Studies on DNA modification in Oscillatoria sp. MKU 178

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Cyanobacterial DNA are generally believed to be resistant to restriction by endonucleases due to extensive modification of the DNA. We report here the absence of a dam-mediated modification in a cyanobacterium, Oscillatoria sp. MKU 178.

Restriction and modification systems of DNA have been well documented in several organisms and the roles which have been postulated for the same include, protection of cells against invasion by foreign DNA, replication and repair of DNA and control of gene expression. However, as the type of restriction/modification is distinct for each organism, the worker interested in using an organism for recombinant DNA work, needs a thorough understanding of its restriction/modification system to be successful in his venture. Our laboratory has been working on the genetic and physiological aspects of cyanobacteria and our present study is aimed at studying the DNA modification in cyanobacteria. We took for our study, Oscillatoria sp., a non-heterocystous cyanobacterium, which exists in a wide range of habitats like streams, ponds, lakes, irrigation canals, rice fields and even sewage and industrial wastewaters; besides, it is useful to agricultural lands because of its soil-binding property and polysaccharide production. These properties make it an ideal choice for the introduction and expression of other agronomically important traits such as production of plant growth regulators and insecticides for its better use in agriculture.

Earlier workers have reported that DNA from several cyanobacterial species is resistant to cleavage by endonucleases. In the course of our investigation on the resistance of cyanobacterial DNA to hydrolysis by restriction enzymes we identified one isolate, Oscillatoria sp. MKU 178, which was susceptible to cleavage by EcoR1. We took up a more detailed study in this isolate.

A fifteen-day-old culture of the strain, grown under a light/dark cycle of 16h/8h, at 25°C, was harvested for DNA extraction. DNA was purified following Sambrook et al. The DNA was treated with Dpn1, Mbo1 and Sau3A enzymes (New England Biolabs, USA). The tubes were incubated at 37°C for 1h and then electrophoresed on a 0.7% agarose gel. The DNA was visualized on a UV transilluminator. As can be seen from Figure 1, Dpn1 failed to cut the DNA (lane 3), while Mbo1 produced a streak showing partial digestion (lane 5) and Sau3A totally digested the DNA (lane 7). The corresponding controls showed no digestion. The above three enzymes are isoschizomers recognizing the sequence GATC. However, the ability to cut depends on the nature of DNA methylation. The action of Dpn1 and Mbo1 is mutually exclusive because Dpn1 cuts the DNA if the adenine residues are

Received 15 July 1992; revised accepted 14 October 1992
methylated in both the strands, whereas \textit{MboI} cuts only if the adenine is totally unmodified. \textit{Sau}3A cuts irrespective of adenine modification, but in the absence of cytidine modification. From our results it is clear that DNA of this organism lacks modification of both adenine and cytidine residues in the GATC sequences. Reports regarding the unusual resistance of cyanobacterial DNA to restriction endonucleases have proposed the involvement of a deoxy-adenosine methylase (Dam) enzyme (Dam methylates the adenine in a GATC sequence) in DNA modification\textsuperscript{6,7}. The results obtained in the present study prompt us to postulate that a \textit{dam}-like system of modification is absent in this cyanobacterium, \textit{Oscillatoria} sp. MKU 178.


ACKNOWLEDGEMENTS. We thank Prof. G. S. Venkataraman for his constant encouragement. K. S. S. was a recipient of a CSIR Senior Research Fellowship.

Received 28 September 1992; accepted 9 October 1992

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\textbf{Apiospora camptospora — A new fungus causing stalk rot on maize in India}

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We observed stalk rot of maize (\textit{Zea mays} L.) during regular survey of diseases. One of the pathogens responsible for disease was identified as \textit{Apiospora camptospora} Penz and Sacc., a new fungus on maize in India.

During the regular survey of diseases, the maize culture Manjri composite was found to show symptoms which resembled stalk rot at the All India Co-ordinated Maize Improvement Project of the Mahatma Phule Agricultural University, Rahuri.

The symptoms are similar to stalk rot caused by \textit{Fusarium}. The lower leaves became flaccid, wilted and rolled inwardly, while upper leaves of such plants became pale green and subsequently the whole leaf sheath becomes chlorotic. The lower internodes developed purple to brownish discoloration. In completely wilted plants, the pith became hollow and developed a pinkish to dirty brown colour. In culture, the fungus is whitish in colour with irregular, patchy growth. Sporodochia appeared ellipsoidal or elongate with black conidiophores. Mother cells measured in the range of 8–12 × 4–7 \textmu m, conidiophores often pale, brown, up to 140 \textmu m long, 3–4 \textmu m thick, conidia round or polygonal in the face view, 20–32 \textmu m diameter, 14–15 \textmu m thick, mid to dark brown with a distinct hyaline, rim or germ.