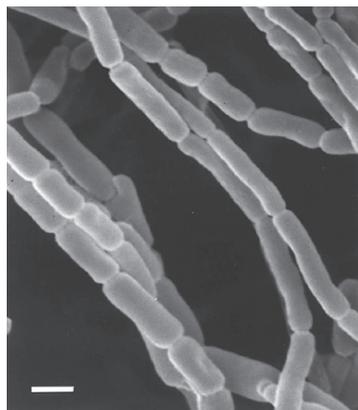


# Staurosporine ©

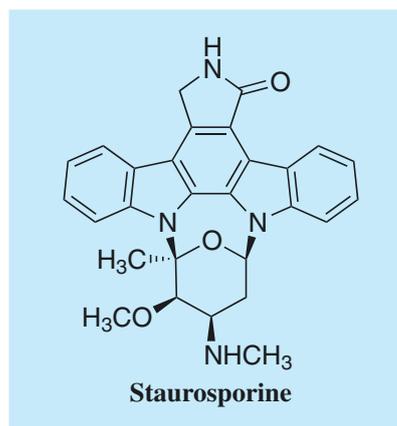
## 1. Discovery, producing organism<sup>23)</sup>

Staurosporine was originally discovered in 1977 from culture of *Saccharothrix aerocolonigenes* strain AM-2282<sup>T</sup> during a screening program for microbial alkaloid using a chemical detection method.<sup>1,2,3)</sup> During the past 30 years, staurosporine and related indolocarbazole natural compounds have been isolated from not only actinomycetes but also *myxomycetes* (slime molds), cyanobacteria and marine invertebrates such as sponges.

In 1986, 10 years after the discovery, staurosporine was found to be a nano molar inhibitor of protein kinases.<sup>4,5)</sup> Staurosporine is commercially available for biochemical research.



*Saccharothrix aerocolonigenes* subsp.  
*staurosporeus* AM-2282<sup>T</sup>  
(*Lentzea albida* AM-2282)



## 2. Physicochemical data and structure

Pale yellow crystals. C<sub>28</sub>H<sub>26</sub>N<sub>4</sub>O<sub>3</sub>; mol wt 466.20; Sol. in DMSO, DMF. Slightly sol. in CHCl<sub>3</sub>, MeOH.

The structure and absolute configuration of staurosporine was revealed by X-ray crystallographic analysis<sup>6,7)</sup> and confirmed by total synthesis by Danishefsky *et al.*<sup>8)</sup> and another group.

## 3. Biosynthesis of Staurosporine

Biosynthesis of staurosporine and rebeccamycin has been revealed by genetic and biochemical studies (Sanchez *et al.*<sup>9,10)</sup>, Onaka *et al.*<sup>11,12)</sup> and Groom *et al.*<sup>24)</sup> see figure).

In staurosporine biosynthesis, StaO initiates the synthesis by catalyzing L-tryptophan to imine form of indole-3-pyruvic acid (IPA imine) and StatD then catalyzes the coupling of two IPA imines to yield chromopyrrolic acid (CPA). The formation of indolocarbazole core of staurosporine is catalyzed by StaP. StaP converts CPA into three indolocarbazole compounds; staurosporine aglycone (K252c), 7-hydroxy-K252-c and acrylflavin A by intramolecular C-C bond formation and oxidative decarboxylation. The presence of StaC directs formation of K252c predominantly. StaG catalyzes a glycosidic bond formation between N-13 and C-6' and then StaN, a P-450 homolog, catalyzes an additional C-N bond formation between N-12 and C-2'. These two enzymes convert Staurosporine aglycone (K252c) to 3' *O*-demethyl, 4'-*N*-demethyl-staurosporine via Holyrine A and Holyrine B. StaMA catalyzes *N*-methylation of 3' *O*-demethyl, 4'-*N*-demethyl-staurosporine and StaMB catalyzes *O*-methylation which result in formation of staurosporine.



## 2) Action on Topoisomerases with ATP binding site

DNA topoisomerases have been shown to be an important target of antitumor drugs or antibacterial agents. Both antitumor drugs in eukaryotes and quinolone antibiotics in prokaryotes stabilize DNA-cleavable complex with topoisomerase. It was shown that staurosporine related structures induce DNA cleavable complex with mammalian topoisomerases. Semisynthetic derivatives of K252a were potent inducer of DNA cleavable complex with Topoisomerase I, whereas rebeccamycin was a weak inducer in the same assay.<sup>16)</sup>

## 3) Anti-microbial and pathogenic activity

Staurosporine was shown to have weak antifungal activity since the discovery by Ōmura *et al.*<sup>1,2)</sup> Later, protein Ser/Thr kinases were identified in many pathogenic organisms including mycobacteria, malaria and viruses.

Staurosporine and K-252 was found to inhibit mycobacterial growth at 5-50  $\mu\text{M}$ .<sup>17)</sup>

In *Plasmodium falciparum*, a cyclin- nuclear division cycle dependent kinase (CDK) PfPK5, was identified as the first such compound not derived from humans. Staurosporine inhibited PfPK5 with  $\text{IC}_{50}$  at 1  $\mu\text{M}$ .<sup>18)</sup> Antiviral properties of indolocarbazoles have been reported including human immunodeficiency virus, cytomegalovirus and Epstein–Barr virus.

## 4) Cytotoxic activity/Acute toxicity

Staurosporine showed a wide range of growth inhibition effects against cultured cells and was extremely cytotoxic in some cases (4pM, Hela-S3, 72 hr exposure ).

Staurosporine induced apoptosis as do anticancer drugs, but via partly distinct pathways. Several cell lines are completely resistant towards different anticancer drugs, but remain sensitive towards staurosporine-induced apoptosis. These finding encouraged further clinical trials for the use of staurosporine derivatives in antitumor therapy.

Acute toxicity (mice, i.p.)  $\text{LD}_{50} = 6.6 \text{ mg/kg}$

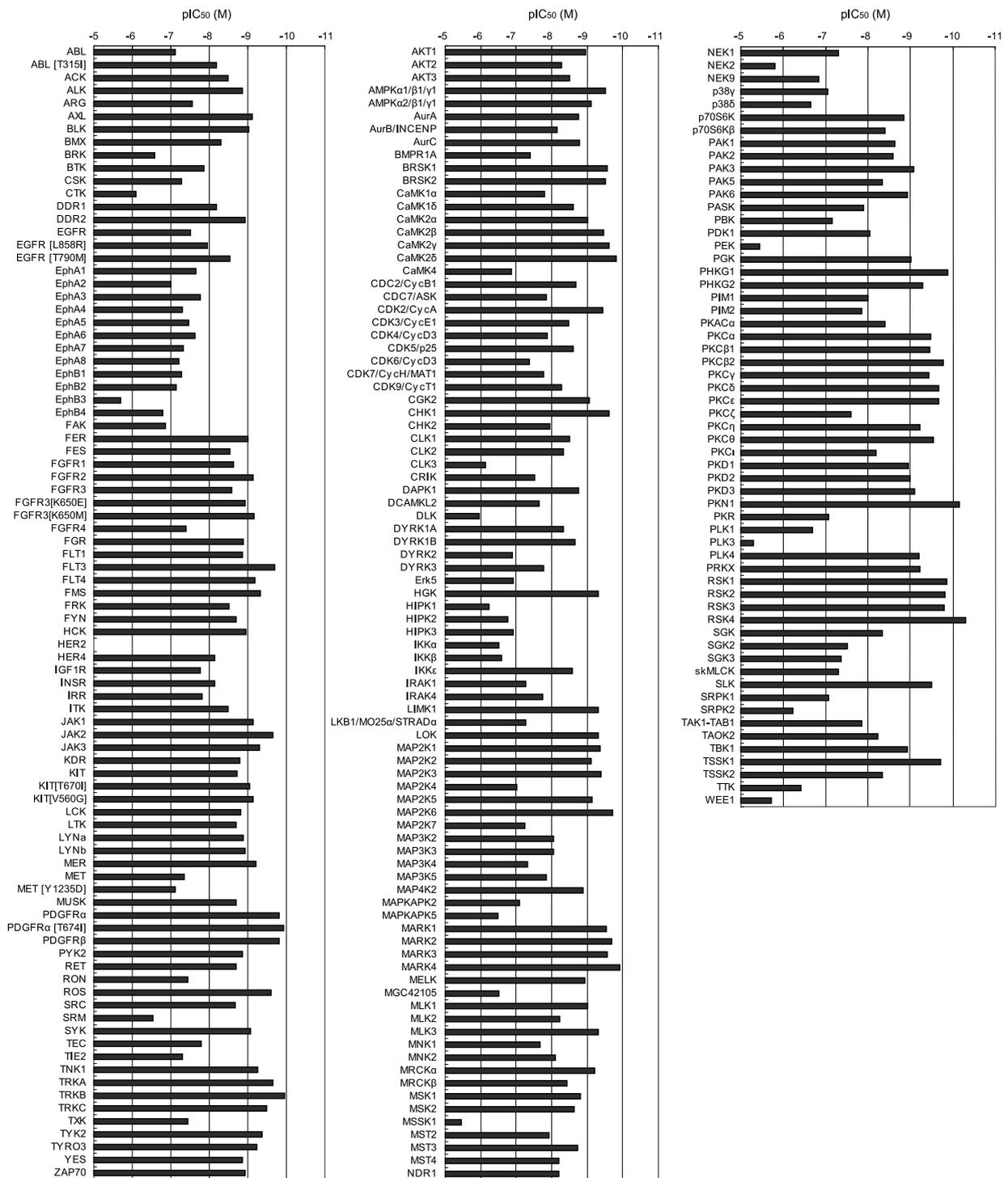
## 5) Other biological activities

Staurosporine and indolocarbazole derivatives showed many other biological activities.

Anti-angiogenesis activity of staurosporine derivatives with modification in its amino sugar moiety was reported<sup>19)</sup>.

Ruboxistaurin (Arxxant, LY333531) is selective inhibitor of Protein kinase C isoforms  $\beta$  and was evaluated in clinical studies Ruboxistaurin has been submitted to FDA for diabetic retinopathy.<sup>20)</sup>

Other reported activities include; inhibition of smooth muscle contraction<sup>21)</sup>, insecticidal activity, hypotensive properties, inhibition of platelet aggregation, activation of macrophages, immunosuppression, inhibition of the osteoclast proton pump, neuroprotection.



Inhibitory activity of staurosporine against protein kinases<sup>22)</sup>

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IC<sub>50</sub>s reported in early era : PKCs (2.7 nM), PKA(15 nM), v-src tyrosine kinase (6 nM) , EGF-R tyrosine kinase(25 nM, insulin receptor tyrosine kinase (IC<sub>50</sub> = 61 nM), phosphorylase kinase (3 nM), S6 kinase (5 nM)
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