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COMMUNICATIONS TO THE EDITOR

The Stereochemistry of Micrococcin P_1 Reinvestigated

Sir:

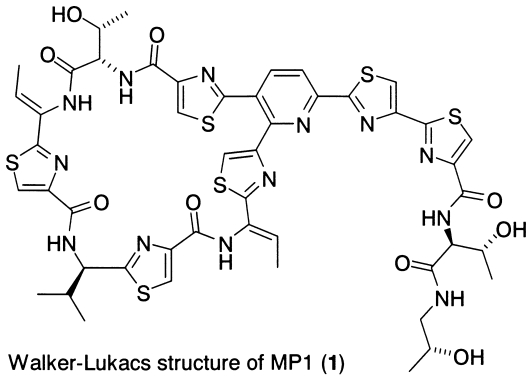
First discovered in 1948 from a strain of *Micrococcus* found in sewage from the city of Oxford,[1] the isolate that was to be named micrococcin[2] is recorded as the first example of a thiopeptide antibiotic,[3] although no work to elucidate the chemical structure of this material was ever reported. An antibiotic with therapeutic activity later obtained from the *B. pumilus* group of spore-bearing bacillus,[4] demonstrated a considerable degree of identity with the antibiotic isolated from *Micrococcus*, and so was named micrococcin P, in spite of the taxonomic implications. This antibiotic complex, which actually consists of two distinct components present in *ca.* a 7 : 1 ratio and designated micrococcin P_1 and P_2, respectively, has more recently been obtained from foodborne *Staphylococcus equorum* isolated from French Raclette cheese[5] and has been shown to inhibit the growth of the malaria parasite *Plasmodium falciparum*[6] and ribosomal protein biosynthesis in Gram-positive bacteria,[7] the latter through binding to the complex of protein L11 and 23S rRNA in the bacterial 50S subunit.[8,9]

A number of exhaustive studies have been conducted to determine the structural identity of the micrococcins, culminating in the WALKER-LUKACS proposal (**1**)[10] which was later revised by BYCROFT and GOWLAND (**2**, **3**) on the basis of chemical and spectroscopic evidence (Fig. 1).[11] However, the synthesis of micrococcin P_1 (MP1) by CIUFOLINI and SHEN in 1999 demonstrated that the BYCROFT-GOWLAND structure **2** did not correspond with the material isolated from the bacillus.[12] Subsequent NMR studies validated the constitution of this thiopeptide antibiotic and concluded that the difference between the natural product and synthetic material was purely stereochemical.[13] A synthetic epimer **4**,[14] that differs in the configuration of the isoalaninol residue, has been prepared but the (*R*)-stereochemistry of this unit is not in doubt.[15] In this manuscript we re-examine some of the original synthetic studies to resolve ambiguities in the absolute configuration of the micrococcin natural products.

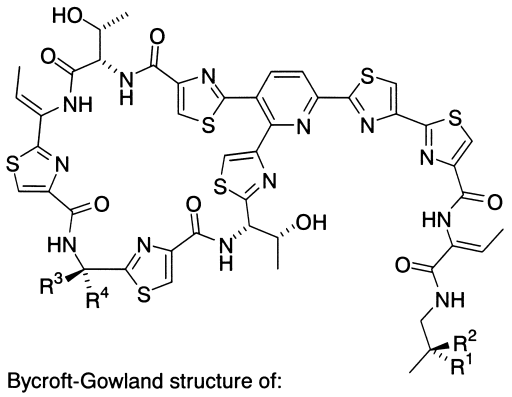
The configuration of the valine-derived thiazole residue in micrococcin was assigned by WALKER following the reproducible isolation of laevorotatory 2-(1-amino-2-methylpropyl)thiazole-4-carboxylic acid hydrochloride (**8**) from the hydrolysis products of MP1.[16] Comparison with synthetic material, prepared by the condensation of L-thioamide **5a** with ethyl bromopyruvate followed by hydrolysis of **6a** in 20% hydrochloric acid at reflux (Scheme 1) led to the configurational assignment of this residue and ultimately to the WALKER-LUKACS hypothesis.

In view of the well documented racemization of chiral α-aminothioamides in Hantzsch reactions[17] and the harsh conditions used by WALKER for hydrolysis of the benzamido *N*-protecting group,[16] we sought to access valine-derived thiazole **8** by a known alternative route[18] to establish its specific rotation and thus verify WALKER's

Fig. 1. Structures of MP1 and MP2.

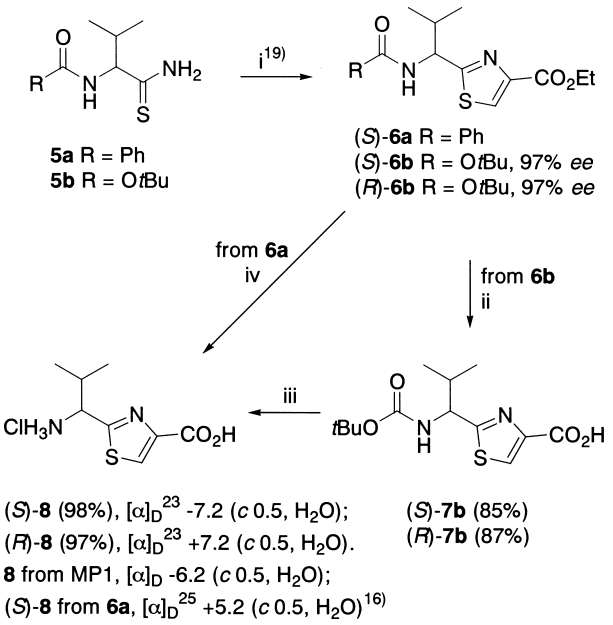


Walker-Lukacs structure of MP1 (**1**)



Bycroft-Gowland structure of:
MP1 (**2**) R^1 = OH, R^2 = H, R^3 = $CHMe_2$, R^4 = H;
MP2 (**3**) R^1,R^2 = O (π-bond), R^3 = $CHMe_2$, R^4 = H;
Shin's synthetic epimer of:
MP1 (**4**) R^1 = H, R^2 = OH, R^3 = $CHMe_2$, R^4 = H;
Structure of:
MP1 (**9**) R^1 = OH, R^2 = H, R^3 = H, R^4 = $CHMe_2$;

Scheme 1. Investigating the stereochemistry of hydrolysate **8**.



Reagents & conditions. (i) From **5a**, ethyl bromopyruvate, EtOH, reflux. From **5b**, $KHCO_3$, ethyl bromopyruvate, DME; then TFAA, 2,6-lutidine. (ii) From **6b**, LiOH, H_2O, MeOH. (iii) HCl in dioxane, RT. (iv) From **6a**, 20% HCl (aq), reflux, 4 h.

original findings (Scheme 1). *tert*-Butoxycarbonyl protected thiazole **6b**[19)] was prepared in 97% *ee* from the corresponding (*S*)-thioamide **5b** and hydrolyzed under basic conditions to give acid **7b** in 85% yield. *N*-Deprotection under acidic conditions gave the required amine hydrochloride, (*S*)-**8**, in 98% yield. The specific rotation of (*S*)-**8**, measured immediately following its isolation,[20)] was found to be laevorotatory, $[\alpha]_D^{23}$ −7.2 (*c* 0.5, H_2O) and closely matched hydrolysate **8** obtained by WALKER from MP1 (Scheme 1).[21)] This contrasts with the specific rotation of synthetic (*S*)-**8** obtained by WALKER and recorded with a small dextrorotation, $[\alpha]_D^{22}$ +2.1 (*c* 2.5, H_2O) or $[\alpha]_D^{25}$ +5.2 (*c* 0.5, H_2O). In order to verify the discrepancy in these findings, (*R*)-thioamide **5b** was also prepared according to the established route *via* (*R*)-thiazole **6b**, isolated in 97% *ee*, to give the hydrochloride, (*R*)-**8**, in 84% overall yield with an equal but opposite specific rotation.

We propose, considering these measurements, that the stereochemistry of the valine-derived thiazole unit in micrococcin P_1 should be corrected to *S*, based upon WALKER's isolation of laevorotatory hydrochloride **8** from the hydrolysis of MP1. It is now our hope that, on the basis of these studies, the validity of the BYCROFT-GOWLAND structure can be verified by total synthesis.

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