# Technology Section

## SYMBIOTIC BIODEGRADATION OF CROP RESIDUES Part I. Biodegradation of Wheat and Rice Straw

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The effect of pre-treatment and reduction in particle size of straws on its microbial digestibility has been investigated. Biodegradation of rice straw was 48.35 % and 69.93 % when *Penicillium* and *B. polymyxa* were employed singly. The combination of *B. polymyxa* with *Penicillium* showed an imp-rovement in the degradability of rice straw upto 71.67 %. The digestibility of cellulose present in wheat straw increased by 54.85 % by symbiotic effect of *B. Cereus* and *Chaetomium*. Reduction in particle size improved the susceptibility to microorganisms.

#### **INTRODUCTION**

Crop residues represent one of the largest potential resources for future energy and food. Their nutritive value. can be increased by growing cellulase producing fungi on them. Recently a large number of rumen and soil micro-organisms have been found which possess the ability to degrade native cellulose into simpler carbohydrates [1-4). Methods for enzymic hydrolysis of cellulose to glucose especially by the cellulase produced by Trichoderma have also been developed [5-9]. Degradation of cellulose has been improved by using mixture of celluloytic microorganisms than individual enzymes due to their synergistic action [10]. A decrease in particle size increased susceptibility of the substrate to the enzymic hydrolysis because of the increase in surface area [11].

#### MATERIALS AND METHODS

Wheat and rice straws were purchased from the local market and were ground to 60, 80, 100, 120 and 200 mesh.

Reese medium[12] containing 0.5 % of wheat or rice straw was fermented with molds and bacteria. The fermented residue was centrifuged at 1185g for 10 min. The precipitate was used for the estimation of undigested cellulose and percentage increase in nitrogen.

Analytical methods for the estimation of nitrogen and cellulose were the same as reported elsewhere [13,14].

#### **RESULTS AND DISCUSSION**

The Effect of Particle Size on the Growth of Micro-

organisms. Effect of particle size on the biodegradation of wheat straw was first studied with the aim of selecting a mesh size most suitable for the purpose. Accordingly wheat straw was ground from 60 to 200 mesh sizes and biodegradation of the material by *Penicillium*, *Chaetomium*, *Streptomyces*, *Trichoderma* and their combinations was studied. It was noted that particle size has an effect both on the degradation of cellulose and increase in nitrogen content. This effect was most conspicuous from 100-200 mesh.

At 200 mesh, *Chaetomium* degraded cellulose (41.26%) while a combinations of *Chaetomium* and *Trichoderma* or *Chaetomium* and *Streptomyces* showed maximum degra-dation (50.29%).

Increase in nitrogen content was also maximum at 200 mesh. Thus *Trichoderma* alone and a combination of *Streptomyces* and *Trichoderma* increased nitrogen content upto 73.01%, and 89.68% respectively.

These observations are in agreement with the findings of other workers [9,11] that by reducing the particle size more surface area is exposed which made the substrate more accessible to the microbial enzymes.

Since grinding of cellulosic material to 200 mesh posed operational problems it was decided to use 100 mesh powder throught out the present studies.

Biodegradation of Straws with Single Strain of Molds, Bacteria and Yeast. The effect of molds, bacteria and yeast on biodegradation of ground straws is shown in Table 1. Maximum biodegradation of cellulose 69.93 % was observed when rice straw was incubated with Bacillus polymyxa. Next in line was Saccharomyces cerevisiae which degraded 50 % cellulose of rice straw and 35.92 % that of wheat straws. The increase in nitrogen content was 87.06 %. Pencillium showed 48.35 % improvement in cellulose degradation and 152.58 % increase in nitrogen contents. Biodegradation of rice straw and wheat straw with *Saccharomyces cerevisiae* was 50.0 % and 35.92 % respectively. Digestibility of various crop residues with cellulose degrading bacteria was reported by various work-ers[15, 16, 17].

Symbiotic Biodegradation of Straws with Molds, Bacteria and Yeast. Biodegradation of rice and wheat straws due to combined effect of molds, bacteria and yeast is given in Table 2. It is evident that 47.2% of the cellulose present in rice straw was biodegraded due to symbiotic effect of Chaetomium and Streptomyces. The increase in nitrogen contents was upto 147.4%. The combination of Chaetomium and Penicillium was also found to be effective in case of wheat straw. The increase in nitrogen in case of wheat straw was 64.51% which was associated with 44.17% degradation of cellulose. Similar results were reported by Peitersen[8] using mixed cultures of cellulolytic fungi.

Symbiosis of *B. polymyxa* with *Streptomyces* degraded 75.85 % of the cellulose present in rice straw with 62.06 % increase in nitrogen content. An increase in nitrogen contents up to 130.17 % and 52.41 % cellulose degradation was observed when *S. cerevisae* was propagated in combination with *Penicillium* on rice straw. Surender and Gupta[16] reported good growth of *Saccharomyces cerevisiae* on urea enriched hydrolysate of cellulosic material. Peitersen[8] while using *S. cereviciae* in combination with *T. viride* reported rapid utilization of glucose and production of cellulase. Symboitic effect of *Pencillium* with *B. cereus*, *B. polymyxa*, *B. sphaericus*, and *B. subtilis* on rice straw resulted in an increased nitrogen contents by 92.24, 70.68, 97.41, 87.06 and 130.07 % respectively.

In wheat straw, maximum biodegradation of cellulose (54.85 %) was observed due to symbiotic effect of B.

	Table 1	1. Biodegradation	of crop	residues by	molds and	bacteria
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	Rice straw		Wheat straw	
Cultures	Degradation of cellulose % age	Increase in nitrogen % age	Degradation of cellulose % age	Increase in nitrogen % age
Pencillium	48.35	152.58	17.99	61.20
Chaetomium	36.36	193.10	26.01	60.48
Streptomyces	34.26	87.06	20.07	42.74
Trichoderma	40.20	172.41	26.69	73.38
Bacillus brevis	43.00	27.58	18.44	31.69
Bacillus cereus	31.46	18.96	12.62	4.83
Bacillus laterosphorous	6.29	91.37	13.59	11.29
Bacillus pumilus	34.26	72.41	31.55	8.06
Bacillus polymyxa	69.93	87.06	5.82	8.87
Bacillus spherieus	33.91	72.41	19.14	16.12
Bacillus subtilis	39.16	60.34	13.59	10.48
Saccharomyces cerevisiae	50.00	18.96	35.92	45.16

Table 2. Symbiotic biodegradation of crop residues by molds bacteria and yeast.

	Rice straw		Wheat straw	
Cultures	Degradation of cellulose % age	Increase in nitrogen % age	Degradation of cellulose % age	Increase in nitrogen % age
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Chaetomium + Trichoderma	36,36	123.41	35.04	67.74
Trichoderma + Streptomyces	46.50	131.03	24.03	83.87
Streptomyces + Penicillium	43.70	185.35	20.35	82.25
Penicillium + Chaetomium	46.15	77.58	44.17	64.51
Trichoderma + Penicillium	25.17	82.75	30.87	66.93
Streptomyces + Chaetomium	47.20	147.41	31.84	67.74
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	43.00	27.58	18.44	31.69
Bacillus brevis + Chaetomium	49.30	39.79	33.99	4.03
Bacillus brevis + Trichoderma	52.00	44.82	32.03	64.51
Bacillus brevis + Streptomyces	56.99	25.86	9.22	33.06
Bacillus brevis + Penicillium	58.39	40.51	15.53	28.22
	31.46	18.96	12.62	4.83
Bacillus cereus + Chaetomium	38.81	8.62	54.85	107.25
Bacillus cereus + Trichoderma	32.86	72.41	33.00	67.74
Bacillus cereus + Streptomyces	42.30	38.76	33.49	29.03
Bacillus cereus + Penicillium	45.80	92.24	22.33	54.03
C				
	6.29	91.37	13.59	11.29
Bacillus laterosporus + Chaetomium	51.04	80.17	18.44	50.00
Bacillus laterosporus + Trichoderma	40.90	81.89	22.33	7.25
Bacillus laterosporus + Streptomyces	34.26	97.41	48.05	61.29
Bacillus laterosporus + Penicillium	39.51	73.27	48.54	34.67
	34.26	72.41	31.35	8.06
Bacillus pumilus + Chaetomium	45.80	124.13	22.33	12.90
Bacillus pumilus + Trichoderma	39.51	81.89	13.10	61.29
Bacillus pumilus + Streptomyces	42.30	80.17	38.34	8.87
Bacillus pumilus + Penicillium	54.54	98.27	48.54	6.45
D				
	69.93	87.06	5.82	8.87
Bacillus polymyxa + Chaetomium	57.34	44.82	50.00	64.51
Bacillus plymyxa + Trichoderma	71.67	62.06	26.69	93.57
Bacillus polymyxa + Streptomyces	75.87	62.06	23.30	78.22
Bacillus polymyxa + Penicillium	71.67	70.68	31.55	101.61
Ducanus porynnynu · 1 chicanun	33.91	92.82	19.41	16.12
Bacillus sphaericus + Chaetomium	35.66	76.72	26.21	41.12
Bacillus sphaericus + Trichoderma	33.91	94.82	27.72	46.77
Bacillus sphaericus + Streptomyces	53.14	72.41	41.74	13.70
Bacillus sphaericus + Penicillium	31.46	97.41	10.19	25.00
Е				
	39.16	60.34	13.59	10.48
Bacillus subtilis + Chaetomium	40.90	1.17	38.34	7.25
Bacillus subtilis + Trichoderma	41.25	72.41	26.25	58.06
	52.44	51.72		
Bacillus subtilis + Streptomyces			29.12	6.45
Bacillus subtilis + Penicillium	45.10	87.06	26.21	8.87
	50.00	18.96	35.92	45.16
Saccharomyces cerevisiae + Chaetomium	41.25	45.68	47.57	52.41
Saccharomyces cerevisiae + Trichoderma	40.50	53.44	33.00	37.90
Saccharomyces cerevisiae + Streptomyces	46.15	41.37	41.74	51.61
Saccharomyces cerevisiae + Penicillium	52.44	130.17	41.26	24.19

cereus and Chaetomium. This was accompanied with 107.25 % increase in nitrogen contents. These results clearly indicated that combinations of Chaetomium with B. brevis, B. polymyxa, B. subtilis and S. cereviciae was more effective than the others. These findings are in accord with that of Han, et al [10] and Peitersen[8] who reported that bacteria in combination with fungus and yeast utilized more cellulose as compared with fungus alone. Improvement in the biodegradation of cellulose with combinations of bacteria and molds than invididual microorganisms is due to the fact that pure culture of single organism had a fixed genetic make up, its physiological capacity and rate of degradation of cellulose is limited but these limitations were overcome by using the combination of bacteria and yeast with molds in the fermentation process.

#### REFERENCES

- H.O.W. Eggins, K.A. Malik, and R.F. Sharp "Some techniques to investigates the celonilization of cellulosic and wood substractes" Proc. of the Inst. Int. Biodeterioration. Symp., 121 (1968).
- 2. H.O.W. Eggins and A.O. Lloyer, Experientia, 27, 749 (1968).
- 3. J.K. Gupta, Y.P. Gupta, and N.B. Das, Agr. Biol.

Chem., 37, 2657 (1973).

- Y.W. Han and V.R. Srimivasan, Appl. Microbiol., 16, 1140 (1968).
- 5. N. Toyama and K. Ogawa, Proc. IVIFS Ferment. Technol. Today, 743 (1972).
- D. Braudt, L. Hontz and M. Maudels AICHE Symp. Ser, 69 (No. 133), 127 (1973).
- 7. W.D. Bellamy, Biotechnol. Bioeng, 16, 869 (1974).
- 8. N. Peitersen, Biotechnol. Buoeng., 17, 1291 (1975).
- Y.W. Han and C.D. Callihan, Appl. Microbiol., 27, 159 (1974).
- 10. Y.W. Han, C.E. Dunlap and C.D. Callihan, Food Techn., 25, 32 (1971).
- E.W. More, J.M. Effland, J. Agr. Food Chem., 20, 1173 (1972). C.A. 28319v vol. 78 (1973).
- 12. H.S. Levinson, G.R. Mandela and E.T. Reese, Arch Biochem. Biophys, 31, 351 (1951).
- 13. R. Markham, Biochem. J., 36, 790 (1942).
- K. Kurschner, and A. Hanak, Z. Untersuch, Lebensun, 59,484 (1930).
- 15. D.M. Undegraff, Biotech. Bioeng., 13, 77 (1971).
- D. Surinder and J.K. Gupta, J. Gen. Appl. Microbiol, 23, 155 (1977).
- 17. Y.W. Han and A.W. Anderson, Appl. Microbiol., 30, 930 (1975).