THE SYNTHESIS OF COMPACTIN (ML-236B) AND MONACOLIN K IN FUNGI

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Compactin (ML-236B) and monacolin K (mevinolin) are specific inhibitors of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase, the rate-limiting enzyme in cholesterol biosynthetic pathway[1~7]. ML-236B was isolated from *Penicillium citrinum*[1] and *Penicillium brevicompactum*[2], and monacolin K from *Monascus ruber*[4] and *Aspergillus terreus*[6].

In the present study, approximately 4,000 newly isolated fungal strains from soil samples collected in the Tokyo Metropolitan area and 1,677 strains obtained from Institute for Fermentation, Osaka (IFO) (comprising of 596 genera, 1,489 species) were grown and assayed for their ability to produce ML-236B and monacolin K. As the result, *Hypomyces chrysospermus* IFO 7798, *Paecilomyces* sp. M2016, *Eupenicillium* sp. M6603, *Trichoderma longibrachiatum* M6735, and *Trichoderma pseudokoningii* M6828 were found to produce ML-236B. *Phoma* sp. M4452, *Doratomyces nanus* IFO 9551, and *Gymnoascus umbrinus* IFO 8450 were new producers of monacolin K.

Fungal strains were aerobically grown at 25°C for 7 days in a medium containing glucose 3.5%, starch 1%, soybean meal 2%, meat extract 0.5%, peptone 0.5%, NaCl 0.2%, KH_2PO_4 0.05%, and $MgSO_4 \cdot 7H_2O$ 0.05% (pH 5.8). In the first screening study, an aliquot (1~50 μl) of the culture filtrate was assayed for inhibition of sterol biosynthesis from [^{14}C]acetate by a rat liver enzyme system, as described previously[8]. As the result, 10 stains of newly isolated fungi and 5 strains from IFO were found to be active in inhibiting the *in vitro* cholesterol biosynthesis. The active strains were then tested for the ability to synthesize ML-236B and monacolin K. Culture filtrates obtained as described above were extracted with ethyl acetate at pH 3~4. The solvent layer was dehydrated over Na_2SO_4 and evaporated to dryness. The residue was subjected to silica gel chromatography and then to HPLC by the methods described previously[1,4,9]. Both ML-236B and monacolin K were identified by the combination of mass and UV spectrometry. Consequently, 4 newly isolated strains were found to produce ML-236B. These strains were identified as *Paecilomyces* sp. M2016, *Eupenicillium* sp. M6603, *T. longibrachiatum* M6735, and *T. pseudokoningii* M6828. *H. chrysospermus* IFO 7798 was also active in the production of ML-236B. Under the experimental conditions described above, 50 μg/ml of ML-236B was produced by *Paecilomyces* sp. M2016 and 1~3 μg/ml by others (Table 1).

As shown in Table 1, *Phoma* sp. M4452 (newly isolated), *D. nanus* IFO 9551, and *G. umbrinus* IFO 8450 were found to be new producers of monacolin K. Of these strains, *Phoma* sp. was the most active in the production of the metabolite.

Of the strains able to produce monacolin K, two strains were identified as already-known

Table 1. Fungal strains that produce compactin (ML-236B) and monacolin K (mevinolin).

Fungal strain	Productivity (μg/ml)	
	ML-236B	Monacolin K
Paecilomyces sp. M2016	49.6	—
Eupenicillium sp. M6603	~1	—
Trichoderma longibrachiatum M6735	1.5	—
T. pseudokoningii M6828	1.5	—
Hypomyces chrysospermus IFO 7798	2.7	—
Phoma sp. M4452	—	1.7
Doratomyces nanus IFO 9551	—	~0.1
Gymnoascus umbrinus IFO 8450	—	~0.1

species; *M. ruber* (M4681) and *A. terreus* (M4865).

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