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"Fairy Ring" Mushrooms Make Protein-rich Grass[1]

WM. A. ALBRECHT, V. L. SHELDON, WM. G. BLUE*

The greener rings of lawn grass provoked by the "fairy ring" mushroom (*Marasmius oreades*) will always attract attention. This is true whether or not we appreciate and understand all that this phenomenon demonstrates. It is not surprising that the casual observer should suspect the ring of grass of greener color as a case where the nitrogen fertilizer was spread on the lawn irregularly by hand, or that others should believe "the fairy ring is in reality a grass disease" (1).

The opportunity to observe a larger lawn area more critically came as the result of assigned parking space necessitating greater distance of walking. Then with two years, 1948 and 1949, of well-distributed and liberal amounts of rainfall, the fairy rings came into bolder prominence and closer attention. They provided a temptation to sample the deeper green grass of the ring advancing ahead of the mushrooms and some grass not so green and not in the ring.[2] These samples were collected for chemical and more refined biochemical analyses in the latter part of the growing season of 1948 in September, and then again the next year, 1949, in both May and July as earlier and later parts of the season when the fairy rings are commonly observed.

Nitrogen Contents of the Grasses. The chemical analyses of the dried grasses included the measurement of (a) the total nitrogen by the usual Kjeldahl procedure, (b) the amino nitrogen as determined by the Van Slyke method used on the acid hydrolyzed samples, and then (c) the amounts of nine of the ten essential amino acids by means of the microbiologic assay method (2).

The darker green color of grass is always associated with larger amounts of nitrogen in the soil for higher concentrations of it in the grass. The sample from the fairy ring with its more luscious and abundant growth proved the truth of this statement very clearly. It contained an equal or higher concentration of total nitrogen when compared with the non-ring grass, as is shown by the data in Table 1. Only the samples taken in May and July 1949 were tested on this point. It is significant that in May both the ring and the non-ring grasses were of about the same—but yet high—concentrations of nitrogen. Also both had more than 50 per cent of their total nitrogen in the amino form, according to the Van Slyke test.

As feed for grazing livestock, for example, this amount of amino nitrogen content would represent more than 200 pounds of high grade protein per ton of dry matter in May. By July, however, the non-ring grass had apparently increased its carbohydrate content through photosynthesis so much more rapidly than it had carried on its conversion of this compound into protein, that the total nitrogen as percentage was much lower. Likewise, the share of the total nitrogen in the amino

[1] Much of this study was made possible through the help of Swift and Company, Chicago, because of their interest in animal nutrition in relation to the soil.

* Professor, Graduate Assistant and Research Assistant in soils, respectively, College of Agriculture, University of Missouri, Columbia, Missouri.

[2] These samples will be spoken of in this discussion as ring grass and non-ring grass.

form was less than 50 per cent. This non-ring summer grass would contain less than 140 pounds of high grade protein per ton. The ring grass at that later date in the season was much higher in total nitrogen than it was in May. The amino nitrogen by the Van Slyke test in late summer was also a larger percentage of the total nitrogen. This late summer grass as dried forage would contain over 290 pounds of the corresponding high grade protein per ton, according to these chemical analyses.

Changes in Carbon-Nitrogen Ratio of Soil Seem to be Responsible. Since this greener ring grass occurs over the mushroom's mycelia extending themselves outward from the ring center, as shown in Figure 1, there must be higher availability of nitrogen in that soil area for more growth of grass and for more nitrogen concentrated in the grass. This must come about as the result of biochemical changes in the soil provoked by the activities of the mycelia. The more pronounced green growth is demonstrated by its photogenic nature, demonstrated in Figure 2. The differences in biochemical activities in the mycelia-bearing volume of the soil as contrasted with those in the other soil volumes deserve consideration.

TABLE 1. *Nitrogen in the "ring" and "non-ring" grasses growing near the mushroom (Marasmius oreades).*

Date sampled	Sample number	Where sampled	Nitrogen Total %	Nitrogen Van Slyke %	Nitrogen Van Slyke × 100 / Total
May 1949	166–236	Non-ring	3.16	1.73	56.2
	167–237	Ring	3.26	1.74	53.3
July 1949	216–238	Non-ring	2.25	1.09	48.7
	217–239	Ring	4.07	2.36	58.4

When the soil's low supplies of total and available nitrogen are so commonly the limiting factors in the growth of grass, and when the *Marasmius* obtains its energy for growth, not from sunlight directly through photosynthesis, but from carbonaceous organic matter in the soil, there is the suggestion that the higher availability of nitrogen in the soil for better nutrition of the ring grass depends on some particular symbiotic effects between the grass and the mycelia coming through the soil organic matter. These effects are seemingly centered about the changes in the carbon-nitrogen ratio of the soil organic matter and their modification of the activities of other soil microorganisms as brought about by the advancing mushroom mycelia.

Dead roots of bluegrass (*Poa pratensis*) and other vegetable residues constituting the organic matter in an old sod apparently represent too little, and too low a rate, of mineralization of their nitrogen contents for stimulation of the grass to more growth and pronounced green color. They do, however, represent the compounds of wider carbon-nitrogen ratio, like those in decomposing strawy manure serving as mushroom compost. It is these compounds of wider carbon-nitrogen ratio, seemingly, which invite the mycelia of *Marasmius* to "run" through them for its nutrition. Apparently while these advancing mycelia consume the more carbonaceous compounds, they are bringing about a favorable reduction in the carbon-nitrogen ratio of the remaining organic matter so far as this serves as a diet for the other soil microbes. These other microbial life forms subsisting on these materials

FIG. 1. Mushrooms on the inner side of their "fairy ring" of taller and greener grass provoked by the mycelia of this fungus advancing through the soil. Photo by the Missouri Agricultural Experiment Station.

FIG. 2. The taller and deeper green grass provoked by the "fairy ring" mushroom suggests some symbiotic relations between these two operating through the soil. Photo by the Missouri Agricultural Experiment Station.

of narrowed carbon-nitrogen ratio for their energy, mineralize nitrogen to higher concentration of it in the soluble forms in the soil. Without this delivery of available nitrogen the larger grass growth of a darker green color would not be possible.

Ratio of Carbon to Other Soil Fertility Elements Involved in the Suite of Amino Acids in Grass. Growth or cell multiplication always involves protein. Life is never found flowing from carbohydrate to carbohydrate molecules, nor from fat to fat. More growth of grass suggests delivery by the soil, as modified by the mycelial effects, of other essential elements of nutrition beside the nitrogen. The elements calcium, magnesium, phosphorus and others, including the trace elements, are required for protein-producing plants like legumes, for example. It is interesting to note the suggestion coming from the higher amino acid contents of the ring grass, as shown in Table 2, that the mycelia "running" through the soil must also be making more available for the grass some other nutrient elements beside nitrogen. This extra amount of these other elements suggests itself as help, in addition to the help by the nitrogen, for the plants' biosynthetic conversion of carbohydrates into proteins. Such conversions are basic in bringing about the higher concentrations of amino acids (and thereby also greater yields per acre) of the grass, especially during the later part of the season.

Even though microbiologic assays of the bluegrass were made for only nine essential amino acids, yet in every case of these for the two later seasons combined, that is September 1948 and July 1949, the comparison of the ring with the non-ring grasses shows a higher concentration for each acid in the grass modified by the effects of the *Marasmius* mycelia growing beneath it. For the grasses in May and for the eight essential amino acids assayed for that date, in only three of them is the grass in the ring higher. The differences in favor of non-ring grass in this early season are not as large as they are in favor of the ring grass in the later seasons.

Here is the suggestion that nitrogen is too slowly available in May for the conversion of the carbohydrates into proteins quickly to prevent a lower concentration of the latter in May. Several limiting factors, including possibly other soil fertility elements beside nitrogen are holding down carbohydrates for growth of non-ring grass and also for the conversion of carbohydrates into proteins in the ring grass making the larger growth. The ratio of carbon to fertility elements other than nitrogen very probably comes into the picture. This can be true even though we have not recognized these ratios as commonly as we have the carbon-nitrogen ratio.

Not only the seasonal variation in the amino acids, but also the specific amounts of certain ones may deserve special attention. Unfortunately, our knowledge of the amount of each of these as a specific growth factor is still too meager to judge them quantitatively. It is interesting to note the suggested higher concentration of tryptophane in the ring grass for the comparison of the two ring and non-ring locations. This is significant when one recalls that tryptophane is commonly too deficient in the grain of maize to serve for good growth of animals in winter, while green grass, in contrast, is most excellent feed in spring. With the higher mineralization of nitrogen within the soil under the ring grass suggesting itself as a procedure for increasing the concentration of this forage's tryptophane content, the question naturally presents itself whether fertilization of pasture grasses with only a mineral nitrogen treatment would also be an effective means of increasing its tryptophane. Even if other fertility elements are needed for this, it still suggests the possibility of properly fertilized grass, quickly dried, as a feed supplement to corn grain to

TABLE 2. *Some amino acid contents of "ring" and "non-ring" grasses growing near the mushroom (Marasmius oreades).*

Date sampled	Sample number	Where sampled	Amino acid contents (mgms./gm. dry matter)								
			Valine	Leucine	Iso-leucine	Threo-nine	Trypto-phane	Histo-dine	Lysine	Argi-nine	Methio-nine
Sept. 1948	92–234	Non-ring	14.7	18.8	31.9	12.1	.........*	6.0	26.0	....	4.3
	93–235	Ring	19.8	23.4	36.2	16.4	....	6.5	28.3	....	4.7
May 1949	166–236	Non-ring	11.8	11.3	17.7	11.4	2.13	8.1	12.2	4.0	....
	167–237	Ring	12.0	13.6	15.9	8.9	2.41	8.1	10.2	3.1	....
July 1949	216–238	Non-ring	11.6	11.2	....	5.6	1.50	2.8	....	2.3	5.0
	217–239	Ring	....	19.6	....	11.7	2.29	4.1	....	7.6	6.9

* Not determined.

balance the deficiency of the latter with respect to this essential amino acid. Of course, the above would be a considered procedure for practice only after it is discovered that such soil treatments under the corn do not serve to increase the tryptophane content of this grain itself.

Since threonine was the last one added to the list of ten essential amino acids, there may be some significance in its higher concentrations in the ring grass in the two determinations on the grass collected in the later seasons, September 1948 and July 1949. The same situation holds for methionine, a sulphur-carrying amino acid. The possible significance of these observations in relation to the growth by animals feeding on the grass remains to be discovered.

Mushroom-Grass Combination, a Possible Symbiosis. The "fairy ring," in terms of the biochemical processes in the soil beneath it, suggests some quite interdependent and intricately balanced relations. They all are parts of what suggests a symbiotic complex including (a) the production in the soil of particular organic compounds represented by the dead grass roots, (b) their specific nutritional services at certain seasons in producing the mushrooms, and (c) the accompanying modification of that soil-organic matter complex, apparently through mineralization of it by other soil microbes, for better nutrition of the grass, at least with respect to nitrogen and possibly other nutrient elements. It suggests that ash analyses, such as total nitrogen ignited in sulfuric acid, have been insufficient tools to give us better understanding of the soil's services in plant nutrition, much less in animal nutrition thereby. It suggests that additional sciences, such as biochemistry, thermodynamics and others, need to be called in before we can understand the processes by which the plants synthesize the many complex chemical compounds composing them, or the physiological interdependence of the different forms of vegetation in a single location. These other scientific helps need to come in for wider use even before we understand fully how possibly the changing ratio of carbon to other fertility elements in the soil is involved in the "fairy ring" as it makes not only more and greener grass, but more protein-rich grass, through the apparent symbiotic performances involved.—UNIVERSITY OF MISSOURI, COLUMBIA, MISSOURI.

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