where cane is grown since the 17th. century in alluvial soils heavier than those of Hawaii and where field methods have been carefully studied, it is not burned. In Puerto Rico with its heavy soils under cultivation for several generations and where the borer exists, the trash should not be burned. Means must be studied to handle the trash in the most economical way which at the same time is the most beneficial to the fields. The questions to be answered are: Is the trash to be buried or left over the surface? Must the trash be lined or spread over the whole surface?"

The increase in sugar cane yield in a field where trash is burned as has been observed in Cuba and Hawaii may be due to a partial sterilization effect whereby the nutrients are rendered more available for the use of the crop. It is a temporary effect which may last till the soil exhausts its natural supplies. Part of the available minerals accumulating on the porous soils of Hawaii after the trash is burned may be lost through leaching.

120 THE JOURNAL OF THE DEPARTMENT OF AGRICULTURE OF P. R.

Agee's comment was reported in 1924 by Mc. George (15) as follows:

"To determine if the current practice of burning the trash in the Hawaiian islands, was depleting the organic carbon of plantation soils, 42 samples of Ewa soils representing cultivated and uncultivated portions of nine fields, were analyzed for total carbon content (little or no carbonate carbon being present in coral soils.) In these fields the organic content in four was lower, and in five higher, in the cultivated than in the uncultivated portions. The variations were, for cultivated fields, from 0.85 to 1.93 per cent with an average of 1.30 per cent and, for the uncultivated, from 0.74 to 2.60 per cent with an average or 1.37 per cent. In these fields the trash is always burnt, and there appears to be no indication that there is any depletion of the organic carbon content, the roots and stubble presumably being sufficient for keeping it up."

Such results, however, could not be considered as significant unless proved to be so, by statistical methods. De Turk (7) says:

"The variability of field soils makes the problem of determining increases in organic matter brought about by a treatment a difficult one, requiring great eare in selecting similar pairs of plots for comparison. It was assumed that composites made up of twelve borings would give samples sufficiently representative of the soil of the plots. The probable error for 12 borings was 0.76 per cent of the mean or 50 to 450 pounds of organic carbon per acre."

The results obtained with the Adeo treatment may also be obtained if the proper compost is prepared by adding a mixture of fertilizer salts to the trash. Adeo, is a mixture of fertilizer salts that serve as a supply of nutrients for microorganisms. The compost is prepared by adding 150 pounds of Adeo to a ton of straw. Sufficient water is added to make about three tons of manure. The question to be settled is whether or not the Adeo treatment is economical. In the preparation of an adequate manure Albrecht (2) recommends:

"A mixture consisting of 45 pounds of ammonium sulphate, 15 pounds of acid phosphate, and 40 pounds of finely ground limestone at the rate of 150 pounds per ton of straw, will, with moisture, convert straw into a brown product having all properties of manures."

According to Noel Deerr: (6)

"It is the custom in Hawaii, Demerara, and Java to burn the trash. In Mauritius, much of the trash is used as bedding for the plantation stock and thus finds its way back to the soil as pen manure. A similar routine is followed in the British West Indies. In Cuba it is the almost invariable custom to let trash rot on the fields, where it remains as a blanket. To this custom the long continued fertility of the Cuban cane lands is to be attributed.

"During the period 1901-13 extensive experiments were made on a Hawaiian plantation, in all 109,990 tons of trash being buried."

The effect of this procedure is thus described: (6)

"Where two rations were formerly the maximum, four are now becoming the rule. The yields, instead of decreasing with each subsequent ration, have inereased. The 1908 crop was the first one to have trash left over its entire ration area. That and the succeeding crops show an average yield of 4.102 tons of sugar per acre; the seven preceding crops gave 3.329 tons of sugar per acre. The 1914 crop to date has yielded 5.2 tons per acre and is expected to go still higher. While all the credit cannot be given to trash, there is no doubt whatsoever that leaving the trash has been the principal factor."

The actual operations in Hawaii followed on a rainfall plantation are described by Larsen: (6)

"After the cane is cut the trash is hoed away from the stools into the furrow. This work requires about two men per acre per day and is called 'palipali-ing'. This is followed by off-barring, which consists of ploughing off or away from the stools. The soil by this operation is thrown against and partly over the trash and assists materially in hastening its decay. A ten, twelve or fourteen-inch plough is used for off-barring. A revolving knife or sharp coulter is attached to the ploughbeam to make a clean cut ahead of the plough. One man with two mules can off-bar 2 to 21/2 acres per day. After off-barring, hoeing is done in the cane lines. In the furrow, that is, between the lines of cane, the weeds in most cases are kept down effectively by the trash. Cultivation between the rows begins from one to two months after pali-pali-ing. After two or three more hoeings in the cane rows as occasion demands and as many more cultivations the trash will have become so thoroughly broken up and disintegrated that the furrow can be small-ploughed without trouble. A small eight-inch plough is run usually four times through the furrow to loosen up the soil and to mix in the trash. After small-ploughing, the cane is hilled. This is done with hoes, ploughs, double mould-boards, or discs. With this operation the rotted and partly rotted trash is thrown toward the cane and is more thoroughly buried and mixed with the soil."

In certain soils in Demerara the presence of decaying trash has, according to Harrison (6) a specific function in neutralizing the effect of the large quantity of alkaline soil water there present. On this point he writes:

"In experiments in which (a) soil water was allowed to evaporate into the air and (b) caused to evaporate in an atmosphere consisting almost entirely of free carbon dioxide it was observed that when the evaporation takes place in air, nearly free from carbon-dioxide gas, practically the whole of the lime salts are deposited as calcium carbonate, while the water is being concentrated to onethird of its original bulk, and the remaining water becomes a saline one, containing large quantities of magnesium salts as chlorides, sulphates and carbonates in solution. The calcium salts, which are known to exercise a profound influence in reducing the highly toxic action of the magnesium chloride and carbonate on plants, are almost wholly removed from solution and the soil water becomes in a condition which is poisonous to vegetation; this is probably what takes place during prolonged periods of dry weather or more or less worn-out cane soils, in which by injudicious cultivation and especially by long-continued destruction of the trash by burning, the normal proportion of organic matter has been largely reduced. When, on the other hand, the evaporation takes place in an atmosphere heavily charged with carbon dioxide, as in the air present in soils containing the proportion of organic matter normal to good soils, the calcium salts remain for a long time in solution until the liquid commences to become a saturated brine, and this for a prolonged period continues to modify the toxic action of the magnesium salts. It is possible on such land that the soil water during drought may become concentrated in the upper layers of the soils, without any material injury to the plant, until by concentration of the soil water the toxic action of the magnesium salts exerts itself.''

Cross, (5) Director of the Tucuman Experiment Station, recommends the following procedure:

"It is practically feasible to leave the trash without burning, in every second middle, cultivating the other one, and every year to alternate the middle so treated. The soil benefits from the nitrogen and organic substances and the sugar yields are increased. The procedure is particularly recommended for plantations invaded by weeds."

In 1914, in Hawaii, special rakes were prepared to handle the trash. (1.) Mr. F. E. Hance, acting chemist of the Hawaiian Sugar Planters Experiment Station, reports in a letter to the writer dated March 12, 1930:

"In Hawaii, no special implements are in use for handling the trash. The custom is to ripen the cane and burn the trash. In the winter time and at all times in some plantations trash remaining on the ground is ploughed under, if the field is to be replanted."

Klinge (9) reports:

"Trash is burned in Hawaii as well as in Peru with the difference that in Hawaii the burning is done in situ and in Peru is done in the rows where the trash is piled after raking it from the field." After cutting the cane in some irrigated fields of Hawaii, the trash is raked towards every other furrow. The intermediate furrow is used for irrigation purposes. The practice of burning the trash followed by those plantations that only depend on rain water for cane growth is very variable. Some plantations burn the trash; while others leave it to rot in the soil. Complete burning cannot always be carried on when the continuous rains keep the trash moist. Under these conditions sufficient unburnt trash remains on the soil."

In Java (19) trash is either transported to the factory to be used as fuel or is burned in the field as a means to clean the soil for the rice crop. It is the custom to get two successive rice crops after the sugar cane crop. Part of the rice straw is incorporated to the soil. The cane is never burned before cutting; special precautions are in fact taken against accidental fires by removing trash at least thirty feet away from nearby roads. Owen (17) reported in 1926 on work done at the Louisiana Experiment Station:

"Adding undecomposed trash to the soil, resulted in a rapid loss of nitrates, but this depressing action upon nitrates disappeared rapidly with the trash decomposition. The addition of fresh trash caused a marked increase in the soil fungi and increased the ratio of fungi to total number of microorganisms. The depressing effect of trash upon the soil nitrates has been traced to the starch and pentose content of the material."

Bonazzi (4), on studies at the Chaparra Experiment Station, Cuba, referred particularly to the influence of the incorporation of sugar cane trash on the moisture content of soil:

"Greatest retention of moisture is to be obtained by allowing the cane leaves to accumulate on the surface of the land. Incorporating the leaves in the soil through ploughing or cultivation does not justify the additional expense. Denitrification is strongly active in presence of cane leaves incorporated with soil. When nitrates were applied in the form of sodium nitrate the losses were very large, whereas in the soils receiving easily assimilable carbohydrates, but no nitrate, the formation of a small quantity of nitrates from the natural soil stores of nitrifiable substance was followed by its early disappearance. The same path was followed by the soils receiving their nitrogen in the organic form.

"The formation of nitrates from tankage in soils containing cane leaves was not cumulative but reached a maximum aften ten days, followed by a rapid disappearance during the second period of incubation. This experiment indicates that great care should be taken not to incorporate cane leaves and trash in the soil at a time of active nitrate formation in the soil. Such a practice would prove deleterious to the growing cane."

A series of plot experiments at the Insular Experiment Station, Puerto Rico, was arranged as follows: (8)

- (1) One and a-half tons air-dried chopped cane trash + 800 pounds (NH₄)₂SO₄ per acre.
- (2) Treatment (1) + 2 tons CaCO₃ per acre.
- (3) Check.
- (4) One and a-half tons of cane trash ± 2 tons CaCO₃ per acre.
- (5) One and a-half tons of cane trash per acre.

The land was kept free from vegetation. The effect of cane trash on soil nitrates was followed for a period of 23 weeks. It was found that the addition of trash alone lowered the level of nitrate concentration in the soil. The addition of sulphate of ammonia to the trash raised the level of nitrate concentration to a maximum of eighty pounds per acre over the check in 21 weeks. The pH of the soil was 7.44. In the first treatment, nitrification was slower at the start, but from the eighth week on surpassed that of the second treatment. In another series of plot experiments at the same Station, the equivalent of $1\frac{1}{2}$ tons of trash per acre was added to a clay soil and sown to soybeans. (8.) The gain in yield of this plot was 201 per cent \pm 33 per cent over the check. Plots with loam soil given a gain in yield over checks of only 160 per cent \pm 12 per cent. The same plots planted to BH-10(12) after incorporating the soybeans did not show such striking difference in yield.

Chemical studies of organic matter decomposition have been greatly based on determination of carbon, nitrogen, ash and loss by ignition. The loss in dry weight was also considered. Such analyses do not give information of what actually happens in the course of organic matter decomposition. To understand the decomposition of sugar cane trash in the soil it is important to gain information concerning the nature of the different groups that form the organic complex. The classification of the various chemical plant constituents into a series of definite groups as proposed by Waksman (27) is a great tool for the study of organic matter decomposition in the sugar cane fields. The various chemical plant constituents are divided into the following groups:

"1. Water-Soluble Constituents.—These include the most readily available nutrients, both for the growth of the plant and for the growth of microorganisms, when the plant is undergoing decomposition. These constituents comprise a number of organic and inorganic substances, such as the sugars, various glucosides, amino acids, and certain simple proteins among the former, while the latter includes nitrates, phosphates, sulphates, chlorides, potassium salts, etc. This group of plant constituents is highest when the plant is young, making up as much as 40 per cent of the dry matter of the total plant material. This percentage decreases with age, so that mature plants may contain only about 5 per cent of water-soluble constituents, these proportions depending of course also on the nature of the plant and available nutrients.

"2. Ether and Alcohol-Soluble Constituents.—They comprise the fats and oils, waxes and resins, tannins, terpenes, alkatoids, and various pigments. They make up only a small portion of the plant, usually 2 to 6 per cent.

"3. Celluloses.—These form in most plants the largest single group of constituents, ranging from 15 to 40 per cent of the dry weight of the plant material. They are polysaccharides which serves the function of protective substances in the plant and they are not hydrolyzed by dilute acids and alkalies. They are rapidly decomposed, however, under certain conditions by various specific bacteria and fungi.

"4. *Bemicelluloses.*—These and allied carbohydrates, not included in the sugars and in the celluloses, play a function of both reserve and protective substances in plants. They are also polysaccharides of the pentose or hexose group and are hydrolyzed by dilute acids. They make up 10 to 30 per cent of the plant constituents. Some of them are decomposed even more rapidly than the celluloses, while others are more resistant.

"5. Lignins.—Lignins are the so-called incrusting substances in plants. Both lignins and celluloses, which form in the plant complexes of a chemical or physical nature known as ligno-cellulose, are low in young plants and increase in proportion with the age of the plant, both in total quantity and in relation to the other plant constituents. The lignins, which make up 5 to 30 per cent of the dry plant material, are most resistant to decomposition in the soil.

"6. Proteins.—These substances play an important function in the nutrition of the plant and in the decomposition of the plant residues in the soil, since they are largely the carriers of the important element nitrogen, as well as of some of the phosphorus and sulphur. In view of the fact that the nitrogen is liberated in the form of ammonia, as a result of the decomposition of the proteins, it was usually assumed that it is sufficient to measure the rapidity of ammonia accumulation from protein decomposition in the soil. However, we come to recognize now that, in the presence of celluloses and hemicelluloses, which are readily used as sources of energy by microorganisms, a part, if not all, of the nitrogen which is liberated in the decomposition of the proteins may be reassimilated by the soil microorganisms and changed into microbial cell substances. The nitrogen (or protein) content of plants is high at an early stage of growth and decreases with an increase in the maturity of the plants, frequently from 18 per cent protein in the young plants to about 1.2 to 1.5 per cent in the mature straw.

"7. Minerals.—These include phosphates, sulphates, chlorides, nitrates, and silicates of potassium, calcium, magnesium, iron, aluminum, etc., some of which are water-soluble and others insoluble. They form the ash content of the plant, although some are also present in the proteins (S, P). They make up from 1 to 12 per cent of the total plant constituents. They are high in young plants and diminish, in proportion to the other plant constituents, with maturity of the plants. The nature of the minerals in the young and old plants differs not only quantitatively but also qualitatively, the soluble minerals predominating in the younger plants and the insoluble in the other plants.

"In these seven groups of complexes we can account for practically 90 to 96 per cent of the plant constituents."

EXPERIMENTAL

COMPOSITION OF SUGAR-CANE TRASH

Analyses were made of two samples of trash which were used in subsequent experiments. The first sample was obtained from a field of POJ-2714, adjoining the United States Field Station at Canal Point, Florida.^{*} It was gathered from the soil surface around cane in the dry form and was further sundried before shipment. The second sample of sugar cane trash was taken at the Everglades Experiment Station, Florida,^{*} as dead leaves from the lower part of the stems of SC-12(4). Before shipping, it was thoroughly dried in the greenhouse. The samples were ground in a No. 1 Wiley mill and sifted through a 1mm. sieve.

^{*} Thanks are extended to Dr. B. A. Bourne, Canal Point, and Dr. R. V. Allison, Everglade Experiment Station for furnishing the samples of sugar-cane trash.

The approximate complete analysis of the sugar-cane trash was carried in duplicate as follows:

Moisture, total nitrogen and ash.

- Six-gram portions were analyzed as follows: (29)
 - (a) Extracted with ether in Soxhlets for 16 hours.
 - (b) Residue was extracted for 24 hours with 100 cc. of cold water.

After filtering, the residue was washed several times with cold water. The filtrate was made to volume and divided into three portions, one used for reducing sugar, one for total nitrogen after evaporating excess of water and one for total soluble organic matter by evaporating and drying to constant weight. Ash determinations were carried on after igniting the organic matter.

- (c) Residue from (b) was treated for 3 hours with 100 ec.
 of hot water over boiling water bath. The solution was analyzed as in (b).
- (d) Residue from (c) was treated twice with 100 cc. of 95 per cent alcohol.
- The flask was placed for 2 hours on a boiling-water bath. The filtrate and washings were evaporated to constant weight. The difference between the weight of the paper with the residue and that of the original paper gives the weight of the plant material from which the ether and water-soluble, as well as the alcohol-soluble portions have been removed.

The sum of the weight of fractions (a), (b), (c) and the weight of the plant substance left after the alcohol extraction was taken as the quantity of the original dry plant material taken for analysis. All calculations were based on such weight.

- (e) The residue from (d) was treated with 100 cc. of a 2 per cent solution of hydrochloric acid and autoclaved for five hours under flowing steam. The solution was filtered off through dried and weighed filter papers. The residue was washed with dilute acid and then with water until free from acid. The filtrate and washings were analyzed for reducing sugars by the Bertrand method, and for total nitrogen. The hemicellulose content was obtaine 1 multiplying the reducing sugars by 0.9.
- (f) The washed residue from the hydrochloric acid extraction was dried to constant weight. Two 1-gram portions of the dry material were treated with 10 cc. of 80 per cent sulphuric acid, for 2 hours, in the cold. The acid was brought in contact with all particles of the material by stirring. After 2 hours, 150 cc. of distilled water were added to each treatment and the contents were autoclaved for 1 hour at

15 pounds pressure. The contents were filtered through small dried and weighed filter papers. The residue was well washed with water to free from traces of sulphuric acid. The combined solution and filtrate was analyzed for reducing sugars. The cellulose content was obtained multiplying the reducing sugars by 0.9. Of the four residues for each original material, two were used for ash and two for nitrogen determinations. Weight of residue $-(ash + nitrogen \times 6.25) = lignin content.$ The cellulose and lignin found in the one gram portion of residue left after the 2 per cent HCl extraction was multiplied by the weight of this residue to obtain the cellulose and lignin content in the original 6 grams of material.

TABLE	I
1 40 005	r

Chemical Constituents	Sample No. 1 POJ-2714 per cent	Sample No. 2 S. C. 12-4 per cent
Ether soluble Fraction Cold Water Soluble Organic Matter Hot Water Soluble Organic Matter Alcohol Soluble Fraction. Hemicelluloses. Celluloses. Lignin (nitrogen & ash free). Water insoluble protein. Total Ash	$\begin{array}{c} 2.34\\ 4.42\\ 2.17\\ .56\\ 25.33\\ 29.71\\ 11.53\\ 2.00\\ 12.79\end{array}$	$\begin{array}{c} 1.85\\ 1.92\\ 1.56\\ 26.32\\ 32.85\\ 16.06\\ 2.25\\ 6.30\end{array}$
Totals	90.85	90.3

COMPOSITION OF OVEN DRIED SUGAR CANE TRASH

The total nitrogen contents by the Kjeldahl method were 0.59 per cent and 0.64 per cent for samples 1 and 2, respectively. The total carbon contained in the first sample was 40.3 per cent as determined by a wet method (23) using KMnO₄ as the oxidizing agent. The pentosan content of the same sample was 25.21 per cent as determined by furfural distillation with 12 per cent HCl and precipitation with phloroglueinol. Total carbon and pentosans were not determined on the second sample. There were no reducing sugars in the water extracts of either sample of trash.

DECOMPOSITION OF SUGAR-CANE TRASH

Trash sample No. 2 (SC-12-4) was used in this experiment. It was chopped in pieces about $\frac{1}{2}$ inch in size. Twenty grams portions in each of 12 one-liter flasks were inoculated with 1 cc. of soil infusion (10 grams of soil to 100 cc. of tap water). Enough water was added

128 THE JOURNAL OF THE DEPARTMENT OF AGRICULTURE OF P. R.

to give composts containing 66, 80 and 88 per cent of water. The treatments were as follows:

1 and 2_____66 per cent water 3 and 4____66 per cent water + 1 gm. (NH₄)₂SO₄ 5 and 6_____80 per cent water 7 and 8_____80 per cent water + 1 gm. (NH₄)₂SO₄ 9 and 10_____88 per cent water 11 and 12_____88 per cent water + 1 gm. (NH₄)₂SO₄

One set was incubated for thirty days at 28°C and a second set for sixty days before analyses were made. Tests for moisture, nitrates, ammonia and total nitrogen were made on the wet decomposed material. Nitrates were determined by the phenoldisulphonic acid method. Ammonia was extracted with normal KCl and determined by distillation with magnesium oxide. Total nitrogen was determined by the Kjeldahl method.

The analysis of the organic constituents was performed on 6-gram portions of the material after drying at 40°C. The procedure previously described was followed with one exception; no cold water treatment was given.

TABLE II

DRY WEIGHT LOSSES OF SUGAR CANE TRASH COMPOSTED FOR 30 AND 60 DAYS WITH OR WITHOUT ADDITIONAL INORGANIC NITROGEN

		66 per ce	nt Water	80 per ce	nt Water	88 per cent Water			
Days Origina Materia		No nitrogen added	Nitrogen added*	No nitrogen added	Nitrogen added*	No nitrogen added	Nitrogen added*		
	gm.	gm.	gm.	gm.	gm.	gm.	gm.		
30	20 20	$\begin{array}{c} 17.9\\ 16\ 4\end{array}$	14.9 14.9	$\begin{array}{c} 17.4\\ 16.4\end{array}$	$\begin{array}{c} 15.9\\ 13.6\end{array}$	$\begin{smallmatrix}&18.2\\&14&8\end{smallmatrix}$	14.8 13.2		

* Figures represent dry weight of residual material plus residual dry weight of (NH4) 2SO4 added.

The information given in terms of dry weight losses is of limited value unless we consider the fate of the different groups which constitute the sugar-cane trash complex.

TABLE III

66 per cent Water 80 per cent Water 88 per cent Water Original Chemical Constituents (1) Material No No No nitrogen Nitrogen nitrogen Nitrogen nitrogen Nitrogen added added added added added added Per cent Ether soluble fraction. 1.85 1,08 1.571.051.04 1.18 0.89 Hot and cold water soluble organic matter (2) 3.483.39 $7.48 \\ 2.29$ 4,14 6.682.945.591.37 1.55 Alcohol soluble fraction. 1.24 2.39 $1.45 \\ 23.91$ 2.34 Hemicelluloses 26.32 20.32 20.86 20.32..... 32.85 31.33 23.4930.90 23.26 32.29 22.65 Celluloses Lignin (nitrogen and ash íree). Water insoluble protein 21.19 18,33 16.0618.10 20.3119.3521.22 $2.25 \\ 0.00$ 3.067.44 3.56 5,943.81 9.44 0.75 Nitrogen as a mmonia.... 0.00 0.830.00 0,75 0.00

COMPOSITION OF SUGAR CANE TRASH COMPOSTED FOR 30 DAYS WITH OR WITHOUT ADDITIONAL INORGANIC NITROGEN

TABLE IV

9.29

93.02

7.40

92.14

9.64

91.89

7.12

90.74

9.16

92.53

6.86

89.36

COMPOSITION OF SUGAR CANE TRASH COMPOSTED FOR 60 DAYS WITH OR WITHOUT ADDITIONAL INORGANIC NITROGEN

		66 per ce	nt Water	80 per ce	nt Water	88 per cent Water			
Chemical Constituents (1)	Original Material	No nitrogen added	Nitrogen added*	No nitrogen added	Nitrogen added	No nitrogen added	Nitrogen added		
	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent		
Ether so'uble fraction Hot and cold water soluble	1.85	1.22	1.51	1,12	1.30	0.86	1,18		
organic matter (2)	3.48	3.50	7.30	3.48	6.69	3.88	6.57		
Alcohol soluble fraction,	1.24	1.02	1.88	1.02	1.80	1.14	1.63		
Hemicelluloses	26.32	24.35	19.33	24.24	18.78	22.29	19.39		
Celluloses	32.85	27.68	17.49	30.64	17.61	27.65	18.12		
free).	16,06	19.13	22.41	19.44	22.81	19.88	24.41		
Water insoluble protein	2.25	5.13	8.63	4.81	10,19	5,81	11.75		
Nitrogen as ammonia	0.00	0.00	0.70	0.00	0.79	0.00	0.77		
Total ash	6.30	8.52	9.78	7.76	10.42	8.24	10.94		
Totals	90.35	90.85	89.03	92,51	90.39	\$9,75	94.66		

⁽¹⁾—All calculations on residual material dried at 100° C; ⁽²⁾—Water soluble organic nitrogen included.

6.30

90.35

Total ash....

Totals.

Results as expressed in Tables III and IV are of a relative value; since the dry weight of the original material does not remain constant during the decomposition processes. Results in terms of actual weight as expressed in Tables V and VI give a much better information of the fate of the chemical constituents during the decomposition processes.

129

TABLE V

TOTAL CONCENTRATION OF THE CHEMICAL CONSTITUENTS OF SUGAR CANE TRASH COMPOSTED FOR 30 DAYS WITH OR WITHOUT ADDITIONAL NITROGEN

	Original	66 per ce	nt Water	80 per ce	nt Water	88 per cent Water			
Chemical Constituents	Material (20 gms)	No nitrogen added	Nitrogen added	No nitrogen added	Nitrogen added*	No nitrogen added	Nitrogen added		
	gm.	gm.	gm.	gm.	gm.	gm.	gm.		
Ether soluble fraction	0.370	0.193	0.234	0.183	0.187	0.162	0.154		
organic matter	0.696	0.608	1.115	0.720	1.062	0.535	0.828		
Hemicelluloses	5.264 6.570	4.326 5.608	0.341 3.028 3.500	0.270 4.209 5.277	0.380	$0.264 \\ 4.352 \\ 5.877$	0.346 3.007		
Lignin (nitrogen and ash	2 919	2 940	2 090	0.011	3.098	5.877	3.352		
Water insoluble protein	0.450	0.548	1,109	0.619	3.309 0.944	3.336	3.141 1.397		
Total ash	$0.000 \\ 1.260$	0.000 1.228	$ \begin{array}{c} 0.124 \\ 1.384 \end{array} $	$0.000 \\ 1.288$	$0.119 \\ 1.533$	$0.000 \\ 1.296$	0.111 1.356		
Totals	18.070	15.996	13.861	16.033	14.609	16.515	13.692		

TABLE VI

TOTAL CONCENTRATION OF THE CHEMICAL CONSTITUENTS OF SUGAR CANE TRASH COMPOSTED FOR 60 DAYS WITH OR WITHOUT ADDITIONAL NITROGEN

	Original	66 per ce	nt Water	80 per ce	nt Water	88 per cent Water			
Chemical Constituents	Material (20 gms)	No nitrogen added	Nitrogen added	No nitrogen added	Nitrogen added	No nitrogen added	Nitrogen added		
	gm.	gm.	gm.	gm.	gm.	gm.	gm.		
Ether soluble fraction	0.370	0.200	0.225	0,184	0.177	0.127	0.156		
organic matter	0.696	0.623	1.088	0.571	0.910	0.574	0.867		
Alcohol soluble fraction,	0.248	0.167	0.280	0.167	.245	0.169	0.202		
Hemicelluloses,	5,264	3,993	2.880	3,975	2.554	3.299	2.558		
Celluloses, Lignin (nitrogen and ash	6,570	4,540	2.605	5.025	2.395	4.092	2.391		
free).	3.212	3.137	3.339	3.188	3,102	2.942	3 999		
Water insoluble protein	.450	.841	1.286	789	1.386	860	1 551		
Nitrogen as ammonia	0.000	0.000	0.104	0,000	0.107	0.000	0.102		
Total ash	1.260	1,397	1.457	1,273	1.417	1.220	1.444		
· Totals	Totals		13.265	15.172	12.293	13.283	12.493		
· · · · · · · · · · · · · · · · · · ·				······································					

Tables VII and VIII give the per cent of each of the original groups left after composting for thirty and sixty days.

TABLE VII

RESIDUAL MATERIAL OF THE VARIOUS CHEMICAL CONSTITUENTS OF SUGAR CANE TRASH COMPOSTED WITHOUT THE ADDITION OF NITROGEN

		66 per ce	nt Water	80 per ce	nt Water	88 per cent Water			
	Original Material	30 days	60 days	30 days	60 days	30 days	60 days		
	gm.	Per cent of original	Per cent of original	Per cent of original	Per cent of original	Per cent of original	Per cent of original		
Total dry material Ether soluble fraction	20.00 0.370	89,50 52,16	82.00 54.05	87.00 49.46	82.00 49.73	91.00 43.79	74.00 34.32		
organic nlatter	$\begin{array}{c} 0.696 \\ 0.248 \\ 5.264 \\ 6.570 \end{array}$	87.36 98.79 82.18 85.36		103.45 108.87 79.96 81.84	$\begin{array}{c} 82.04 \\ 67.34 \\ 75.51 \\ 76.48 \end{array}$	76.87 106.45 82.67 89.45	82.47 68.15 62.67 62.28		
Lighins (nitrogen a n d ash free) Water insoluble protein Total ash	$3.212 \\ 0.450 \\ 1.260$	$100.87 \\ 121.78 \\ 97.46$	97.67 186.89 110.87	$104.83 \\ 137.56 \\ 102.22$	$99.25 \\ 175.33 \\ 101.03$	$103.86 \\ 154.00 \\ 102.86$	91.59 191.11 96.83		

TABLE VIII

RESIDUAL MATERIAL OF THE VARIOUS CHEMICAL CONSTITUENTS OF SUGAR CANE TRASH COMPOSTED WITH THE ADDITION OF NITROGEN

		66 per ce	nt Water	80 per ce	nt Water	88 per cent Water		
	Original Material	30 days	60 days	30 days	60 days	30 days	60 days	
	gm.	Per cent of original	Per cent of original	Per cent of original	Per cent of original	Per cent of original	Per cent of original	
Total dry material Ether soluble fraction	20.00 0.370	$74.50 \\ 63.24$	74.50 60.81	79,50 50,54	68.00 47.84	74.00 41.62	66:00 42:16	
organic matter	$\begin{array}{c} 0.696 \\ 0.248 \\ 5.264 \\ 6.570 \end{array}$	$160.21 \\ 137.50 \\ 57.52 \\ 53.27$	$156.32 \\ 112.90 \\ 54.71 \\ 39.67$	$152.59 \\ 153.23 \\ 63.01 \\ 56.29$	$130.75 \\ 98.79 \\ 48.52 \\ 36.45$	$118.97 \\ 139.52 \\ 57.12 \\ 51.02$	124.57 81.45 48.59 36.39	
Lignins (nitrogen an d ash free)	$\begin{array}{c} 3 & 212 \\ 0.450 \\ 1.260 \end{array}$	94.21 246.44 109.84	$103.95\ 285.78\ 115.63$	$104.89 \\ 209.78 \\ 121.67$	96.58 308.00 112.46	$97.79\ 310.44\ 107.62$	$100.31\ 344.67\ 114.60$	



The rate of conversion of the available forms of nitrogen into less available forms of nitrogen constituting the cells of microorganisms active in the decomposition processes is shown in Tables IX and X

TABLE IX

"NITROGEN TRANSFORMATION IN THE DECOMPOSITION OF SUGAR CANE TRASH"

(In per cent of total residual material)

		No Nitrogen Added							Nitrogen Added					
Form of Nitrogen		66\$ Water		80% Water		88% Water			66% Water		80≸ Water		885 Water	
	0 days	30 days	60 days	30 days	60 days	30 days	60 days	0 days	30 days	60 days	30 days	60 days	30 days	60 days
Total nitrogen Nitrogen as antmonia Soluble in hot and cold water Hydrolyzable by 2 per cent HCI "Humin" nitrogen, not acted by au- toclaving with 6 per cent H ₂ SO ₄	.64 0 .28 .12 .22	.68 0 .12 .19 .22	92 0 10 26 29	.70 0 .13 .24 .25	.86 09 .26 .28	.76 0 .15 .23 .26	1,06 0 .13 .38 .38	1.70	2.25 .83 .93 .43 .37	2.25 .70 .87 .61 .44	2.08 .75 1.07 .47 .37	2.46 .79 .83 .70 .49	2.30 .75 .79 .52 .42	2.67 .77 .79 .73 .49

TABLE X

TOTAL CONCENTRATION OF THE NITROGEN TRANSFORMATION OF SUGAR CANE TRASH DECOMPOSING FOR 30 AND 60 DAYS AT DIFFERENT MOISTURES, WITH AND WITHOUT ADDITIONAL NITROGEN

		No Nitrogen Added							Nitrogen Added					
Form of Nitrogen	materia	06K H_O		80% H2O		885 H _e O		materia		665 H ₂ O		О-Н	885 H_0	
	Original 20 gms	30 days	60 days	30 days	60 days	30 days	60 days	Original 20 gms	30 days	60 days	30 days	60 days	30 days	60 days
	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	mgm.	ngm.	mgm.	mgm.	ingm.	mgm.	mgm.
Total Nitrogen Nitrogen as anunonia Soluble in cold and hot water Hydrolyzable by 2 per cont HC1	$128 \\ 0 \\ 56 \\ 24$	122 0 21 34	$151 \\ 0 \\ 16 \\ 33$	$122 \\ 0 \\ 23 \\ 42$	141 0 15 43	$138 \\ 0 \\ 27 \\ 42$	157 0 22 34	340 212	335 124 138 64	335 104 130 91	331 119 170 75	335 107 113 95	340 111 117 77	352 102 104 96
claving with 6 per cent H ₂ SO ₄	44	39	48	44	46	47	38		55	66	59	67	62	65

DECOMPOSITION OF CANE TRASH AS MEASURED BY THE EVOLUTION OF CARBON DIOXIDE

The decomposition of cane trash as measured by the evolution of carbon dioxide with or without the addition of minerals was studied in three different soil types brought from Puerto Rico by special permission of the United States Quarantine Board. These soils were among the first ones classified in the Soil Survey of Puerto Rico

134 THE JOURNAL OF THE DEPARTMENT OF AGRICULTURE OF P. R.

which started in the fall of 1928 under the supervision of the United States Bureau of Soils. The type's are:

- 1. Río Piedras elay. This type belongs to the Río Piedras series, deep phase. It is typically developed on the hills of the Puerto Rico Insular Experiment Station. The sample was taken at Km. 2.25 Leprocomio road, Trujillo. This soil is used for sugar cane and other crops.
- Bayamón clay loam. This type belongs to the Bayamón series. It is strongly acid. The sample was taken at Km. 2.2, Sanatorium Road, Río Piedras. This soil is mainly used for citrus, sugar cane and other crops.
- 3. Múcara clay. It has the characteristics of Río Piedras clay but is higher in organic matter content. The sample was taken at Km. 2.8 Carolina–Juncos Road. This soil is used for sugar cane and other crops.

The Río Piedras clay and Bayamón elay loam are types representing poor sugar-cane soil. The Múcara clay represents a better sugar-cane soil.

All the soil samples were taken to a depth of six inches in a suitable place in the field. They were air-dried and sifted through a 2mm, sieve. The soils were free from carbonates.

To a duplicate set of seven 300 cc. Florence flasks the following materials were added:

- (2) Trash + $(NH_4)_2HPO_4 + K_2HPO_4$.
- (3) Trash + NaNO₃ + K_2 HPO₄.
- (4) Soil.
- (5) Soil + trash.
- (6) Soil + trash + $(NH_4)_2HPO_4 + K_2HPO_4$.
- (7) Soil + trash + NaNO₃ + K_2 HPO₄.

The first three treatments were inoculated with 1 c. c. soil infusion prepared by adding 100 c.c. of tap water to 10 grams of a rich soil (Sassafras sandy loam). The amount of trash added represented 5 grams of the dry material. Ground trash sample No. 1 (POJ-2714) was used. It was kept at optimum moisture by adding 200 per cent of its own dry weight as water.

The amount of soil used represented 100 grams of dry soil. Water was added to bring the moisture content up to 25 per cent. The equivalent of 42.3 mgm. of nitrogen and 100 mgm. of K_2HPO_4 were added as mineral sources. The complete set was connected to a carbon dioxide apparatus, as described by Waksman and Starkey. (24.)

⁽¹⁾ Trash.

The carbon dioxide given off was collected in one-sixth normal $Ba(OH_2)$. The excess of barium hydroxide was titrated back with one-sixth normal oxalic acid to determine the carbon dioxide absorbed, using phenolphthalein as an indicater. The experiment lasted 28 days. The system was aerated daily for four hours. Titrations were made every day during the first week and every other day, afterwards. Analyses for pH, nitrate and ammonia were made at the end. The quinhydrone electrode was used for pH determinations. Nitrates were determined by the phenoldisulphonic acid method. The ammonia was collected in 300 c. c. Florence flasks after consecutive leachings with 100, 50, and 50 c. c. portions of normal KCl and 50 c. c. water. The ammonia was distilled over after neutralizing with magnesium oxide.

TABLE	\mathbf{XI}
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EVOLUTION OF CARBON DIOXIDE FROM CANE TRASH ADDED TO PUERTO RICO SOILS WITH OR WITHOUT THE ADDITION OF MINERALS

	After 28 days								
T. extments	Total mgm. C as CO ₂	рИ	Nitrate mgm. of nitrogen in flask	Ammonia mgm. of nitrogen in flask					
Trash. Trash + $(NH_4)_2HPO_4 + K_2HPO_4$. Trash + $NnNO_3 + K_2HPO_4$. Múcara clay + trash. Múcara clay + trash. + $(NH_4)_2HPO_4 + K_2HPO_4$. Múcara clay + trash + $NaNO_3 + K_3HPO_4$. Río Piedras clay + trash. Río Piedras clay + trash. Bayamón clay + trash. Bayamón clay + trash. Bayamón clay + trash.	124 445 330 37 361 441 441 452 27 303 207 292 292 292 292 27 225 260 317	$\begin{array}{c} 7.16\\ 9.08\\ 7.02\\ 7.25\\ 4.6\\ 7.50\\ 5.3\\ 5.3\\ 5.3\\ \end{array}$	None None 15.3 9.2 None None None None None 2.4 None 20.1	None None None 1.8 None 1.7.7 None 28.6 11.6 None 82.0 None					

INFLUENCE OF SOIL TREATMENT UPON THE MICROBIAL POPULATION

In the previous treatment given to the Puerto Rico soils the number of fungi, bacteria and actinomyces were determined after fifteen and thirty days by the plate method. Peptone-Glucose acid Agar (21) was the medium used for fungi. Sodium Albuminate Agar (22) was the medium used for bacteria and actinomyces. Five plates were poured for each treatment. The plates were incubated at 28° C. Fungi were counted after three days. Bacteria and actinomyces were counted after ten days in the same plate.

136 THE JOURNAL OF THE DEPARTMENT OF AGRICULTURE OF P. R.

	Number of Fungi X 1,000															
Treatment	Río Piedras Clay			Bayamón Clay Loam				Múcara Clay				Toa Silt Loam				
	15	days	30	days	15	days	30	days	15	days	30	days	15	days	30 	days
Soil alone. Soil $+$ CaCO ₃ . Soil $+$ dried blood. Soil $+$ (NH ₄) ₂ SO ₄ . Soil $+$ (NH ₄) ₂ SO ₄ $+$ CaCO ³ . Soil $+$ cane trash Soil $+$ cane trash $+$ (NH ₄) ₂ HPO ₄ .		27.0 70 433 50 57 700 933		29.0 28 220 28 15 103 230		134.0 36 447 70 90 697 837		40 39 346 139 62 856 836		$46 \\ 31 \\ 216 \\ 45 \\ 40 \\ 236 \\ 560$		$28 \\ 22 \\ 236 \\ 26 \\ 24 \\ 180 \\ 166$		$\begin{array}{r} 40\\ 28\\ 290\\ 43\\ 25\\ 206\\ 510 \end{array}$		42 35 496 58 11 286 454
	Number of Bacteria X 100,000															
Treatment	Río Piedras Clay			ras	Bayamón Clay Loam				Múcara Clay				Toa Silt Loam			
	15	days	30	days	15	i days	30	days	15	days	30	days	15	days	30	days
Soil alone Soil + CaCO ₅ . Soil + dried blood. Soil + (NH ₄) ₂ SO ₄ . Soil + (NH ₄) ₂ SO ₄ + CaCO ₅ Soil + cane trash Soil + cane trash + (NH ₄) ₂ HPO ₄ .		63 180 163 91 180 490 700		$170 \\ 261 \\ 354 \\ 208 \\ 58 \\ 222 \\ 236 \\$		$227 \\ 134 \\ 1, 130 \\ 140 \\ 158 \\ 1, 246 \\ 1, 695 $		69 89 250 82 93 640 235		$34\\ 80\\ 24\\ 51\\ 42\\ 280\\ 264$		83 146 334 54 84 100 448		69 75 128 61 99 96 174	:	351 281 864 672 530 468 748
	Number of Actinomyces X 109,000															
Treatment	Río Piedras Clay			Bayamón Clay Loam				Múcara Clay				Toa Silt Loam				
	15	days	30	days	15	5 days	30	days	15	days	30	days	15	days	30	days
Soil alone. Soil + CaCO ₃ . Soil + dried blood. Soil + (NH_4) :SO ₄ + CaCO ₃ . Soil + (NH_4) :SO ₄ + CaCO ₃ . Soil + cane trash. Soil + cane trash + (NH_4) :HPO ₄ .		$29 \\ 61 \\ 360 \\ 46 \\ 68 \\ 267 \\ 363$		$\begin{array}{r} 40 \\ 52 \\ 154 \\ 62 \\ 24 \\ 88 \\ 144 \end{array}$		$ \begin{array}{r} 48 \\ 14 \\ 110 \\ 6 \\ 14 \\ 47 \\ 210 \\ \end{array} $		20 13 150 8 27 100 85	1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1	$ \begin{array}{r} 8 \\ 224 \\ 34 \\ 16 \\ 100 \\ 116 \end{array} $		$46 \\ 29 \\ 206 \\ 42 \\ 17 \\ 20 \\ 152$		17 23 220 29 27 60 116		$32 \\ 26 \\ 306 \\ 47 \\ 33 \\ 96 \\ 112$

TABLE XII

ABUNDANCE OF MICROORGANISMS IN SOILS KEPT AT OPTIMUM MOISTURE

DISCUSSION OF RESULTS

The two samples of trash differ considerably in composition, as shown in Table I. This may be due to differences in the varieties, to age effects or to the methods of sampling the materials. S.C. 12-4trash was taken as dead leaves from the lower part of the stems. P.O.J. 2714 trash was gathered from soil surface around the cane. Waksman and Tenney (26) have shown that the composition of plant remains is different with different plants and the same plant at different stages of growth. The younger the plant and the less mature it is the greater will be the content of readily decomposable material. The S.C. 12-4 has 2.81 per cent less water soluble organic matter, 1 per cent more hemicelluloses, 3.14 per cent more cellulose, 4.53 per cent more lignin and 6.49 per cent less ash than the P.O.J. 2714. The furfural determination shows that almost all the hemicelluloses of the P.O.J. 2714 were pentosans.

If both samples of trash were placed under similar environmental conditions, it would seem likely that the composition of the variety P.O.J. 2714 would be such as to result in its more rapid decomposition.

Celluloses and hemicelluloses add 55.24 per cent for P.O.J. 2714 and 59.17 per cent for S.C. 12(4). It is very important, therefore, to understand the role of soil microorganisms in decomposing such complexes for energy purposes. Organisms capable of decomposing celluloses under aerobic conditions are found among various groups of fungi, among the actinomyces and among certain specific bacteria. However, under anaerobic conditions, the fungi and actinomyces do not thrive and bacteria alone are entirely concerned in the process. Many fungi are capable of decomposing hemicelluloses. Certain specific bacteria and lower animals are also capable of assimilating hemicelluloses.

Certain hemicelluloses are more readily decomposed than others. The degree of decomposition varies with the nature of the plant and the type of hemicelloluse. Pentosans are more readily decomposed than hexosans.

Table 2 gives the dry weight losses of sugar-cane trash composted for thirty and sixty days with or without additional inorganic nitrogen. The trash was inoculated with soil infusion to provide for the presence of the soil variable microflora and microfauna. No deduction was made for the dry weight increase due to the addition of $(NH_4)_2SO_4$ because of the complicated nature of the chemical changes affecting this salt during microbial activities.

In the compost material containing 88 per cent water, evil smelling substances, seemed to indicate that some anaerobic decomposition was going on. Such evil smelling substances were not noticed in the other treatments. The presence of 88 per cent moisture represents the addition of 733 per cent of water to the dry material. The degree of saturation seems to be too high for the free circulation of air. As conditions were partly anaerobic, it is advisable, not to compare the results obtained in the 88 per cent H_2O treatment with those obtained in the 66 and 80 per cent H_2O treatments whose average value in this discussion is taken to represent aerobic decomposition. The nature of the processes involved would be different in loth cases.

It is obvious, that the information obtained from losses of dry weight, is of a very limited value. The individual groups constituting the trash complex are not affected at the same rate. Some groups tend to increase; others tend to decrease and others tend to remain more or less constant. The increase in some of the groups compensates the decrease in others and therefore an appreciable change of the total dry material is not noticed.

The ether and alcohol soluble fractions constitute a heterogeneous group of complexes such as oils, waxes, resins, tannins, terpenes, alkaloids, etc. As the percentage of these two groups in sugar-cane trash is quite small, namely around 3 per cent, not much stress is to be laid on their fate during the processes of decomposition. It seems, however, that about 50 per cent of the ether soluble fraction is of such nature as to decompose during the first thirty days. The rest seems to be more resistant to decomposition; even with the addition of nitrogen.

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The celluloses and hemicelluloses disappeared more rapidly than could be accounted for by the decrease in total dry weight. The decrease in weight due to their decomposition is balanced by the accumulation of proteins. Their disappearance may be due to actual loss of earbon as carbon dioxide or to transformation into the new complexes synthesized by the microorganisms. It must be remembered that in such a biological process as, composting sugar-cane trash, decomposition and synthesis take place side by side. The increase in water insoluble protein corroborates the synthesis of new protein cell material.

The lignin fraction remains more or less constant. Waksman and Tenney (28) studied the decomposition of composted rye straw under aerobic conditions for a period of 386 days. They found that the decomposition of celluloses and hemicelluloses accounted for most of the decomposed plant materials. This was accompanied by an increase of water insoluble protein and lignins.

Further studies on sugar-cane trash decomposition for a period over sixty days would give more evidence of the fate of the lignin fraction. It also will show up to what limit the celluloses and hemicelluloses become resistant to decomposition.

The addition of nitrogen in both, aerobic and partly anaerobic conditions, activated the heterotrophic organisms in decomposing more celluloses and hemicelluloses. This was accompanied by a larger increase of water insoluble protein and water soluble organic matter.

The total ash content remained more or less constant in all the treatments to which nitrogen was not added. The increase in ash content due to the addition of one gram of $(NH_4)_2SO_4$ varied only from 0.06 to 0.25 gram. This supports the statement previously made:

"No deduction was made in Table 2 for the dry weight increase due to the addition of $(NH_4)_2SO_4$ because of the complicated nature of the chemical changes affecting this salt during microbial activities."

Tables IX and X show that with the progress of decomposition there was a decrease of the nitrogen soluble in cold water and a corresponding increase in the nitrogen hydrolizable by dilute hydrochloric acid and in the more resistant or so-called "humin" nitrogen. This points, once more, to the building up by microorganisms of proteins and other complex organic nitrogenous compounds.

The 212 milligrams of nitrogen added to the compost was in excess to that required by the active microorganisms. Under aerobic conditions, after thirty days, an average of 122 milligrams of nitrogen as ammonia was recovered. After sixty days, 106 milligrams were thus recovered. The ratio of available nitrogen used to hemicelluloses and celluloses decomposed was as follows:

	66 per cent	80 per cent	88 per cent			
	water	water	water			
30 days	1:39	1:28	1:38			
50 days	1:28	1:39	1:22			

Practically all the nitrogen was recovered in the residual material. This shows that there was no loss of nitrogen either by volatization or reduction processes. These results throw further light upon the problem of synthesis of new protein material as a result of the activities of the microorganisms which bring about the decomposition of the celluloses and hemicelluloses. In the absence of additional nitrogen, the increase in the amount of water insoluble protein took place at the expense of the water soluble simple nitrogenous compounds. Some of the nitrogenous substances synthesized by the microorganisms, are only slowly available sources of nitrogen for soil microorganisms, otherwise the celluloses and hemicelluloses would have undergone a much more rapid decomposition. The slight increase of the total nitrogen fraction in the compost to which no nitrogen was added may be due to nitrogen fixation. Energy for the nitrogen fixers may have been taken from the disintegration products resulting from the decomposition of celluloses and hemicelhuloses.

At the end of the experimental periods the trash to which nitrogen was added was more brittle and readily crumbled between the fingers. Probably such a material would not interfere with the plowing operation. The decomposed material resembled soil humus in color.

Further studies extended over longer periods of time on decomposition of cane trash in the presence of available minerals, especially nitrogen and phosphorus, should give information on how to use the minerals in the most economical way.

Carbon dioxide as an index of soil fertility has been discussed by Waksman and Starkey (24).

Results given in Table XI indicate that the Múcara clay liberated the greatest amount of carbon dioxide in all the treatments. The rapidity of trash decomposition depends on the rate by which soil microorganisms utilize available minerals present, especially nitrogen. It seems that the Múcara clay supplies minerals at a faster rate than the two other soils. The Bayamón clay loam seems to have the lowest mineral spuplying power. From field returns it can be said that the Múcara clay is a better sugar-cane soil.

Carbon dioxide evolution in the Río Piedras clay was not stimulated by the addition of minerals. Probably some physical factor must be responsible for repressing microbial activity. May be this soil, at optimum moisture conditions, would behave differently.

Microorganic activity was repressed when $NaNO_3$ was added to the trash in the absence of soil. Probably this was due to high alkalinity (pH 9.0) or to toxicity of the sodium or nitrate ion. The buffer effect of the soil seems to check such effect.

No definite explanation can be given for the reduction of nitrates to ammonia when NaNO₂ and trash were added to the Río Piedras clay. This might have been due to an involuntary addition of $(NH_4)_2HPO_4$ for NaNO₃: but the increase to (pH 7.4) seems to indicate that there was not such an error.

Fungi, bacteria and actimomyces were most abundant when dried blood and cane trash were added. The organic matter in the form of those materials offered a good energy source for the heterotrophic soil organisms which develop well in the plate method.

When straw is added to the soil, the microorganisms decomposing

celluloses and hemicelluloses use the available nitrogen present in the soil, up to a certain limit. It seems dangerous to leave trash on a nitrogen starving soil planted to cane. The ratio between the amount of cellulose decomposed and the amount of nitrogen assimilated is about 30:1 in the case of fungi and aerobic bacteria. However, in the soil, where the cells of microorganisms freshly synthesized are constantly decomposed by other organisms, the ratio is 50-60:1 (25). In other words; for every unit of nitrogen that can become available in the soil in a definite period of time, about 50 to 60 units of cellulose will be decomposed. Such a ratio has not been experimentally established for hemicelluloses; but it is believed that an identical ratio prevails.

To hasten the decomposition of sugar-cane trash in the sugar-cane fields it is evident that available nitrogen must be given to the soil microorganisms. Nitrogen is supplied with great cost to most crops and especially so, to sugar-cane. It is a standard practice in Hawaii. Java, and Puerto Rico to apply heavy applications of inorganic fertilizers to fields of sugar cane. Hawaii leads with applications of 600 to 1,000 pounds of a complete fertilizer per acre followed by 400 to 600 pounds of sodium nitrate or ammonium sulphate. Puerto Rico is approaching Hawaii and applies 400 to 800 pounds of a complete fertilizer followed by 300 to 400 pounds of ammonium sulphate. Java growers are strong supporters of ammonium sulphate alone. They apply from 100 to 1,200 pounds per acre. Cuba is also using large amounts of fertilizers.

In the tropics, especially in the regions of high rainfall, there are great losses of available nitrogen in the run-off and percolating waters. Sugar-cane trash would serve as a store of available nitrogen. The nitrogen deposited as proteins in the cells of soil microorganisms would not leach so readily. Further microbial activity would convert the stored nitrogen into forms available for the sugarcane plant.

In 1927, Albrecht (2) reports:

"Some recent work by the Oregon Experiment Station indicates that artificial manure can be made successfully with ten pounds of nitrogen per ton of straw. It is customary in England to add about fourteen parts of nitrogen per ton of straw."

The average nitrogen per acre added to the sugar-cane fields of Hawaii and Puerto Rico is about 180 and 148 pounds, respectively. The addition of a part of this nitrogen to the sugar-cane trash would hasten its decomposition in the soil. Such addition of nitrogen to the trash would tend to improve the trash physical condition for the plowing operation.

A farmer in the United States adding every year an average cf 2,000 pounds of organic matter per acre is doing quite well. Such an average for the tropics would be too low because the decomposition of organic matter is greatly hastened by optimum climatic conditions.

Considering that thirty tons of cane per acre gives about ten tons of green straw and assuming that the green straw contains about 75 per cent of water; 5,000 pounds of dry material would remain in the soil. Deducting for ash, 10 per cent of the dry weight; 4,500 pounds of organic matter will be left for enriching the soil supplies of organic matter.

Nitrogen could be supplied, as a means of hastening trash decomposition, by a quick growing leguminous plant. Nitrogen could also be supplied by scattering over the trash part of the fertilizer added to the sugar-cane plant. Perhaps, it would be practical to adopt a wet spraying method.

The economic advisability of applying a source of available minerals, especially nitrogen, as a means of hastening trash decomposition must be established by adequate experimentation. The questions to be answered are: What form of nitrogen is the mostadequate? What amount of nitrogen must be added to make this practice a paying proposition? How should this nitrogen be added? How long will it take for the nitrogen transformed into the complex proteins of microbial protoplasm to become available for the use of the plant?

SUMMARY

The decomposition of sugar-cane trash on the basis of its chemical composition has been studied when kept at different moisture contents with or without the addition of inorganic nitrogen for a period of thirty and sixty days at 28°C. Results lead to the following conclusions:

- (1) The addition of inorganic nitrogen to the composted trash hastened the decomposition processes.
- (2) The ether and alcohol fractions account for a very slight portion of the total organic matter decomposition.
- (3) The decomposition of the celluloses and hemicelluloses account for most of the decomposed organic matter. As such groups represent a good source of energy for the heterotrophic organisms their disappearance may be due to actual loss of carbon dioxide or to transformations

into the new complexes synthesized by the microorganisms. This synthesis is further proven by an accumulation in crude protein and an increase in the water soluble matter.

- (4) The lignin and ash fractions remain more or less constant.
- (5) Practically all the nitrogen present and added was recovered in the residual material. There was no loss, therefore, of nitrogen either by volatilization or reduction processes. The slight increase of the total nitrogen fraction in the compost to which no nitrogen was added may be due to nitrogen fixation. Energy for the nitrogen fixers may have been taken from the disintegration products resulting from the decomposition of celluloses and hemicelluloses.
- (6) The addition of nitrogen improved the physical conditions of the trash. It was more brittle and readily crumbled between the fingers.
- (7) Addition of trash to three soil types from Puerto Rico increased the rate of carbon dioxide seven to nine times.
- (8) Influence of soil treatment upon the microbial population showed an increase in the numbers of fungi, bacteria and actinomyces when dried blood or sugar-cane trash was added to the soils.

ADDENDA

After this manuscript was sent to press the author received a reprint of Owen's and Denson's (31) recent work at Louisiana ou "The effect of plowing under cane trash upon the available nitrogen of the soil." The investigation was outlined in the following heads:

- 1. "The various organic constituents of cane trash.
- 2. The rate of decomposition of cane trash and its various fractions as measured by the evolution of carbon dioxide.
- 3. The effect of age upon the depressing action of cane trash upon soil nitrates.
- 4. The influence of cane trash upon the soil microflora.
- 5. The utilization of forms of nitrogen other than nitrates for the decomposition of cane trash in soils.
- 6. The cycle of nitrogen incorporation and release from microbial protoplasm.
- 7. The influence of cane trash upon plant growth.
- 8. The effect of turned under cane trash upon moisture retention in the soil.

9. The agronomic phases of plowing under cane trash." The authors conclusions were:

1. "The addition of fresh cane trash to soils containing nitrates causes a very rapid transformation of the nitrogen into organic forms.

144 THE JOURNAL OF THE DEPARTMENT OF AGRICULTURE OF P. B.

- 2. The depressing effect of cane trash upon nitrates, diminishes at a fairly constant rate when this crop residue is in contact with the soil, and under ordinary conditions it has perhaps lost most of its pernicious effects at the time the fertilizer is applied to cane.
- 3. The addition of fresh trash retards appreciably the rate of growth of corn plants, but the incorporation of trash which has been partially buried for several months, increased the yields and the initial rate of growth.
- 4. The addition of cane trash to soils increase their water holding capacity, and decreases their rate of drying, so that in seasons of drought this may be quite a factor in conserving the moisture of the soil.
- 5. The addition of cane trash to soils results in appreciable gains of total nitrogen, indicating the stimulation of the nitrogen fixing bacteria by the products resulting from the decomposition of organic matter incorporated in the soil.
- 6. The fact that field experiments over a period of several years at this Station, have shown no decrease in crop yields from the practice of turning under cane trash it would appear that the nitrate nitrogen immobilized by the presence of the organic matter, is in excess of the immediate requirements of the growing crop. This would make the practice of turning under trash, consistent with the necessity of conserving the surplus available nitrogen in the soil.
- 7. Whether the continued practice of incorporating such large quantities of organic matter with a C:N ratio of approximately 1:40 would tend to so increase the accumulation of the more resistant constituent lignin, as to unfavorably affect the composition, and the productiveness of a soil is a question, the importance of which warrants further study."

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