THE COMPOSITION OF TURFGRASS
THATCH AND THE INFLUENCE OF
SEVERAL MATERIALS TO INCREASE
THATCH DECOMPOSITION

Thesis for the Degree of M. S. MICHIGAN STATE UNIVERSITY DAVID P. MARTIN 1970

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ABSTRACT

THE COMPOSITION OF TURFGRASS THATCH AND THE INFLUENCE OF SEVERAL MATERIALS TO INCREASE THATCH DECOMPOSITION

By

David P. Martin

The composition of turfgrass thatch was investigated to determine the chemical nature of the thatch layer and possible causes for its accumulation. Analyses of total cell wall, hemicellulose, cellulose, and lignin content on two layers of Toronto creeping bentgrass,

Merion Kentucky bluegrass, and Pennlawn red fescue thatch were made and used as an indication of the state of thatch decomposition. The same determinations were also made on the individual leaf, stem, and root components of these three turf species. Comparisons could then be made to ascertain which plant component comprised most of the thatch layer.

Total cell wall content was significantly lower in Toronto creeping bentgrass thatch than the other two species, as were hemicellulose and cellulose in the lower thatch layer. Lignin, the most resistant plant constituent

to microbial activity, was found in greatest quantities in the thatch layer nearest the soil, decreasing upward. Red fescue thatch contained higher percentages of lignin than the other two species.

Total cell wall content was considerably higher in the roots than the leaves in all species studied, and also higher than the stems in creeping bentgrass. Hemicellulose and cellulose constituents were variable between leaf, stem, and root material depending on the species. Lignin content was highest in the roots, followed in order by the stems and leaves. Total lignin content of the plant was in the order of red fescue>creeping bentgrass>Kentucky bluegrass.

In a second investigation, carbon dioxide evolution was utilized as an indicator of microbial activity under in vitro conditions. An attempt was made to increase biological degradation of thatch by the addition of (a) two enzymes, pectinase and cellulase, (b) sucrose, and (c) ferulic acid, a lignin precursor. In addition, pH levels ranging from three to nine were briefly investigated for their effect on microorganism activity. A sealed environment was maintained at 24 C for 24 hours at which time gas samples were analyzed for CO₂ content using a gas chromatograph. Turf plugs were incubated with the same four materials in a controlled environment in order to investigate degradation under more natural conditions. Weight

loss of the thatch and total cell wall content determinations were used as an indication of decomposition.

Increased carbon dioxide evolution resulted as rates of material were increased. Peak production did not occur until the third day when the two enzymes were added at high rates. Maximum increases in carbon dioxide evolution were obtained from the additions of pectinase and sucrose. Although conclusive proof was lacking for the added carbon source to have been completely utilized, the trends strongly suggest that additional thatch decomposition resulted from the addition of these materials.

A pH of six was optimum for microbial activity, with five and seven also favorable. Acidic conditions were superior to alkaline conditions for thatch decomposition. In the controlled environment chamber study, total cell wall determinations indicated thatch treated with pectinase and ferulic acid was decomposing at a more rapid rate than the untreated thatch.

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Ву

David P. Martin

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INTRODUCTION

The primary turfgrasses established in the coolhumid climatic regions of the United States for home
lawns, recreation areas, and golf courses are creeping
bentgrass (Agrostis palustris Huds.), Kentucky bluegrass
(Poa pratensis L.), and red fescue (Festuca rubra L.). In
recent years, a great deal of emphasis has been placed on
the development of improved turfgrass varieties and cultural practices for the maintenance of high quality turfs.
Included in these practices are frequent and close defoliation, high fertilizer applications, intensive irrigation,
and the use of fungicides, herbicides, and insecticides.

Because of these practices, resulting from the demand for superior quality turfgrass areas, the grass plant is being grown in an unnatural, stress environment. Turfgrass plants growing in this environment slough off leaf, stem, sheath, and root parts more rapidly than under natural conditions. This organic debris often accumulates between or around the crown of the plant and the soil surface, creating a layer which has been termed thatch.

A certain amount of thatch is desirable for resiliency on athletic fields and golf course putting greens.

An excessive thatch layer on turf areas is objectionable

since it causes (a) increased disease susceptibility, (b) restricted rooting, (c) reduced air and water exchange, and (d) less resistance to environmental stress conditions for the grass plant.

In the past, methods of preventing thatch accumulation have emphasized mechanical procedures. Included are coring, grooving, slicing, spiking, topdressing, and fertilizer and lime combinations. At best, these have been temporary solutions with the thatch problem reoccurring after a period of time.

The objectives of this study were to determine (1) the chemical composition of thatch in various phases of decomposition, (2) the relationship of the chemical composition of various plant parts and turfgrass species to the accumulation of thatch, and (3) the potential of using various organic materials for inducing or stimulating microorganism activity to increase the decomposition of thatch.

LITERATURE REVIEW

Factors Influencing the Decomposition of Organic Matter

Organic matter in the soil is composed primarily of plant materials, plus some animal residues and cells of microorganisms. The decomposition of organic matter is influenced primarily by (a) the chemical nature of the plant material, (b) the physical and chemical properties of the soil, and (c) certain climatic factors, according to Millar, et al. (16). More mature plant tissues are more resistant to decomposition. The soil factors influencing decomposition are aeration and cultivation, fertility level, and pH, while the principle climatic factors influencing decomposition are moisture and temperature (1).

The composition of the parent plant material is usually the predominant factor affecting the nature of the organic matter (17). Thus, organic matter will be composed primarily of lignin, protein, polysaccharides, and other carbohydrates. Lignin is the plant constituent most resistant to biological attack. Consequently, lignin is found in soil organic matter in proportionately higher amounts than in the undecomposed plant. The soil humus may be composed by as much as 40-45% lignin and lignin complexes (16).

The decomposition of organic matter occurs primarily by the degradative processes of the soil microbes. Rangaswami (22) lists the chief soil factors influencing the microbial population as being the fertility level, moisture, aeration, temperature, organic matter, pH, and soil cultural practices. Thus, the microorganism population is influenced by the same factors which influence decomposition of organic matter. The optimum temperature of most microorganisms is about 25-30 C and the mesophilic range 20-30 C according to Alexander (1).

Carbon dioxide is evolved by respiration of soil microorganisms (32). This provides a method for measuring decomposition rates of organic matter. Three possible techniques are suggested by Alexander (1): (a) measurement of carbon dioxide evolution or oxygen uptake, (b) determination of the decrease in organic matter either chemically or by weight loss, and (c) observation of the disappearance of a specific constituent such as cellulose or lignin.

There is a certain amount of native carbon present in the soil as well as the carbon added to the organic matter from plant residues. Hallman and Bartholemew (12) used radioactively labeled carbon to ascertain the carbon source. By monitoring carbon dioxide evolution, they could distinguish between the loss of native soil carbon and that from the added plant residues. Carbon dioxide evolution by microorganisms was directly correlated with

the microbial population in the soil and can be used as an estimate of the population.

The cultural practices on turfgrass areas results in a considerably altered micro-environment. This probably causes a radical shift in the microflora and microfauna populations. No quantitative or qualitative studies of these populations of thatched turfs have been reported.

Cell Wall Constituents of Plants

The chemical composition of plant tissues is very complex. This review will cite a few references which relate to the major plant constituents of concern in this investigation.

Alexander (1) divides the organic constituents into six classifications as follows: (a) cellulose, (b) hemicellulose, (c) lignin, (d) water-soluble fraction (simple sugars, amino acids, etc.), (e) ether and alcohol soluble fraction (fats, oils, waxes, etc.) and (f) proteins. The first three are quite important in organic matter decomposition.

According to Meier (15), hemicellulose, meaning half-cellulose, is not a very precise term, since it is a collective name referring to those plant cell wall polysaccharides other than cellulose.

Fahn (11) reports the presence of hemicellulose in both primary and secondary cell walls. It ranks second

only to cellulose in the amount found in living plant tissues. When returned to the soil as organic matter, hemicellulose decomposes rapidly at first, but then persists for a fairly long time (30). Starkey (25) found the hemicellulose content of rye (Secale cereale L.) plants to be 17% when young and 23% when the plant matured.

Cellulose is also present in both the primary and secondary cell walls (11) and is by far the most common organic compound found in living plants. About one-third of the carbon transformed into organic compounds by plants is made into cellulose according to Bonner and Varner (5). Cellulose comprises about 50% of wood and more than 90% of cotton cloth. Armstrong and others (3) found the cellulose content was much higher in grasses than either legumes or Sullivan (27) reported an average cellulose content of 23% for Kentucky bluegrass (Poa pratensis L.) grown as a forage grass and cut successively over the summer period. Phillips and his coworkers (21) reported that Kentucky bluegrass increased in cellulose content from 22% during the vegetative stage to 30% at the time of flowering. Starkey (25) found young rye plants contained 18% cellulose and the mature plants 36%. Finally, Patton (19) reported a cellulose content of 18.5% for young tall fescue (Festuca arundinacea Schreb.) plants and 46.8% for mature, dry plants. From these several papers and the reports of other authors it is quite clear that the

cellulose content of plants is very high and increases quantitatively as the plant matures.

The third most abundant plant constituent is lignin (1, 6). It is a collective term used for a group of high molecular weight, amorphous compounds which (a) cement and anchor cellulose fibers together and (b) stiffen and protect them from chemical and physical attack.

According to Bonner and Varner (5), lignin is quite resistant to chemical degradation and even more so to enzymatic digestion. Lignin is a major component of the middle lamella which acts as a cement to hold cells together to form tissues (11). Most authors report the presence of lignin only in the middle lamella and secondary cell wall. However, Wardrop and Bland (33) report lignification beginning at the cell corners of the primary. wall and extending to the middle lamella and secondary walls. Lignin encrusts cell walls, forming woodlike tissues and serves as a major component of conducting vessels, fibers, and tracheids which strengthen plants (5). decomposes more slowly than any other plant constituent and is a major factor in the rate of organic matter decomposition.

Lignin is a major component of wood, accounting for 22-34% (5). As plants mature, the lignin content increases rapidly due to the continuing lignification process. Percentages of lignin in Kentucky bluegrass

increased from 3.4% to 7.14% as the plant matured (21). Sullivan (26) reported lignin contents of 4.7% in the first cutting of Kentucky bluegrass and 6% in the second Phillips and Smith (20) reported a higher lignin content in mature plant heads of timothy (Phleum pratense L.) while Soluski and coworkers (24) found the average lignin content in leaves, stems, and heads of orchardgrass (Dactylis glomerata L.), bromegrass (Bromus inermis Leyss.), and wheatgrass (Agropyron spp.) to be 5.3, 7.8, and 8.9% respectively. The lignin content of rye increased from 9.9% in young plants to 17.1% in mature plants (32). lignin content of tall fescue plants increased during maturation from 4% to 20% according to Patton (19). Under turf conditions, Ledeboer (14) reported a 14.8% lignin content for a composite grass sample of colonial bentgrass (Agrostis tenuis Sibth.), red fescue, and common Kentucky bluegrass.

Control of Turfgrass Thatch

Turfgrass thatch is a somewhat special form of organic matter in that it accumulates under modified environmental conditions on the surface of the soil rather than being incorporated into the soil. Many reasons have been advanced for the thatch problem as well as numerous solutions. To date, most remedial attempts to control thatch have involved mechanical methods such as topdressing, cultivation, and vertical mowing.

Thompson and Ward (28, 29) reported the most effective practice to reduce thatch accumulations was frequent topdressing with soil. Frequent coring coupled with vertical mowing reduced the accumulation of thatch and also the number of topdressings required. Individual cultural practices had little positive effect except for topdressing.

The greatest reduction in thatch resulted from topdressing and groove cultivation in a long term study by Engel and Alderfer (10). Annual liming and core cultivation were also desirable cultural practices, while high nitrogen levels and wetting agents appeared to increase thatch. Engel (9) also attributed the puffiness in bentgrass putting greens to excessive nitrogen rates which caused crowding and superficial rooting.

Many sources also refer to increased thatch problems being associated with acidic soils. Edmond and Coles
(8) reported the development of thatch wherever the soil
pH was five or less. The decomposition of organic residues
in a velvet bentgrass turf was increased with each additional increment of limestone applied. This area of investigation is not clearly understood, due to the lack of
information regarding the original pH and failure to
isolate this factor as a single variable.

In a more detailed experiment, Ledeboer (14) found no significant decomposition of thatch with treatments

utilizing combinations of limestone, gypsum, sucrose, and nitrogen. Schery (23) reported fertilizer rates had little effect on thatch accumulations, while thatch was increased slightly where clippings were retured. Starkey (25) suggested increased thatch decomposition was influenced primarily by temperature, moisture, aeration, and liming of acidic soils. He further stated that decomposition probably could not be accelerated by the addition of microorganisms, but that they naturally develop according to a particular set of conditions.

Ledeboer (13) reported the cellulose, lignin, and organic matter content was higher in the upper part of the thatch layer in comparison to the lower layer. The opposite result would have been expected. The cellulose content of the thatch was lower than in the living plant, indicating rapid loss when the plant died. The lignin content of the thatch was 26-27%. This is several times higher than the amount normally found in living plants. This increase in the percent lignin in thatch is a result of its resistance to microbial degradation.

MATERIALS AND METHODS

Source of Thatch Samples

Thatch samples of 'Toronto' creeping bentgrass, 'Merion' Kentucky bluegrass, and 'Pennlawn' red fescue were obtained from mature turfgrass areas established in 1962. The thatch layers had accumulated under cultural practices typical of those normally utilized for these three species in Michigan.

The cultural practices for 'Toronto' creeping bentgrass turf were representative of golf course putting green conditions. It was mowed six times per week at 0.25 inch with clippings removed. The nitrogen fertilizer level was in the range of 6 to 8 pounds per 1,000 square feet per year. Supplemental watering was frequently practiced during the hot summer months, and at other times of the year when required to prevent wilting. Fungicide applications, cultivation, and topdressing were performed only when needed. Lead arsenate was applied every two years for annual bluegrass control and MCPP annually for broadleaf weed control. The soil at this site was classified as a loamy sand with a pH of 7.3. The phosphorous and potassium levels were 51 and 97 pounds per acre respectively. The thatch had accumulated to a depth of

approximately one inch, but was intermixed with topdressing soil and living plant parts. These two components were undesirable for the types of studies to be conducted and were difficult to separate from the thatch.

The 'Merion' Kentucky bluegrass turf received a total of 6 pounds of nitrogen per 1,000 square feet per year in three equal applications. The soil in this area was a sandy loam with a pH of 6.8. The respective tests for phosphorous and potassium were 68 and 241 pounds per acre. The bluegrass was mowed twice weekly at 1.5 inches with clippings returned. Supplemental watering was applied as needed to prevent wilting. The area received no cultivation, liming, topdressing, or fungicide applications. Occasionally the herbicide 2,4-D was applied for broadleaf weed control, and chlordane was applied once for insect control. The thatch accumulation was 0.5 to 0.75 inches thick.

The Pennlawn red fescue was mowed twice weekly at 1.5 inches with clippings returned. Nitrogen fertilization totaled 3 pounds per 1,000 square feet per growing season applied in two equal applications spring and fall. The pH of the sandy loam soil was 7.3 with 92 pounds of phosphorous and 191 pounds of potassium per acre. Irrigation of the area was practiced as needed to prevent wilt. The area received no cultivation, liming, topdressing, or fungicide applications. Chlordane was applied

once for insect control and 2,4-D twice for broadleaf weeds. The thatch layer had accumulated to a depth of 1.5 inches.

A number of sod pieces were harvested from each location. The soil was removed from the bottom of the thatch and the vegetation on top carefully cut off, leaving only the intact layer of thatch. The red fescue thatch was cut into three equal layers, each being 0.5 inches thick. Since the thatch layer of bentgrass and bluegrass was thinner, it was divided into two layers of equal thickness.

The preparation of the thatch samples was similar for all species. The individual thatch layers were torn apart by hand and a small quantity placed in a Waring blender containing 300 ml water. The blender was operated for one minute to further disperse the thatch and to place the finer soil particles in suspension. The contents of the blender were filtered using a 40 mesh sieve. larger thatch pieces were retained on the sieve and thus could be separated from the larger soil particles. filtrate was then re-filtered through a 100 mesh sieve to remove some of the finer soil particles but to retain most The larger mineral particles were removed of the thatch. by hand. The thatch was dried at 40 C and ground in a Wiley Mill equipped with a 1 mm screen. The ground thatch samples were placed in a 40 C drying oven for 24 hours and then held at a constant 30 C temperature for later analysis.

Preparation of Living Plant Components

Leaf, stem, and root components of the three grass species were also obtained from the same turfgrass areas during June 1969. The leaves were collected by mowing at the standard cutting height of each species. The leaves were dried at 40 C, ground in a Wiley Mill having a 1 mm screen, and stored for later analysis at 30 C.

Representative pieces were harvested for the isolation of stem and root samples. The stem tissue referred to in this experiment includes the plant from the base of the crown to the top of the first leaf sheath, and in the bentgrass, some stolons were also included. This part of the plant was separated by hand so that it contained only stem tissue. The root component was obtained by washing the soil through a screen with water under pressure so that only the roots, and rhizomes remained. The roots and stems were dried at 40 C, ground in a Wiley Mill with a 1 mm screen, and stored at 30 C for later analysis.

<u>Determination of Carbon-Nitrogen</u> Ratio of Thatch

A certain minimum level of nitrogen is needed for optimum microorganism activity in an organic layer. Since a thatch layer had accumulated on the areas utilized for this investigation, it was desirable to determine the carbon to nitrogen ratio to ascertain whether nitrogen was

limiting. Samples of the several thatch layers were ground very fine to pass through an 80 mesh sieve. The carbon content was determined by dry combustion, by placing 10 mg samples in an induction furnace carbon analyzer. A direct carbon reading was obtained. Total nitrogen was determined by the Kjehdahl method. The ratio of the two elements was then calculated.

Quantitative Determination of Cell Wall Constituents

Total cell wall, hemicellulose, cellulose, and lignin determinations were made on duplicate samples of the various thatch layers, and on the leaf, stem and root components of the three turfgrass species. The analytical procedures followed were those outlined by Van Soest (25) and Van Soest and Marcus (26). One gram samples were placed in 250 ml erlenmeyer flasks, and 0.5 g sodium sulfite, 2.0 ml decahydronaphthalene, and 100 ml neutral detergent fiber (NDF) solution added. This mixture was boiled for one hour and suction filtered through a previously weighed 50 ml filter crucible. The residue was washed twice with boiling water and several times with The crucible containing the washed thatch was dried at 100 C for 15 hours, weighed, ashed, and reweighed so that the original one gram sample could be corrected for the mineral content. This was particularly important for the thatch samples which still contained a significant amount of soil.

The same procedure was repeated on identical samples, using the same materials except for the substitution of acid detergent fiber (ADF) for NDF. Following digestion, filtering, drying and weighing, the ADF residue was filtered for one hour with 72% sulfuric acid to obtain acid detergent lignin (ADL). The residue was dried, weighed, and ashed so that a correction could be made for mineral matter.

The following calculations were made after correction of the original samples for mineral content. Quantitative estimates were obtained as follows: (a) total cell wall (NDF-ash), (b) hemicellulose (NDF-ADF), (c) cellulose (ADF-ADL), and (d) lignin (ADL-ash). Each constituent was expressed as a percent of the total sample. The precision of the technique was such that values between replications were generally within .5%.

<u>Preparation of Materials for</u> <u>Increasing Thatch Decomposition</u>

This phase of the investigation was designed to study the feasibility of increasing the biological degradation of thatch by adding certain materials which stimulate microorganism activity. Only red fescue thatch was used for this study. The thatch was separated from the soil and above ground plant parts and passed through a Wiley Mill having a 1 mm screen. A large quantity of thatch was prepared so that sub-samples could be drawn from a uniform sample for all experiments and duplicates.

Pectinase, cellulase, sucrose, and ferulic acid were selected for detailed study based on a preliminary experiment involving 14 materials. Pectinase,* a commercially prepared enzyme, was diluted with distilled water to concentrations of 1:1000, 1:500, 1:200, 1:100, and 1:20. Sucrose, a disaccharide sugar, was also diluted to the same concentrations. Cellulase,* another commercially prepared enzyme, and ferulic acid, a lignin precursor, were prepared to dilutions of 1:2000, 1:1000, 1:200, and 1:100. In addition, a series of buffered solutions ranging from pH three to pH nine was prepared to investigate the effect of pH.

Measurement of Microorganism Activity

Thatch decomposition, as influenced by the addition of various materials, was determined by the following in vitro technique. Ground thatch samples weighing 1.5 g were placed in 25 by 200 mm glass tubes. Five ml of the prepared solutions were added per treatment. An hour later, 1 ml of distilled water was added to provide the optimum moisture level which had previously been determined. In addition to the treatments prepared in triplicate, a 6 ml distilled water plus thatch treatment was included to provide a control. After setting for several

^{*}These two enzymes were obtained from Nutritional Biochemicals Corp., Cleveland, Ohio.

hours, the thatch was completely moist and a 16 mm sleeve type rubber stopper was placed over the mouth of the tube to seal it. Each tube was held at an angle and the bottom of the tube gently tapped. The thatch sample did not remain completely on the bottom, but settled at an angle from the bottom to 5 cm up the side of the tube. The racks containing the tubes were placed in a controlled environment chamber in the light at 24 C for 24 hours.

After a 24 hour incubation period, the carbon dioxide content in the tubes was measured as an indication of the degree of microbial activity. This procedure was accomplished by using a 2.5 cc syringe, flushing it once in each treatment before withdrawing a 1 cc gas sample for analysis. The 1 cc sample was then injected into a gas chromatograph instrument. With the aid of a flowing stream of helium gas, the sample passes through a network of tubes packed with silica gel which separates the gases. The CO₂ is passed over a hot wire. The wire becomes hotter or cooler depending on the atmosphere and the concentration of CO2. The change in resistance of the wire alters the flow of current which is amplified and recorded on a potentiometer. By also measuring a stock gas sample, a factor was derived which was used to calculate the percent carbon dioxide in the treated samples.

The rubber stoppers were removed from all tubes to allow a complete exchange of air, followed by resealing

the tubes and placing in the controlled environment chamber for another 24 hour period after which the measurements were repeated again.

Determination of Carbon Source for Microorganisms

It was necessary to ascertain whether the thatch was still serving as the substrate in this system or whether the microorganisms were utilizing only the carbon added by the various materials. This possibility was checked in two ways:

- 1. The total carbon evolved as carbon dioxide was calculated and compared to the amount of carbon added by the treatment material. In this way it could be determined whether the added carbon supply had been exhausted.
- 2. Thatch samples were also analyzed for total cell wall content to determine whether additional thatch decomposition had occurred in comparison to an untreated control.

Controlled Environment Chamber Study

This investigation was designed to determine the possibility of increasing thatch decomposition under more natural conditions. Plugs 10 cm in diameter and 7.5 cm in depth were cut from the red fescue turf having a 1.5 inch thatch layer. These samples were inserted in waxed paper containers and placed in a controlled environment chamber

having a day/night temperature of 24/17 C and a light intensity of 2,000 foot candles for 16 hours.

The design was completely randomized with eight replications, and five treatments consisting of a control (water), pectinase, cellulase, sucrose, and ferulic acid. The treatments were diluted with distilled water to a ratio of 1:100. In order to avoid any foliar burn, the treatments were applied by using a syringe to inject the material directly into the thatch layer at four locations in each plug. A quantity of 2 cc of each material was applied to each sample six times per week.

The experiment was terminated after six weeks and results obtained by two methods. The thatch sample was isolated by removing the soil and live grass plants, oven dried, and then weighed. In addition, total cell wall determinations were made according to the procedure previously outlined.

RESULTS AND DISCUSSION

Chemical Nature of Thatch

Carbon-Nitrogen Ratio

In order for the microflora in organic matter to effectively utilize the carbon in the organic residue, a minimum amount of nitrogen must be present. This relationship is expressed as the carbon-nitrogen ratio. If the ratio is too high, nitrogen could be a limiting factor in thatch breakdown, resulting in additional thatch accumulation. The C:N ratios determined for the turfgrass thatch used in this investigation are listed in Table 1.

Table 1.--The C:N ratios for the thatch layers of Toronto creeping bentgrass, Merion Kentucky bluegrass, and Pennlawn red fescue.

Species	Thatch layer	Carbon-nitrogen ratio
Creeping bentgrass	Upper Lower	2 4 27
Kentucky bluegrass	Upper Lower	41 48
Red fescue	Upper Middle Lower	29 26 28

The ratios range from 24-48. Millar et al. (16) report a narrow range of 15-20 is needed to meet the minimum nitrogen requirements of the microorganisms. Apparently, the turfgrass thatch examined in these studies is lacking in nitrogen required for optimum decomposition rates. This may be somewhat surprising in view of the high nitrogen rates applied to turfgrass areas, however, rapid leaching of nitrogen may also be a factor, particularly under intense irrigation. In addition, the rates at which plant parts are sloughed under turfgrass conditions is very high, resulting in a high return of organic debris to the surface soil. Consequently, the C:N ratio may be one reason for thatch accumulation.

Cell Wall Analysis of Thatch

The thatch samples prepared for chemical determinations were analyzed for total cell wall, hemicellulose, cellulose, and lignin content (Table 2). The total cell wall content of Kentucky bluegrass and red fescue thatch was quite high compared to creeping bentgrass and also increased from top to bottom within the thatch layer. This would be expected since the more soluble plant constituents are no longer present in the lower layer. The results of creeping bentgrass thatch are quite variable. This can be explained in part by the very poor sample obtained for this species since the thatch layer had a much higher percentage of living plant material and also

Table 2.--Quantitative determinations of cell wall constituents for several Ø layers of thatch from three turfgrass species expressed as percentage of the total thatch dry weight.

Species	Thatch Layer	Total Cell Wall	Hemicellulose	Cellulose	Lignin
Creeping bentgrass	Upper Lower	59.0a [†] r‡ 54.8a [†] r	28.9a r 20.7b r	20.8a r 20.8a r	9.3a r 13.3b r
Kentucky bluegrass	Upper	75.3a s	34.9a s 34.9a s	30.0a r 27.5a s	10.4a r 13.2b r
Red fescue	Upper Lower	71.0a s [‡] 74.8a s	25.8a r 26.8a t	31.la r 31.7a s	14.1a s 16.3a s
hsd .05 hsd .01		4.9	3.75.5	5.0	2.0

diffollowed by a different letter (a, b, c) show significant level between leaf, stem, and root within a species using † Means ferences at 1% Tukey's test.

[‡]Means followed by a different letter (r, s, t) show significant dif-s at 1% level comparing leaf to leaf, etc. of different species using ferences at 1% Tukey's test. had a large quantity of soil topdressing. This made it difficult to free the thatch of mineral matter and perform a representative analysis. In addition, the quantity of living material present probably caused lower values because of the higher percentage of soluble cellular constituents in the living plant.

Differences in the hemicellulose and cellulose content of the thatch layers were quite variable. Bluegrass and red fescue had a significantly higher percentage in the lower thatch layer than bentgrass. One would have expected the less complex, more soluble hemicellulose and cellulose fractions to decline in the lower thatch layers as compared to the upper layers since they have been exposed to degradative processes for a longer period of time. Apparently lignin blocks the entrance of microbes to these more soluble compounds and retards decomposition as indicated by Bonner and Varner (5).

The differences in lignin content were significant between species and also between layers within a species. The lignin content of the lower thatch layer was higher than the upper layer for all three species. This would be explained by the more soluble plant constituents being previously decomposed in the lower, older thatch layer, consequently, the lignin as a percentage of the thatch remaining would be higher. The upper thatch layer has not been exposed to microbial action for as

long a period of time and in addition, contains a greater amount of living tissue. Both factors would lower the percent lignin content present in the upper thatch layer. These percentages for lignin (Table 2) are considerably lower than the figures reported for turfgrass thatch by Ledeboer (14).

Cell Wall Analysis of Grasses

A great deal of data is available on cell wall constituents for forage grasses, but very little information is available for turfgrasses. This information would be desirable in comparing the differences in the rate of thatch accumulation between species. Ledeboer (14) reported the organic matter content of thatch for a mixed stand of turfgrass species as 84.8% and a lignin content of 14.8%. No other data is currently available.

The percent total cell wall (TCW), hemicellulose, cellulose, and lignin are given in Table 3 for the leaf, stem, and root components of Toronto creeping bentgrass, Merion Kentucky bluegrass, and Pennlawn red fescue. TCW content of the leaves was in the order creeping bentgrass> red fescue>Kentucky bluegrass. Creeping bentgrass leaves had the highest TCW content of the three species. This is attributed to the large number of parallel vascular strands found in creeping bentgrass leaves. Further, all species had a much higher TCW content in the roots than in the leaves. The TCW of stems was also much higher

Table 3.--Quantitative determinations of cell wall constituents for leaf, stem, and root components of three turfgrass species expressed as a percentage of the total sample on a dry weight basis.

Species	Total Cell Wall	Hemi- cellulose	Cellulose	Lignin
Creeping bentgrass Leaf Stem	56.3a [†] r [‡] 56.5a [†] r	34.2a r 29.8b r	18.5a 22.5b r	3.7a r 4.2a r
Root Kentucky bluegrass Leaf Stem Root	76.3b ^T r 45.6a r [‡] 71.7b 70.8b r	35.8a r 25.7a s 39.3b s 33.9c r	26.6c r 17.6a r 27.5b s 27.1b r	13.9b s 2.3a r 4.8a r 9.8b r
Red fescue Leaf Stem Root	50.6a r [‡] 75.2b s 79.6b r	26.7a s 28.9a r 33.9b r	21.2a s 35.2b t 33.1b s	2.7a r 11.1b s 12.6b
hsd .05 hsd .01	12.5 16.6	2.4 3.2	2.4 3.2	2.0 2.7

^{*}Means followed by a different letter (a, b, c) show significant differences at 1% level between leaf, stem, and root within a species using Tukey's test.

^{*}Means followed by a different letter (r, s, t) show significant differences at 1% level comparing leaf to leaf, etc. of different species using Tukey's test.

than the leaf material except for creeping bentgrass.

The stem tissue and the roots in particular, are composed of much older tissue than the leaves which have a very rapid turnover under turfgrass conditions of repeated mowing. This partially explains the large difference in cell wall material.

More hemicellulose and cellulose was found in the roots than in the leaves of these three species. For some reason, hemicellulose occurred in greater amounts than cellulose. This is in contradiction with the commonly accepted thesis that cellulose is the most abundant organic compound in living plants. Whether this contrasting result is real or an artifact produced because the technique used did not isolate these two compounds adequately is not known.

The lignin content of the creeping bentgrass,

Kentucky bluegrass and red fescue leaves was 3.7, 2.3,

and 2.7% respectively. Initially, these percentages appear to be very low in comparison to the importance sometimes attributed to clippings in affecting thatch

accumulation. However, there is a rapid turnover of leaf material in turfs mowed regularly. Consequently, lignification would not have a chance to proceed to any significant degree. In addition, some leaf material is found in the surface thatch layer, but almost completely decomposes and disappears in the deeper thatch layers

according to Ledeboer (14) and confirmed by observations of the author. Therefore, it is unlikely that turfgrass clippings make a significant contribution to long-term thatch accumulation.

While the lignin content of stems was higher in all species than in the leaves, the high level of 11.1% lignin in red fescue stems is of great interest. Apparently the papery, fibrous sheath surrounding red fescue stems is quite high in lignin compared to the other two grasses. The persistence of stems in red fescue thatch again correlates with detailed physical observations of the author and of Ledeboer (13). The papery, fibrous sheath and stem tissue of red fescue persisted in a more intact form deeper into the decomposing thatch layer, indicating a resistance to microbial degradation.

Many fine, fibrous roots are observed throughout the thatch layer, but particularly in the upper portion. Some of the larger, primary roots extended into the soil beneath the thatch. After separating the living roots from the soil and thatch, they were found to contain high percentages of lignin--13.9, 9.8, and 12.6% for bentgrass, bluegrass, and red fescue respectively. Lignin is an encrusting substance in cell walls and highly resistant to microbial degradation (5). From the results of this investigation, it would appear that the roots of turfgrasses, along with the stems of red fescue would be the primary contributors to an accumulation of thatch.

Increasing Decomposition of Thatch

Effects of Several Materials on Microorganism Activity

This phase of the investigation was designed to determine whether adding certain materials would induce or stimulate microbial activity and increase decomposition of the thatch. Because of the many factors influencing thatch decomposition under field conditions, it was necessary to conduct these investigations in vitro in order to isolate some of the variables which would otherwise mask or confound the results.

The desired result is the mineralization of thatch or organic matter into simple inorganic elements and compounds via the degradative activities of microorganisms. The activity of microorganisms can be measured by determining the quantity of carbon dioxide evolved from microbial respiration. This was done by maintaining a sealed environment following treatment of the thatch and measuring the composition of a gas sample taken from the test tube after a prescribed period of time. The effects of the addition of four materials on the mineralization of red fescue thatch are shown in Table 4. The data is expressed as the increase of carbon dioxide evolution over controls treated only with water.

The carbon dioxide evolved increases as a higher rate of material is added. The response decreases with

Table 4.--Carbon dioxide evolved from microorganism respiration on red fescue thatch as influenced by pectinase, cellulase, sucrose, and ferulic acid at several rates. Figures represent increase over control treatment which was water plus thatch.

	Rate	Percent CO ₂ accumulated in 24 hours		
Treatment	per tube (mg.)	Day 2	Day 3	Day 7
Pectinase	5 10 25 50 250	0.28 1.06 2.26** 8.35** 4.38** (1.73)†	0.14 0.63 1.58 3.85** 11.50** (2.85)	0.24 0.45 1.03 2.17 9.84** (2.92)
Cellulase	5 10 25 50	0.03 0.60 1.00 2.33** (1.68)	 0.68 1.08 2.78** (1.31)	0.11 0.34 0.86** 1.46** (0.85)
Sucrose	5 10 25 50 250	 0.94 3.83** 7.03** 6.83** (1.83)	0.11 1.12** 3.20** 4.57** 6.24** (0.82)	0.11 1.14 3.29** 6.83** (1.66)
Ferulic Acid	5 10 25 50	0.63 1.74** 1.87** 4.89** (1.13)	0.39 1.65** 2.87** 4.25** (1.60)	 0.17 0.26 2.04** (1.41)

^{**}Significant at 1% level.

[†]Significance values computed by Dunnett's procedure.

time. Where high rates are applied, there is a delay in maximum response which does not occur until the third day. At very low concentrations there is practically no response.

From this data it is apparent that pectinase, sucrose, and ferulic acid produced the greatest response. At very low rates, ferulic acid caused the highest increase in carbon dioxide. Although cellulase and pectinase are both enzymes, pectinase seemed to give a much greater response. This may have been due to a differential in pH requirement of the two enzymes, or to greater stimulation of microorganisms specific for pectic substances in comparison to those stimulated for cellulose which is more resistant to decomposition. Sucrose, also increased microbial respiration in this study. It has been reported that sucrose caused no effect under field conditions when used at 5 and 10 pound rates per 1,000 square feet (13). Organic matter content was used in that study as a measurement of decomposition.

Carbon Source for Microorganisms

In view of the favorable response reported above, the following question must be asked: Is the increase in carbon dioxide due only to the added carbon source, or is there additional carbon utilized from the thatch resulting in increased thatch decomposition? This question may be answered by: (1) Calculating the quantity of carbon

utilized in relation to that added to see whether all the exogenous carbon had been exhausted, and (2) Analysis of total cell wall content following the incubation period to determine whether or not additional decomposition had occurred in comparison to the check treatment.

The first possibility was investigated using sucrose since the quantity of carbon added to the treatment
can be readily calculated. The carbon evolved as carbon
dioxide after one week did not exceed the amount added in
the sucrose, although it was very nearly so for several
rates. Had the experiment been conducted for a longer
period of time, the carbon dioxide may have surpassed the
maximum amount possible from the exogenous carbon.

The results of the total cell wall measurements for the second possibility are listed in Table 5. A greater response was obtained at higher rates. Only a few of the higher rates are listed and only sucrose was significant at the 5% level. This gives substantial proof however that these materials have stimulated microorganism activity in vitro, resulting in additional thatch decomposition in comparison to the control. Whether or not similar results could be obtained in the field over a period of years was beyond the scope of this study. However, it proves the possibility exists and that further investigations are warranted.

Table 5.--Analysis of the total cell wall content to determine the carbon source for microorganism respiration. The red fescue thatch was treated for two weeks.

		Daniel Late 1
Treatment	Rate per tube (mg.)	Percent total cell wall
Check		58.4
Pectinase	250	57.3
Cellulase	10	57.7
Sucrose	250	55.7*
Ferulic Acid	10	57.4

 $[*]LSD_{.05} = 1.7$

Effects of pH on Thatch Decomposition

Many observations have been reported in the literature on the effects of pH in relation to the accumulation and decomposition of thatch. Most of the information merely reports observations and is not the result of carefully controlled experiments. Therefore, the results are probably biased by many other factors. An attempt was made to get more accurate information by measuring the direct effects of buffered solutions on the activity of microorganisms.

The pH levels from three to nine were tested on red fescue thatch and are reported in Table 6. Under the conditions of this experiment, a pH of six was most

desirable followed by five and seven. Also, acidic conditions were definitely superior to an alkaline environment. A carefully controlled field experiment is necessary to further elucidate the effects of pH on thatch decomposition.

Table 6.--Effect of pH on microorganism activity of red fescue thatch as measured by carbon dioxide evolution.

	Percent CO ₂ accumulat	ed in 24 hour period
рН	First day	Second day
3	2.00	5.27
4	1.75	6.62
5	2.46	8.06
6	4.35	10.41
7	3.01	6.13
8	1.30	1.89
9	1.60	1.80

Controlled Environment Chamber Experiment

As a rapid check under more natural conditions, plugs of red fescue thatch were treated in a controlled environment chamber. Treatment applications were made six times per week by injection of the material directly into the thatch layer. At no time during the 6 week period was the turf adversely affected by the treatments. At the

completion of the experiment, measurements were made for:
(1) remaining weight of the thatch samples and (2) total
cell wall determinations, as reported in Table 7.

Table 7.--Averages of eight replications showing the effects of four treatments on red fescue thatch in a controlled environment chamber for 6 weeks.

Treatment	Rate/plug (mg./week)	Average weight (gms.)	Percent total cell wall
Check		45.2	48.9
Pectinase	120	42.7	47.0*
Cellulase	120	43.0	48.2
Sucrose	120	44.3	48.9
Ferulic Acid	120	44.1	46.7*

 $[*]LSD_{.05} = 1.9$

All four treatments caused a loss in total thatch weight, but the decrease was not significant. The figures do indicate a trend in the desired direction, however.

The decrease in percent total cell wall was significant at the 5% level for ferulic acid and pectinase. Ferulic acid, with a decrease of 2% in comparison to the control, gave the best indication of thatch breakdown. No explanation is available for the lack of response from sucrose, except that the rates used may have been too low.

Both the <u>in vitro</u> and controlled environment chamber studies suggest a potential for biological controls of

The addition of the proper material at the proper thatch. rate is very important and needs further study. or not the rate of thatch decomposition with biological controls would be fast enough in the field is not proven by this experiment. However, the rate of microorganism activity in vitro suggests it would be if initiated before a thick layer accumulates. Additional detailed studies are needed on the specific microbial populations involved and their response to the additions of organic stimulants in the field. The effects of pesticides on the macrodecomposers also needs to be investigated. This may be one of the greatest limiting factors in the initial degradative process because of the high levels of pesticides applied to intensively managed turfs. The problem of thatch on golf courses, athletic fields, and lawns in the cool, humid climatic region of the United States dictates that these types of investigations be initiated in the near future.

CONCLUSIONS

- 1. The carbon-nitrogen ratio, although not excessively high, may be a limiting factor in thatch decomposition.
- 2. Lignin content increases in the thatch layer from top to bottom and is higher in red fescue thatch than creeping bentgrass or Kentucky bluegrass thatch.
- 3. Total cell wall content is much higher in the roots of the three turfgrass species than in the leaves which contain more soluble cellular material. Hemicellulose and cellulose in most cases are also considerably higher in the roots than the leaves.
- 4. The lignin content of creeping bentgrass, Kentucky bluegrass, and red fescue roots is three to four times greater than the leaves. A much higher lignin content is found in red fescue stems.
- 5. Chemical measurements of cell wall constituents indicate the clippings of turfgrasses contribute very little to thatch accumulation. This is also confirmed by detailed physical observations.
- 6. Adding pectinase, sucrose, and ferulic acid to thatch increased microorganism activity. Higher rates increased this activity. Total cell wall content determinations indicated increased thatch decomposition occurred.

- 7. A slightly acidic micro-environment resulted in greatest microorganism activity under the laboratory conditions of this investigation.
- 8. A controlled environment chamber study with turf plugs confirmed the possibility of increasing thatch decomposition by adding pectinase and ferulic acid.
- 9. The results of this investigation warrant a field experiment to further investigate the possibilities of thatch control through biological means.

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