

# Structural and functional studies on Vc-YaeO a Rho inhibitor.

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Efficient and accurate transcription termination is required for the correct regulation of bacterial gene expression. Transcription termination is the process by which a nascent RNA is released from its complex with RNA polymerase and DNA template. In bacteria, two main mechanisms of transcription termination have been described. These mechanisms, commonly referred to as Rho-independent and Rho-dependent termination, are essential for the regulation of bacterial gene expression. Rho-independent termination occurs at a GC-rich self-complementary region that forms a stem-loop structure believed to cause the RNA polymerase to pause, allowing the release of the RNA. Rho-dependent termination, on the other hand, requires the presence of a hexameric helicase, Rho. Rho is an essential transcription factor that binds nucleic acids at specific termination sites (rut) and translocates along the RNA until it reaches the transcription complex. There, it facilitates the termination by unwinding RNA/DNA heteroduplexes upon hydrolysis of ATP.

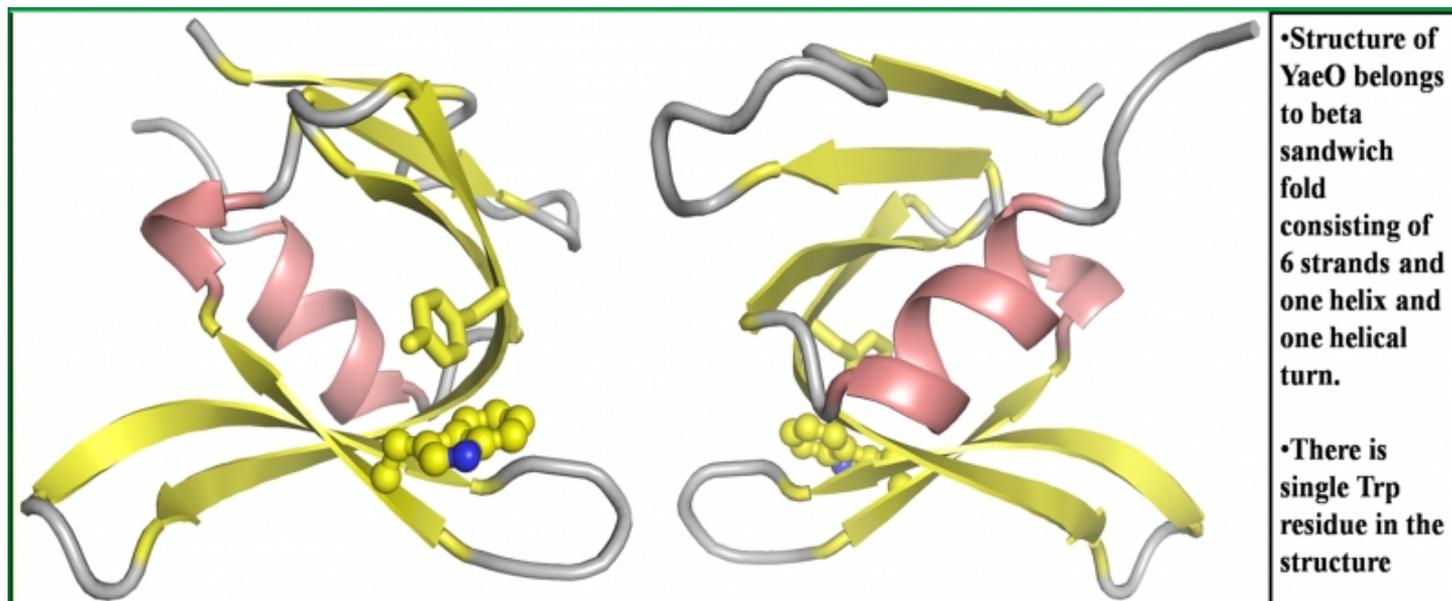
Currently, only two Rho-specific inhibitors of transcription termination have been reported. The first to be described is a 21.3-kDa protein encoded by gene *so* of the satellite bacteriophage P4 and which structure has been solved (1). The second inhibitor is the product of gene *yaeO* from *Escherichia coli*, which has been shown to reduce termination of both the Rho-dependent bacteriophage terminator tL1 and autogenously regulated gene *rho* (2). Overexpressed *yaeO* also suppressed some temperature-sensitive mutations in division genes *ftsA* and *ftsQ*, in chaperone gene *groEL* and in co-chaperone gene *grpE*. *E. coli* homolog of *Vibrio cholerae* O395 YaeO (Vc-YaeO) is an RNase P-like protein whose functional characteristics and exact nature of the interaction with Rho has not been elaborately studied.

We have solved the crystal structure of the YaeO from *Vibrio cholerae* O395 (Vc-YaeO) at atomic resolution (1.8Å) and quantified the interaction with N-terminal part of Rho. Structural similarity of Vc-YaeO with HFQ proteins and RNase P-like protein indicated by DALI search promoted us to decipher the interaction of Vc-YaeO with NTPs and oligonucleotides, through intrinsic fluorescence quenching assay as the protein has only one *trp* residue. Significant interaction with oligonucleotides and NTPs unlike its *E. coli* homolog (3) proves that it interact nearer to *trp* residue. A further revelation of its RNase P-like protein property or HFQ-like RNA chaperone and transporter property would reveal its novel multitasking functions in addition to its Rho-dependent anti-termination property.

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**Keywords:** [Vc-YaeO](#), [Transcription anti-termination](#), [fluorescence quenching](#)