INHIBITION OF THE CRISPR-CAS SYSTEM BY AN ANTI-CRISPRPROTEIN

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The ongoing co-evolutionary arms race between bacteria and bacteriophages has led to the development of various defense and attack strategies. One of the bacterial defense systems against phages is CRISPR-Cas. This system detects and integrates invading DNA into the CRISPR array. The transcript of the CRISPR array is processed into small crRNA molecules, which together with Cas proteins form ribonucleoprotein effector complexes. Guided by crRNAs, these complexes recognize and cleave complementary phage DNA, thereby providing adaptive immunity [1]. To overcome CRISPR-Cas–mediated defense, bacteriophages have evolved anti-CRISPR (Acr) proteins that inhibit CRISPR-Cas activity, most commonly by preventing target DNA binding or nuclease function [2]. Beyond their biological relevance, Acr proteins have attracted considerable interest as regulators of CRISPR-Cas–based genome editing technologies [3,4]. Despite the rapid expansion in the number of identified Acr proteins, detailed molecular mechanisms of inhibition have been elucidated for only a small subset [4].

In this study, we analyse the inhibition mechanism of a previously uncharacterized AcrIF protein targeting the type I-F CRISPR-Cas system. *In vivo* activity assays demonstrate that this AcrIF protein inhibits the CRISPR ribonucleoprotein effector complex. The protein was purified using liquid chromatography, and its inhibitory activity was assessed through a series of biochemical assays. Our current data indicates a unique mode of action for this AcrIF protein, the molecular basis of which we aim to elucidate.

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