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Abstract

Actinomycetes are a group of Gram-positive bacteria of which many representatives are prominent for being prolific producers of bioactive natural products including antibiotics, fungicides, antitumor agents, or immunosuppressants. *Streptomyces* antibiotic regulatory protein (SARP) family of transcriptional regulators are widely distributed among actinomycetes, especially in streptomycetes and are known to activate antibiotic biosynthesis. The set of genes responsible for the production of natural products, including pathway-specific transcriptional regulators like SARPs, are typically located in contiguous regions of the genome known as "biosynthetic gene clusters" (BGCs). The aim of this study was to activate expression of antibiotic BGCs in selected actinomycetes strains upon heterologous expression of the SARP-type regulator PapR2 from *Streptomyces pristinaespiralis*. Here, it was shown that PapR2 activates the undecylprodigiosin (Red) BGC in *Streptomyces coelicolor* A3(2) and thereby substitutes for RedD, the native SARP regulator of Red biosynthesis. In this study, overexpression of *papR2* increased the production of predominantly unknown antimicrobial compounds in more than half of the selected actinomycetes strains, as observed by bioassays against different test strains including bacteria and fungi. For strain *Streptomyces* sp. TÜ4106 it was found that *papR2* expression is associated with increased bioactivity and the production of a so far unknown blue pigmented substance. In summary, this study confirmed that activation of antibiotic BGCs can be successfully achieved by heterologous expression of SARP family regulators, thereby representing a powerful approach for novel bioactive natural product discovery.

Zusammenfassung

Aktinomyceten sind Gram-positive Bakterien, von denen mehrere Vertreter dafür bekannt sind eine Vielzahl von bioaktiven Naturstoffen zu produzieren, wie z. B. Antibiotika, Fungizide, antitumorale Agenzien oder Immunsuppressiva. Transkriptionsfaktoren aus der Familie der "*Streptomyces* antibiotic regulatory protein" (SARP) sind unter den Aktinomyceten, insbesondere bei den Streptomyceten, weit verbreitet und aktivieren bekanntermaßen die Antibiotika-Biosynthese. Die für die Antibiotika-Biosynthese verantwortlichen Gene, darunter auch die Gene, die für SARPs kodieren, liegen in der Regel gruppiert in spezifischen Regionen des Genoms vor, den sogenannten "Biosynthese-Genclustern" (BGCs). Ziel dieser Arbeit war es, die Antibiotika-Biosynthese in ausgewählten Aktinomyceten Stämmen durch heterologe Expression des SARP-Regulators PapR2 aus *Streptomyces pristinaespiralis* zu aktivieren. Es konnte gezeigt werden, dass PapR2 spezifisch das Undecylprodigiosin (Red)-BGC in *Streptomyces coelicolor* A3(2) aktiviert und damit RedD, den nativen SARP-Regulator der Red-Biosynthese, ersetzt. In der vorliegenden Arbeit wurde gezeigt, dass die Expression von *papR2* die Produktion von überwiegend unbekannten antimikrobiellen Substanzen in mehr als der Hälfte der ausgewählten Aktinomyceten Stämmen aktiviert, was durch Bioassays mit verschiedenen bakteriellen und pilzlichen Teststämmen festgestellt werden konnte. Für den Stamm *Streptomyces* sp. TÜ4106 wurde infolge der Expression von *papR2* eine erhöhte Bioaktivität verbunden mit der Produktion einer bisher unbekannten, blau pigmentierten Substanz nachgewiesen. Zusammenfassend wurde in dieser Studie gezeigt, dass Antibiotika-BGCs erfolgreich durch heterologe Expression von SARP-Transkriptionsfaktoren aktiviert werden können. Dies stellt einen leistungsstarken Ansatz für die Entdeckung neuer bioaktiver Naturstoffe dar.

Contents