Aflatoxin biosynthesis and regulation

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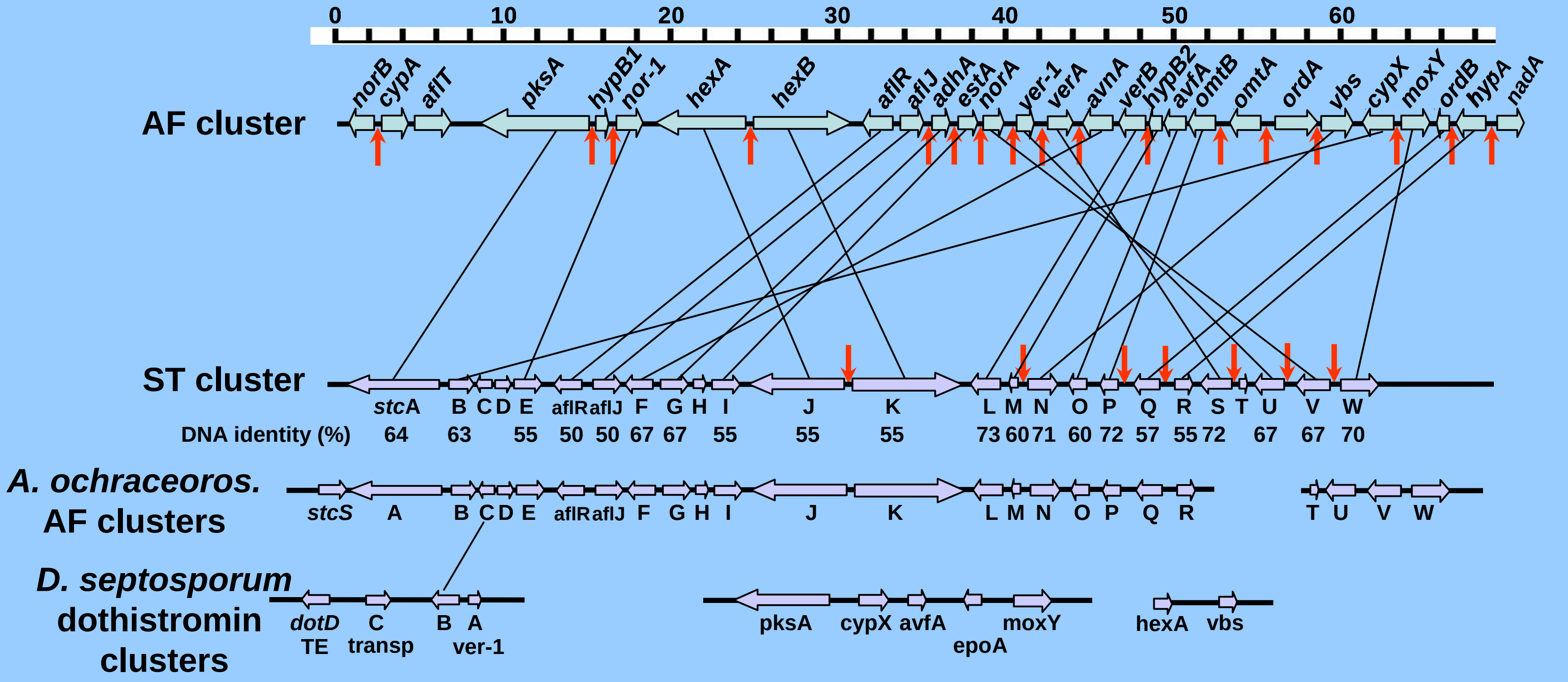
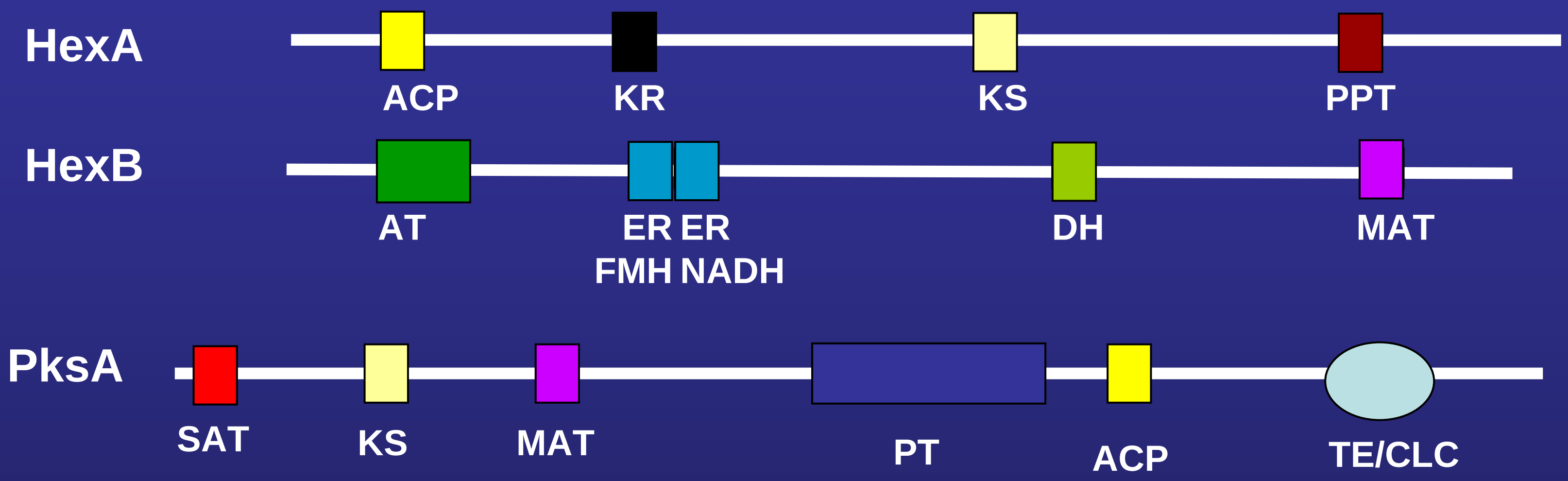
Introduction

- AF/ST biosynthesis involves expression of at least 25 genes
- Genes are organized in a cluster or clusters
- Genes are coordinately regulated
- The pathway-specific transcription factor, AflR, and globally acting transcription factors regulate biosynthesis
- Regulation of biosynthesis is different in different AF-producing species

Comparison of AF/ST clusters

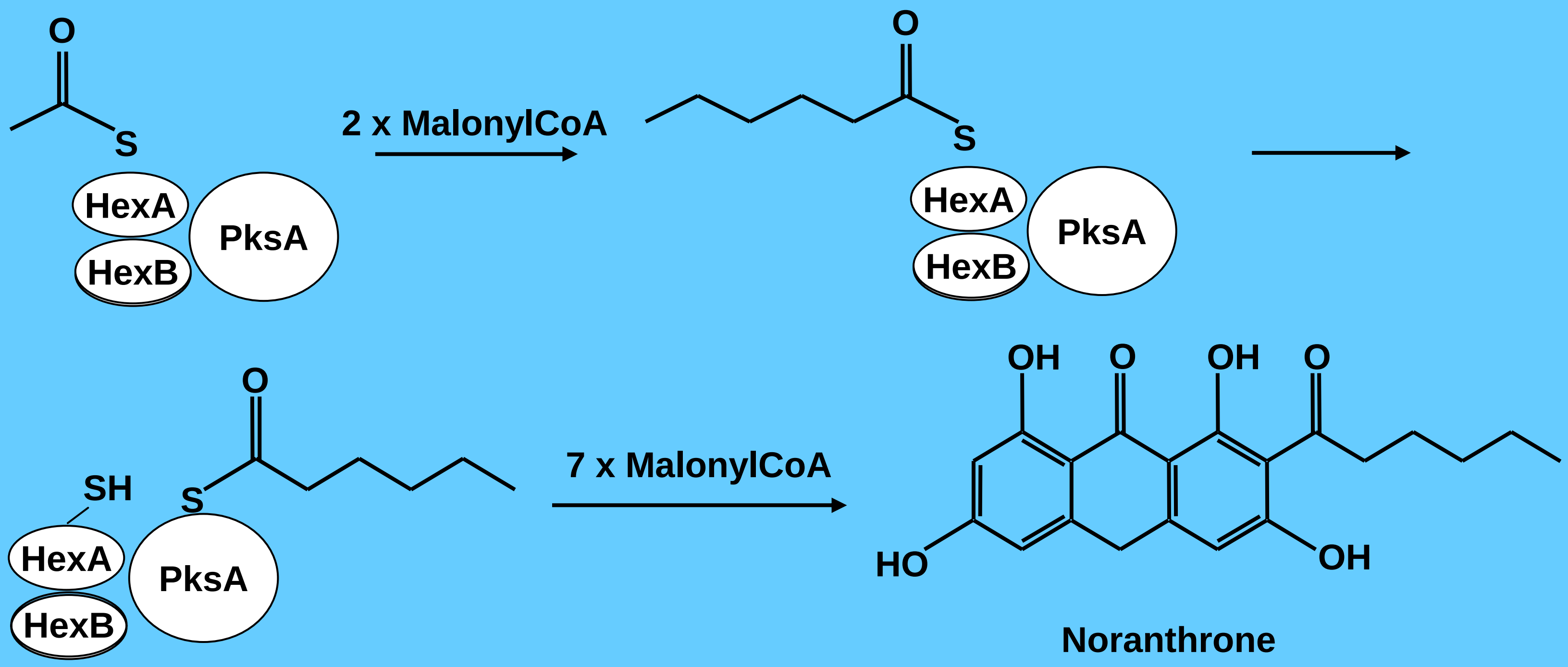


Polyketide synthesis



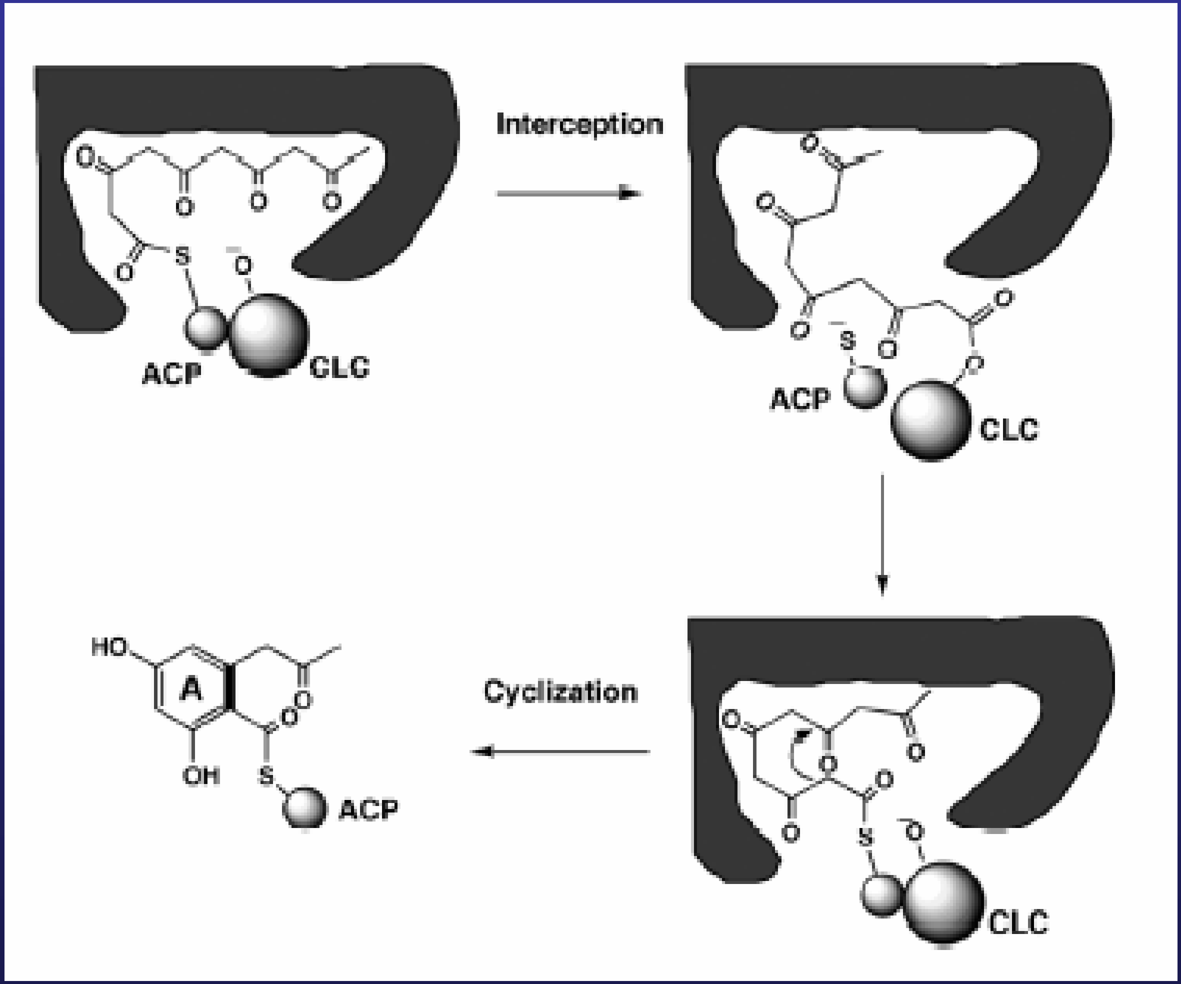
ACP	acyl carrier protein
KS	keto synthase
MAT	malonylCoA ACPtransacylase
PPT	phosphopantetheinyltransferase
SAT	starter unit ACP transacylase
PT	product template
TE/CLC	thioesterase/claisen-like cyclase
ER	enoylreductase
DH	dehydratase
KR	ketoreductase
AT	acetyltransacylase

NorS complex



Noranthrone

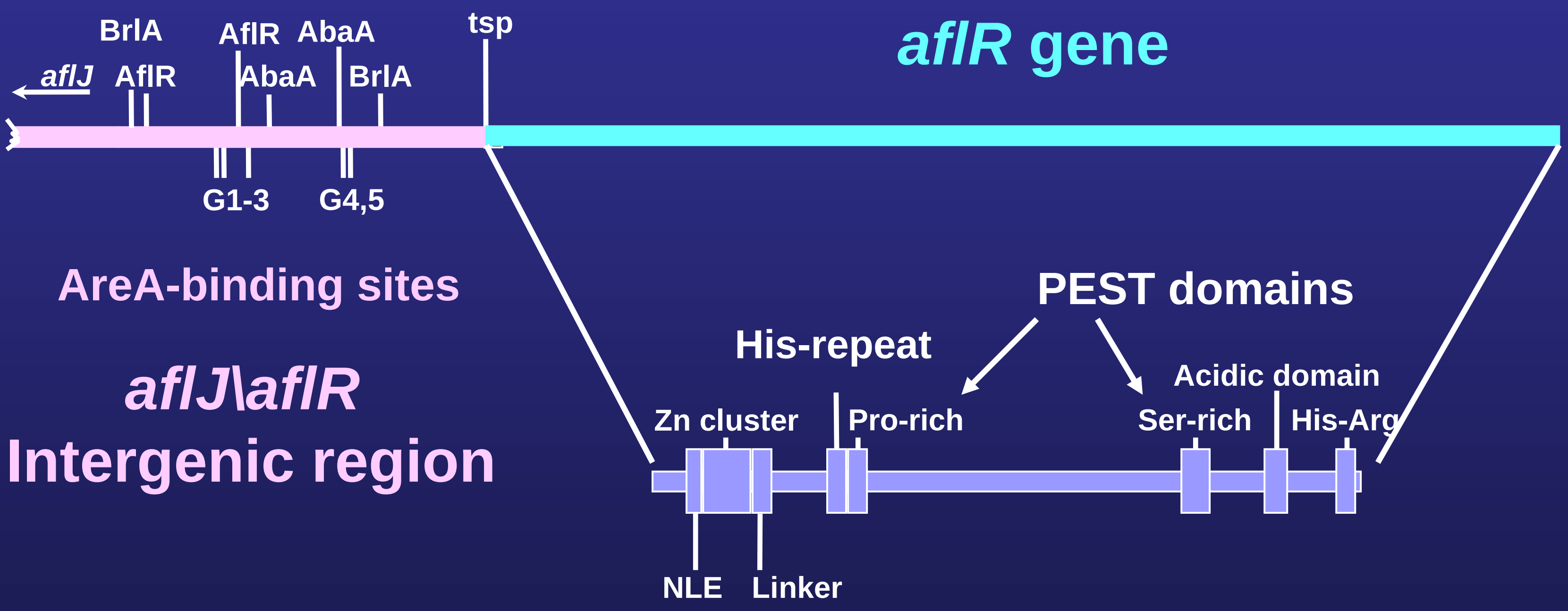
Control of PKS size by CLC domain



Watanabe&Ebizuka
2004

Overview of AflR and *aflR* intergenic region

Promoter with transcription factor binding sites



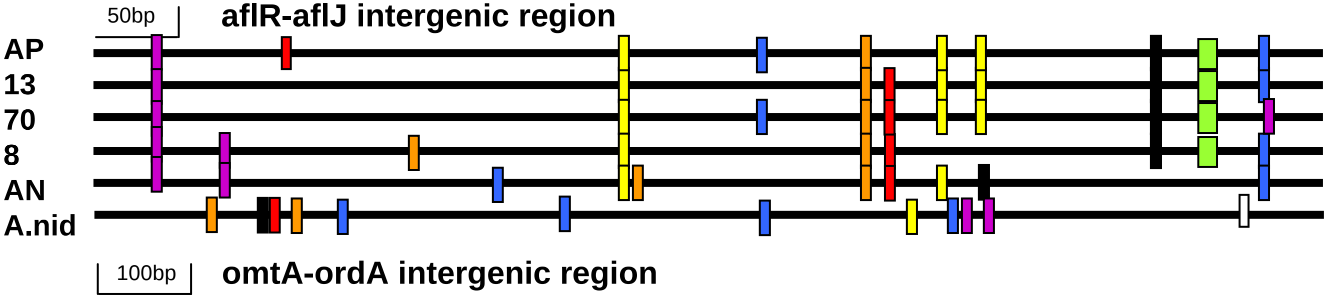
AreA-binding sites

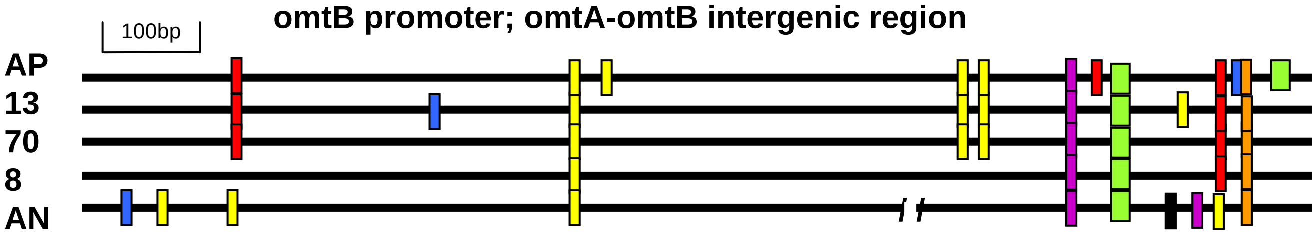
aflJ\aflR
Intergenic region

AflR protein

Variability in putative transcription factor binding sites in three AF cluster genes

⬜ TATA
⬛ CAAT
🟩 AflR
🟨 AreA
🟥 PacC
🟦 BrlA
🟧 AbaA
🟪 CreA





AflR coding domain variability

Strain	His-rich region (No. of His)	PEST region
A. flavus (Grp I)	6	No
A. parasit.	6	No
A. flavus (Grp II)	2	No
A. pseudotamar.	6	No
A. nomius	4	Yes
A. bombycis	4	Yes
A. nidulans	0	Yes
A. ochraceo.	0	Yes

Other levels of regulation

LaeA	**protein methyltransferase: affects only 2º metabolite clusters; may specifically affect pre mRNA stability, translation**
PKA	**mediated by G-protein signaling factors: FlbA and FluG**
VeA	**possible transcription factor: mediates formation of both sclerotia and AF; target is unknown.**

Epigenetic regulation

Chromosomal position of gene cluster

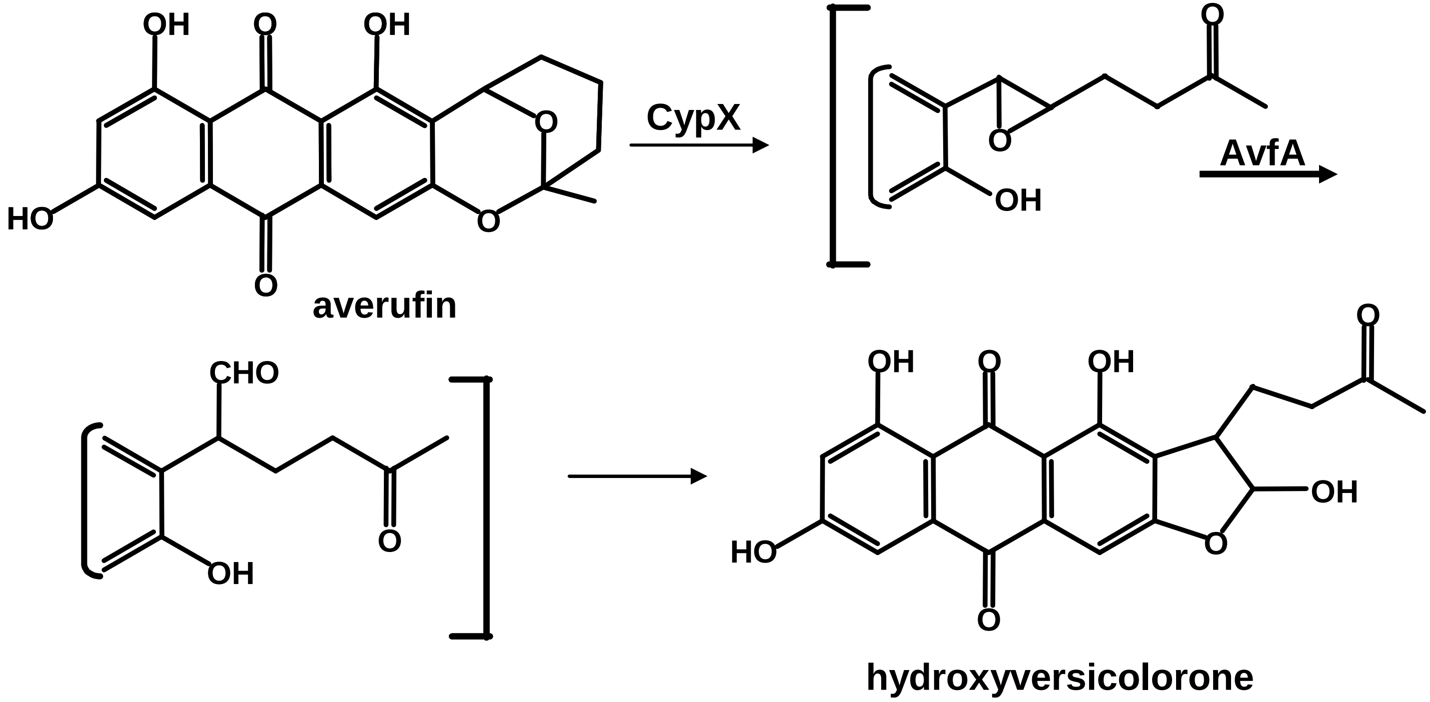
DNA methylation of GC-rich regions in subtelomeric region

Histone modifications: acetylation, methylation; phosphorylation

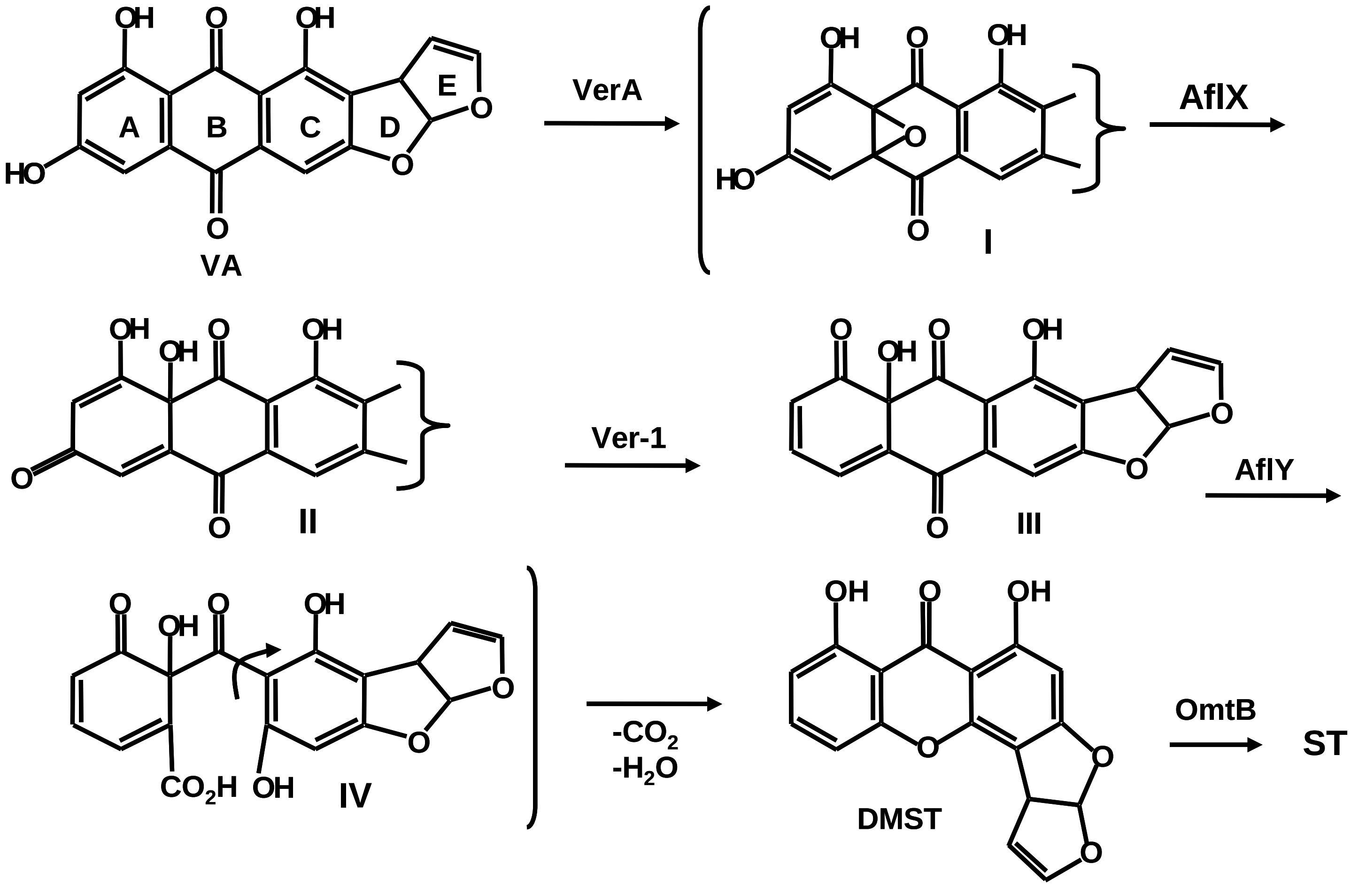
Conclusions

- Polyketide formation utilizes a Fas-derived starter unit and an iterative PKS
- Regulation of AF biosynthesis involves AflR and global transcription factors
- Some biosynthesis steps involve several oxidative enzymes, but no stable intermediates are detectable

Conversion of Avf to HVN



Conversion of VerA to ST



AF formation