The sensitivity of DNA cleavage by SpeI and ApaLI to methylation by M.EcoK

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During work on site-directed mutagenesis of the human interleukin-2 gene an EcoK site (1) was created which overlapped recognition sequences for SpeI (1) and ApaLI (1) (Fig. 1a). Isolation of the DNA from m+ strain DH1 (2) and m– strain HB101 (3) and subsequent incubation with the two restriction endonucleases revealed that EcoK methylation completely or almost completely protected both DNA strands from cleavage by SpeI, but did not prevent cleavage of either strand by ApaLI (Fig. 1b). Thus, methylation of only one of the 5'-terminal A's of the SpeI site is sufficient to protect it against SpeI, whereas methylation of one of the two A's of the ApaLI sequence does not interfere with its cleavage by ApaLI.

REFERENCES