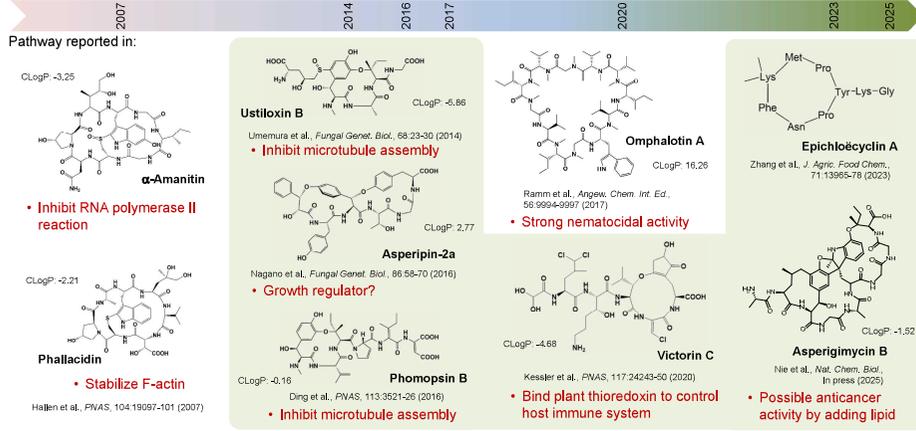


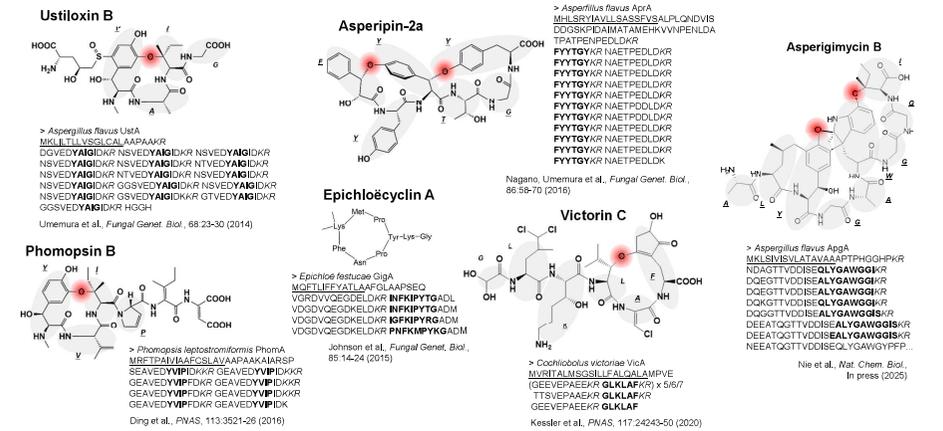


# History of fungal RiPP biosynthetic pathways



Water-soluble amphiphilic cyclic peptides, except omphalotin

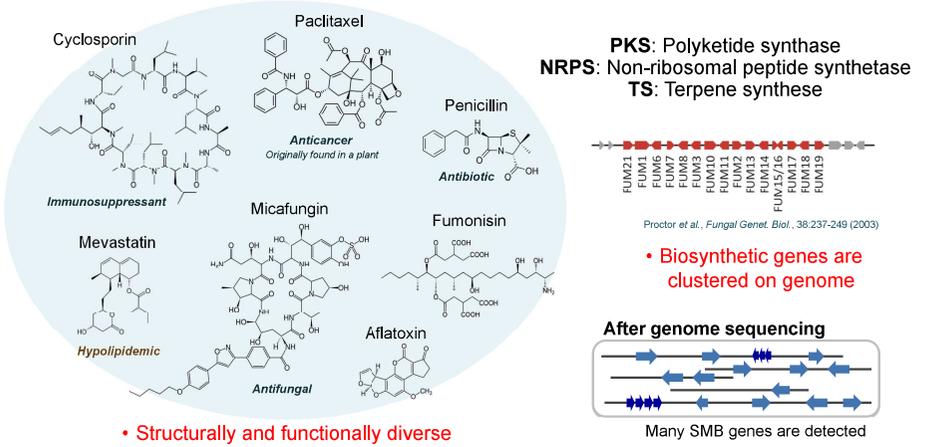
# Dikaritins: ascomycete RiPPs (so far)



Same class with cyclophane cyclic structure via ether bond to aromatic ring

# How did we discover the pathway?

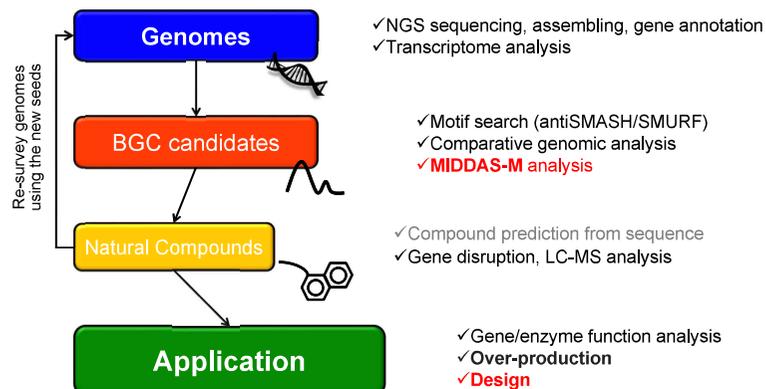
# Fungi produce various natural products



Structurally and functionally diverse

Rich repository for bioactive compounds

## Platform to find and utilize natural compound biosynthetic gene information



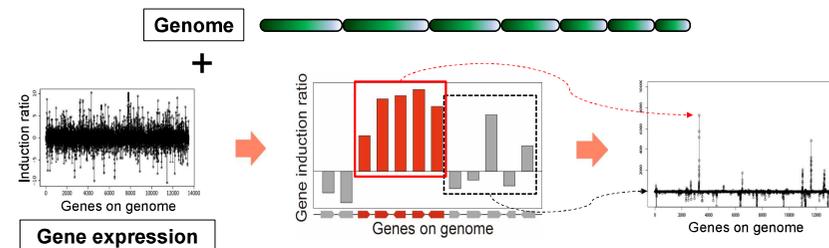
Whole utilization of genome and biological omics information

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## MIDDAS-M:

Motif-Independent De novo Detection Algorithm for SMB gene clusters

Using genome and transcriptome information, based on the co-regulated gene expression, sharply detect functional biosynthetic gene clusters (BGCs)



Detect motif-unknown BGCs, as well as known ones

Umemura et al., *PLoS One*, 8(12), e84028 (2014) 11

## Principle of MIDDAS-M algorithm

### Comprehensive creation of virtual gene clusters

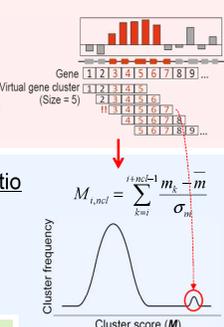
- For continuous genes on genome
- Change cluster size from 1 to 30

### Evaluation of induction ratio for virtual gene clusters

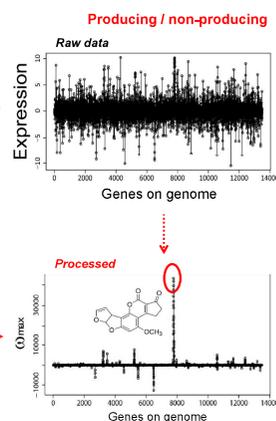
- Sum induction ratios of cluster member genes, followed by normalization

### Emphasis on deviation from normal distribution

- Introduce score probability  $P$



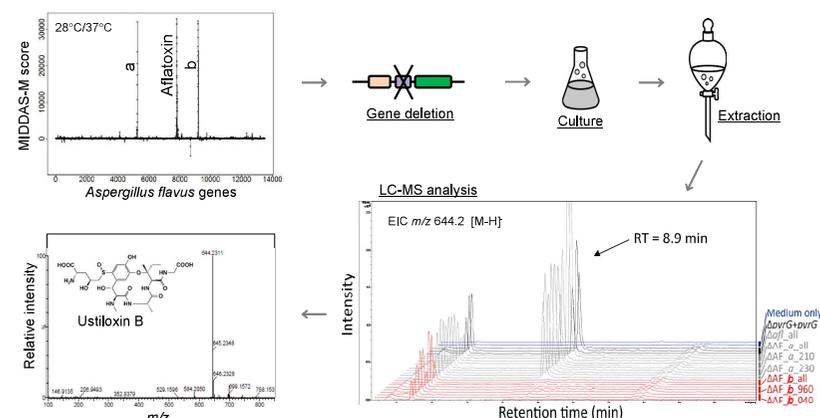
$$\omega_{i,nc1} = \left( \frac{M_{i,nc1} - M_{nc1}}{\sigma_{M,nc1}} \right)^j \log P_{i,nc1}$$



Umemura et al., *PLoS One*, 8(12), e84028 (2014)

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## Identification of ustiloxin BGC

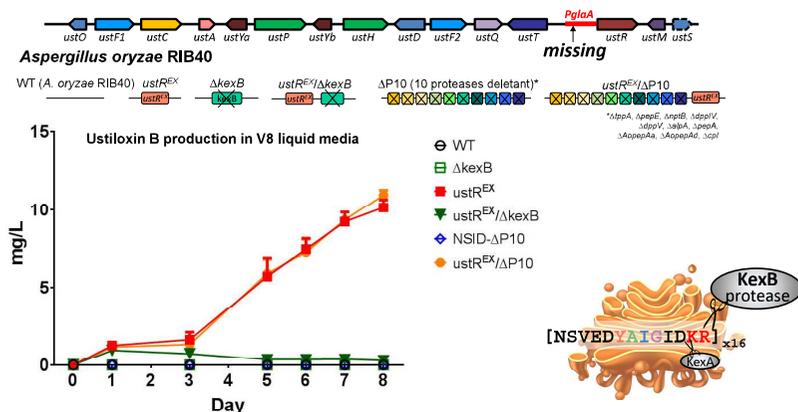


Cluster b deletants do not produce ustiloxin B

Umemura et al., *Fungal Genetics and Biology*, 68, 23-30 (2014) 15



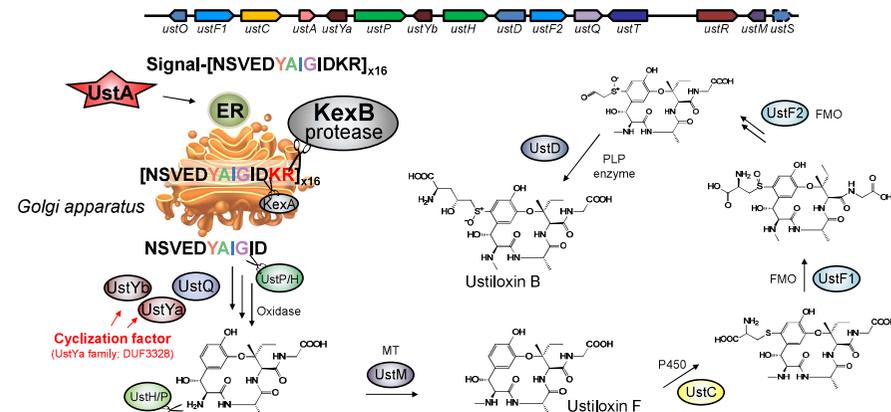
## Precursor protein is processed by KexB



KexB is indispensable for ustiloxin biosynthesis

Yoshimi et al., *AMB Express*, 6:9-16 (2016) 20

## Ustiloxin biosynthetic mechanism



After cyclization, tyrosine is modified with norvaline by 4 enzymes

Ye et al., *Angew. Chem., Int. Ed.*, 55:8072-8075 (2016) 23

## Distribution of dikaritin biosynthetic pathway in the fungal kingdom

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## KEP survey in the fungal kingdom

KEP: Kex2-processed repeat protein



Phyla	#Strain	Note	#KEP	per strain
Ascomycota	1024	Filamentous fungi	5372	5.25
Basidiomycota	317	Mushroom	1680	5.30
Mucoromycota	39	Mucor	357	9.15
Microsporidia	39	Spore-forming unicellular parasites	68	1.74
Chytridiomycota	16	Saprobic	178	9.37
Zoopagomycota	3	Obligate parasites	185	11.56
Cryptomycota	2	Lack chitinous cell walls	14	4.67
Blastocladiomycota	2		12	6.00
Not classified	2		12	6.00
<b>Total</b>	<b>1461</b>		<b>7878</b>	<b>5.39</b>

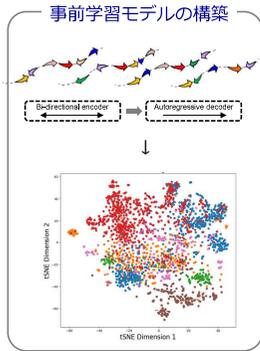
Latest assemblies registered in NCBI on Oct 9, 2019 were used

Umemura, *Fungal Biology and Biotechnology*, 7:11 (2020)

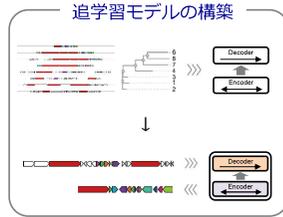
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# 進化軸情報に基づく 化合物生成遺伝子クラスターの生成的デザイン

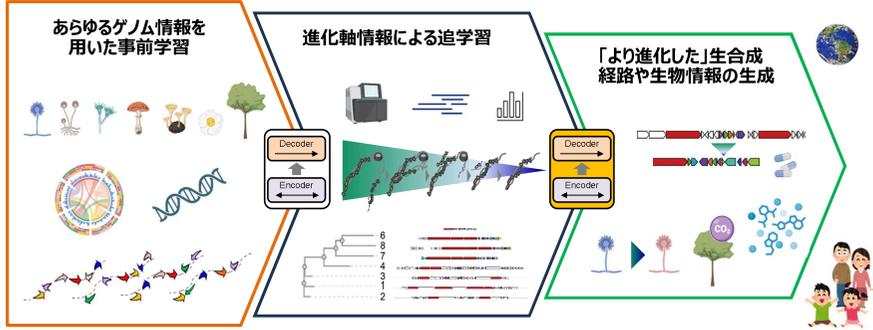


生物オミクス情報  
+  
大規模言語モデル  
=  
生成遺伝子  
クラスター生成  
=  
新規化合物の創出



ゲノム配列上の機能モチーフの集積情報を事前学習させたモデルに対し、  
進化軸の方向を追学習させることで、「より進化した」生成遺伝子クラスターを生成

# 「より進化した」生物情報生成モデルの構築



生産性・化合物物性/構造、さらに微生物特性等を望みの方向に設計できる学習モデルを構築  
あらゆる生物オミクス情報を組み入れたAI設計手法により、  
環境低負荷な物質生産系と有用微生物を創出し、炭素循環型社会の実現に貢献