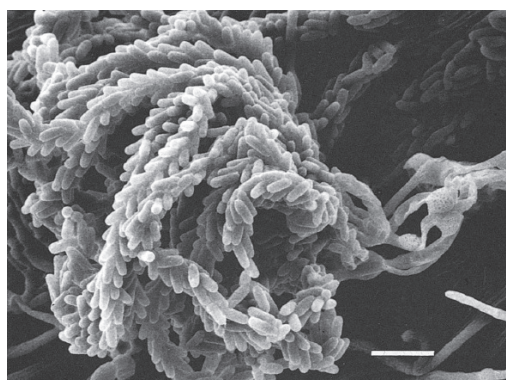


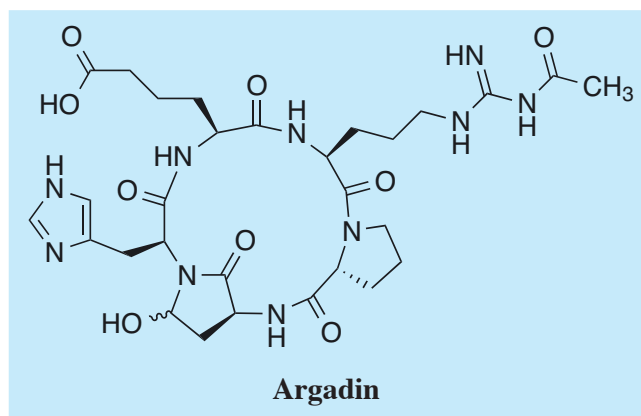
Argadin

1. Discovery, producing organism and structure^{1,2,7-9)}

Argadin was isolated from the culture broth of fungal strain *Clonostachys* FO-7314 and identified as a chitinase inhibitor. It is a cyclic pentapeptide composed of *N*⁰-acetyl-L-arginine, D-proline, L-aspartic β-semialdehyde, L-histidine, and L-2-aminoadipic acid in which the aldehyde carbon of aspartic β-semialdehyde is bonded to the histidyl α-amino residue. The stereochemistry of C_α and C_γ of aspartic β-semialdehyde and C_α of histidine were elucidated from the crystal structure of the argadin-chitinase complex. Argadin is the first peptide inhibitor of glycosidase showing nanomolar inhibition, and the argadin-chitinase complex revealed how the argadin backbone and side chains mimic the interactions of the enzyme with chito oligosaccharides. The first total synthesis of argadin was reported by Eggleston *et al.*³⁾, and it is shown to exist as a 5:1 mixture of diastereoisomers at C_γ of aspartic β-semialdehyde.



Clonostachys sp. FO-7314
Bar: 10 μm



2. Physical data

White powder. C₂₉H₄₂N₁₀O₉; mol wt 674.72. Sol. in acidic H₂O, acidic DMSO. Slightly sol. in H₂O, DMSO. Insol. in MeOH, acetone, CHCl₃.

3. Biological activity

1) Chitinase inhibition^{1,2)}

	Temperature	IC ₅₀ (μM) ^a			
		Argadin	Argifin ^b	Riboflavin ^c	Allosamidin ^d
<i>Lucilia cuprina</i> chitinase ^e	20°C	0.0034	0.103	N.T.	0.0004
<i>Lucilia cuprina</i> chitinase ^e	37°C	0.15	3.7	2.1	0.0023
<i>Streptomyces griseus</i> chitinase	37°C	1.9	14.8	>200	0.016
<i>Bacillus subtilis</i> chitinase	37°C	1.9	19.0	>200	0.025
<i>Serratia marcescens</i> chitinase B ^a	37°C	0.0192	32.5	N.T.	0.45

^a *S. marcescens* chitinase inhibition data are reported as K_i values.

^b Argifin is a new compound isolated in Kitasato (see Argifin).

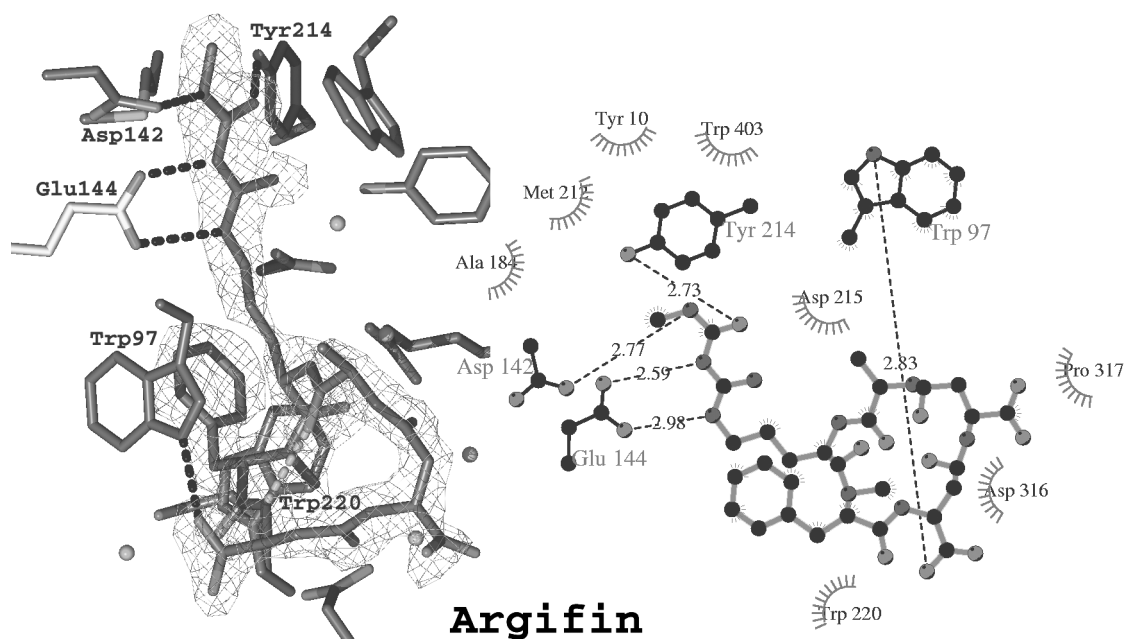
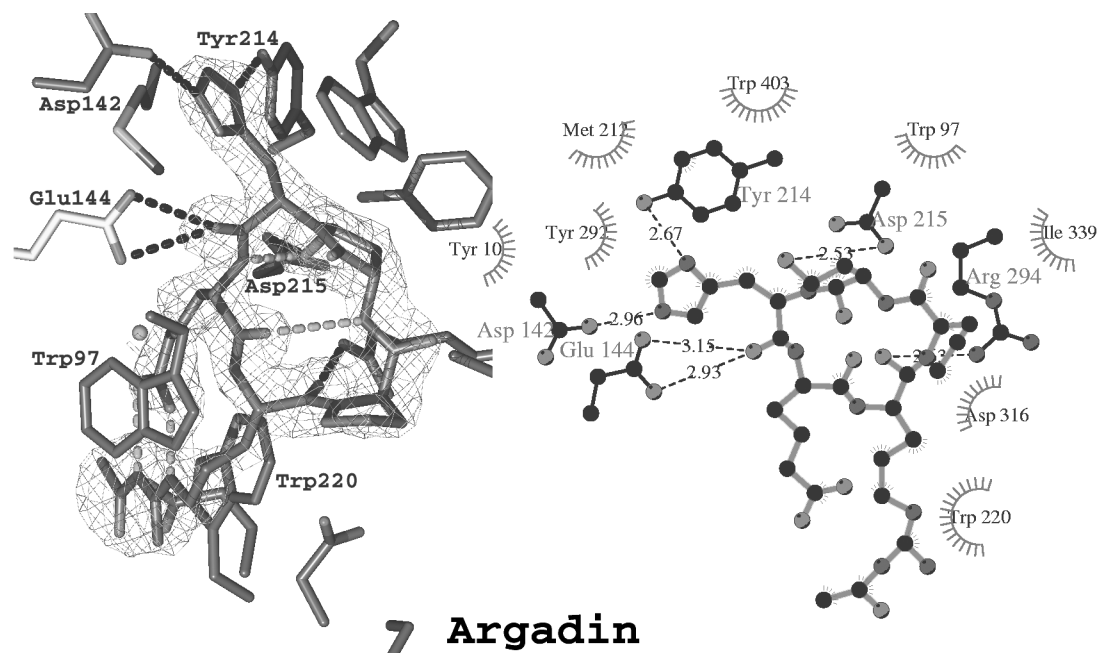
^c Riboflavin was isolated from a *Penicillium* strain as a chitinase inhibitor in Kitasato⁴⁾.

^d Allosamidin is a representative chitinase inhibitor.

^e *Lucilia cuprina* is sheep blowfly.

2) Argadin-chitinase complex and argifin-chitinase complex^{2,5,6,10)}

The structures of argifin and argadin in complex with *Serratia marcescens* chitinase B were resolved (2.0 Å resolution). These structures give an unprecedented view of how peptide based inhibitors inactivate carbohydrate-processing enzyme. For example, the carbonyl oxygen of the histidine in argadin occupies almost the same position as the scissile oxygen in the chitinase B-chitooligosaccharide complex and hydrogen-bonds to the catalytic acid (Glu144), while Glu-144 makes hydrogen bonds to the guanidinium group of the arginine side chain in argifin. The structures of argifin and argadin in complex with *Aspergillus fumigatus* chitinase and human bacterial-class chitinase (chitotriosidase) were also resolved⁵⁾.



3) Insecticidal activity¹⁾

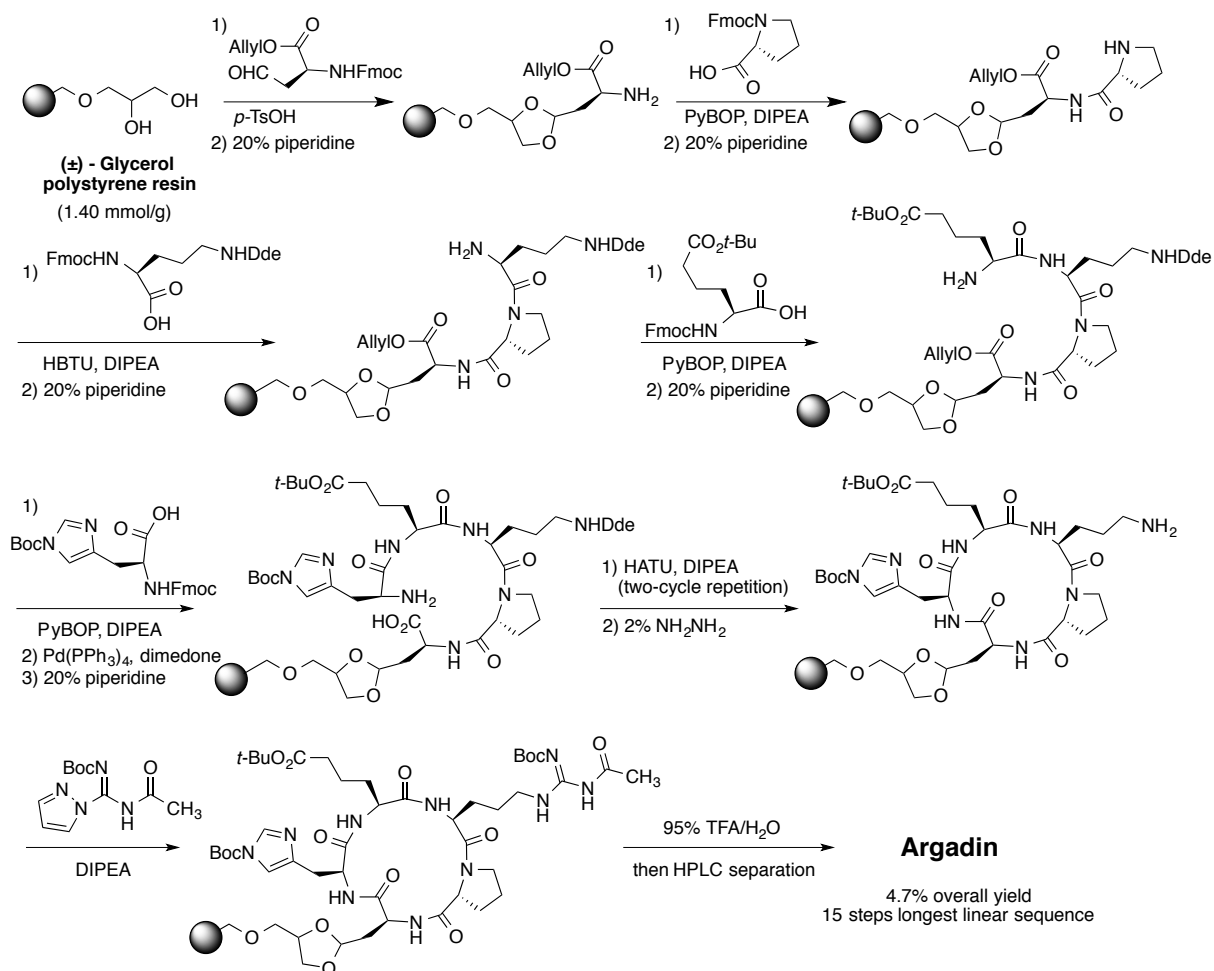
Argadin was injected into American cockroach (*Periplaneta americana*) larvae and compared with mock injected controls. No mortality was observed for control cases after day 1, while 60% and 38% mortality was observed for cockroaches injected with 20 μg and 2 μg of argadin, respectively.

4) Other biological activity¹⁾

Argadin did not inhibit the growth of tested bacteria, yeast, or fungi at 10 $\mu\text{g}/\text{disc}$ (paper disc method). Argadin also did not inhibit the growth of P388, KB, or HL-60 cells at 25 $\mu\text{g}/\text{ml}$.

4. Total synthesis^{3,11)}

The total synthesis of argadin was reported by two groups^{3,11)}. Below is a scheme achieved by Ōmura *et al.*¹¹⁾ (See Appendix-I).



5. References

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