Signaling Cascades of Antibiotic Biosynthesis in Actinomycetes

Takuya Nihira

International Center for Biotechnology, Osaka University
2-1 Yamadaoka, Suita, Osaka 565-0871
e-mail:nihira@icb.osaka-u.ac.jp

It is well known that members of Actinomycetes produce a great many antibiotics and other classes of biologically active secondary metabolites. Actinomycetes make some two-thirds of the known antibiotics that are produced by microorganisms, and amongst them nearly 80% are made by members of the genus *Streptomyces*, with other genera trailing numerically. If secondary metabolites with biological activities other than anti-microbial are included, actinomycetes are still out in front, with over 60% in which *Streptomyces* spp. again account for 80% of these.

Antibiotic production in streptomycetes is generally growth phase-dependent. In liquid culture, it begins as the culture enters stationary phase. In agar-grown cultures, it coincides with the onset of morphological differentiation, and the occurrence of mutants deficient in both antibiotic production and the formation of aerial hyphae indicates at least some common elements of genetic control. Most antibiotics are the products of complex biosynthetic pathways, with a cluster of genes (generally 20-30) dedicated to the synthesis of any one compound. These gene clusters usually contain pathway-specific regulatory genes which act as transcriptional activators, and which are themselves subject to control by pleiotroic regulatory genes.

Streptomyces species possess characteristic low-molecular-weight signal compounds named (-butyrolactone autoregulators which mainly regulate onset of secondary metabolism/biosynthesis of antibiotics. The (-butyrolactone autoregulators are *Streptomyces* hormones having a common 3-hydroxymethyl butanolide skeleton. Based on minor but essential structural differences C-2 side chains, the 11 (-butyrolactone autoregulators so far identified can be classified into three groups: i) virginiae butanolide (VB)-type possessing 6S-hydroxyl group, ii) IM-2-type possessing

6R-hydroxyl group, and iii) A-factor-type possessing 6-keto group. The VB signal in *S. virginiae* is transmitted into the cells via binding to a VB specific receptor, BarA, and the IM-2 signal in *S. lavendulae* is transmitted via binding to an IM-2 specific receptor, FarA. Several lines of evidence derived from gel-shift assays, surface-plasmon-resonance analyses, DNase I footprinting and S1 nuclease analyses have revealed that, in the absence of an autoregulator, the autoregulator receptors act as a transcriptional repressor by binding to specific DNA sequences overlapping the -10 to -35 region of target genes. The binding was usually abolished by binding with autoregulators, which resulted in transcriptional derepression of target genes. Several target genes which participate in the control of antibiotic biosynthesis have been found under the control of receptors. Using the VB-BarA system of *S. virginiae* in controlling the virginiamycin biosynthesis as the model case, signal transduction pathway/mechanism of antibiotic biosynthesis will be presented.