Pergamon

Tetrahedron Letters 39 (1998) 6625–6628

TETRAHEDRON LETTERS

# Absolute Configuration of the Polyazamacrolides, Macrocyclic Polyamines Produced by a Ladybird Beetle

Frank C. Schröder[a]*, Jay J. Farmer[a], Scott R. Smedley[b], Thomas Eisner[c], and Jerrold Meinwald[a]

[a]Baker Laboratory, Department of Chemistry, Cornell University, Ithaca, NY 14853, USA.
[b]Department of Biology, Trinity College, Hartford, CT 06106, USA.
[c]Section of Neurobiology and Behavior, Mudd Hall, Cornell University, Ithaca, NY 14853, USA.

Received 2 June 1998; accepted 30 June 1998

**Abstract:** The absolute configuration of the polyazamacrolides, oligomeric macrocycles from the pupal defensive secretion of *Epilachna borealis*, was determined by comparison of derivatives of the natural material with enantiomerically pure synthetic samples. Samples of a mixture of three (ω-1)-(2-hydroxyethylamino)alkanoic acids and of the corresponding aza-lactones were synthesized from (*R*)-alaninol. Gas chromatographic comparison of MTPA-amides of the synthetic aza-lactones with the MTPA-amides of aza-lactones prepared from the natural material established that the polyazamacrolides have the (*R*)-configuration at all stereogenic centers. © 1998 Elsevier Science Ltd. All rights reserved.

The pupal defensive secretion of the coccinellid beetle *Epilachna borealis* consists largely of a combinatorial library of macrocyclic polyamines, the polyazamacrolides (**1**-**6**, PAML's).[1,2] The PAML's are derived from an apparently non-selective oligomerization of the three (ω-1)-(2-hydroxyethylamino)alkanoic acids, **7**, **8**, and **9**, forming macrocycles with up to 200 and more members.[3] Here we report the assignment of the absolute configuration of the three building blocks, **7**, **8**, and **9**, of the PAML's.

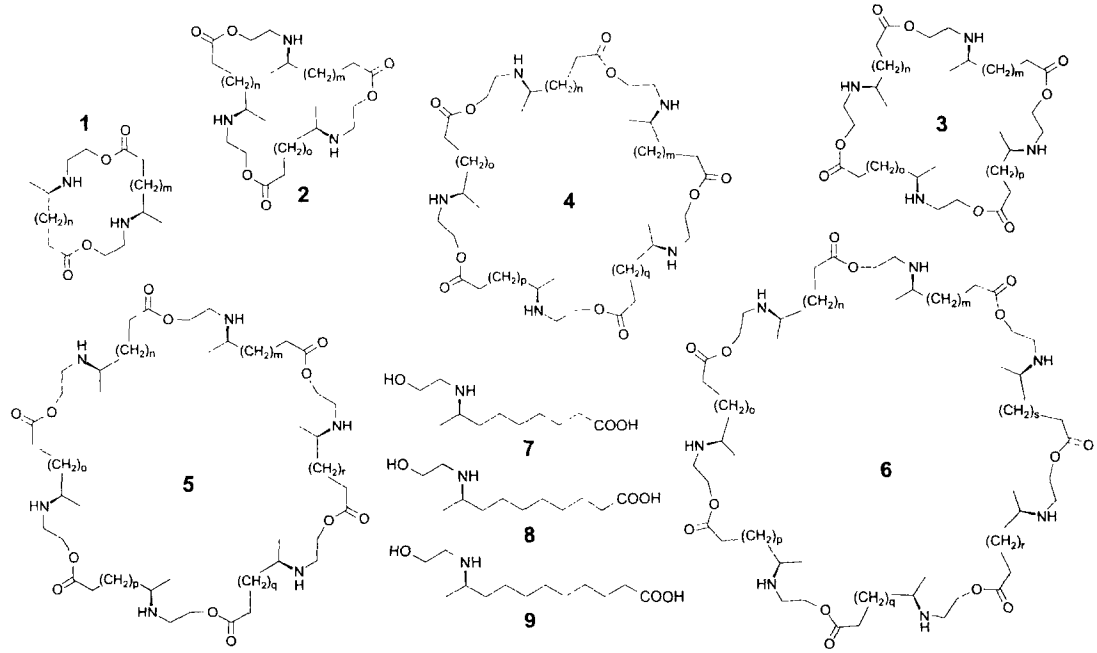



**Fig. 1** Library of PAML's, **1**-**6**, from the pupal defensive secretion of *E. borealis*, along with their building blocks, **7**, **8**, and **9**. In these formulas, each of the variables m...s can have the values 5, 6, or 7.


*PII:* S0040-4039(98)01404-X

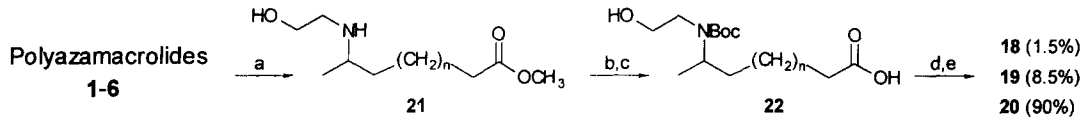



**Fig. 3** Synthesis of the aza-lactones **18**, **19**, and **20** from the natural mixture of PAML's (n = 5, 6, and 7). - a: NaOMe, MeOH, 10 h/0° C. - b: $Boc_2O$, THF, 2 h/25 °C. - c: 1N aq. KOH, MeOH, 6 h/25 °C. d: 2-chloro-1-methylpyridinium iodide, triethylamine, $CH_3CN$, 80 °C, high dilution.[6] - $CF_3COOH$, $CH_2Cl_2$, 2 h/0 °C.

Gas chromatographic comparison of the MTPA-amides of the synthetic lactones **18-20** with the MTPA-amides of samples of **18-20** that had been synthesized from the natural PAML's, indicated that the PAML-derived lactones and, therefore, the building blocks **7**, **8**, and **9**, show the (*R*)-configuration with at least 99% enantiomeric excess (Fig. 4). Interestingly, the configuration of the PAML's is opposite that of the structurally related lactone epilachnene (**23**), the major component of the defensive pupal secretion of a congeneric species, *E. varivestis* (Fig. 5).[8]

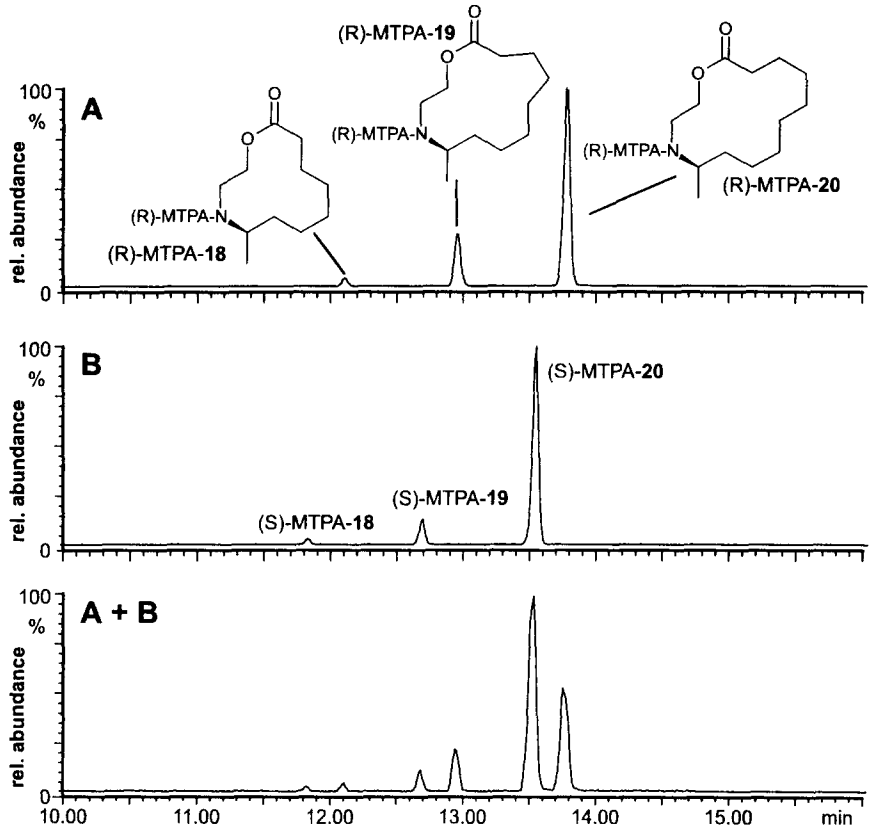



**Fig. 4** Assignment of absolute configuration of the lactones **18, 19** and **20** by gas chromatographic comparison of the corresponding Mosher-derivatives.[7] - **A**: (*R*)-α-methoxy-α-trifluoromethylphenylacetyl (MTPA) derivatives of synthetic (*R*)-lactones **18-20**. - **B**: (*S*)-MTPA derivatives of lactones **18-20** derived from the PAML's. - **A+B**: Mixture of samples **A** and **B**. - GC-column: J&W Scientific 29 m fused silica DB5-MS, film 0.25 µm, i.d. 0.25 mm; temperatures: starting at 150 °C, then increased 10 °C/min to 290 °C.

The (*R*)-enantiomers of the three (ω-1)-(2-hydroxyethylamino)alkanoic acids **7**, **8**, and **9** were synthesized as shown in Fig. 2. (2*R*)-2-Amino-1-propanol (**10**) was converted to the aziridine **11**, which was opened with 7-octenyl cuprate yielding intermediate **12**. After introduction of the 2-hydroxyethyl side chain and hydroxylation of the terminal double bond of **12**, the resulting alcohol **14** was oxidized with PCC in ethyl acetate, furnishing the acid **17** along with trace amounts of the chain degradation products **15** and **16**.[4,5] This usually undesirable side reaction in our case greatly facilitated matters. Prolonged reaction of **14** with PCC afforded a mixture of the three homologous acids **15**, **16**, and **17** in a ratio similar to the natural ratio of the (ω-1)-(2-hydroxyethylamino)alkanoic acids **7**, **8**, and **9** in the PAML's. This mixture was then employed to determine the absolute configuration of the natural material. Since volatile derivatives of the enantiomers of **7**, **8**, and **9**, such as *bis*-N,O-acetyl- or trifluoroacetyl derivatives of the corresponding methyl esters, did not separate well on our chiral GC-columns, the mixture of the homologous acids **15**, **16**, and **17** was converted into the lactones **18**, **19**, and **20**.[6] As shown in Fig. 4, the diastereomeric α-methoxy-α-trifluoromethylphenylacetyl (MTPA) amides of **18-20** are well resolved by gas chromatography.[7,8]

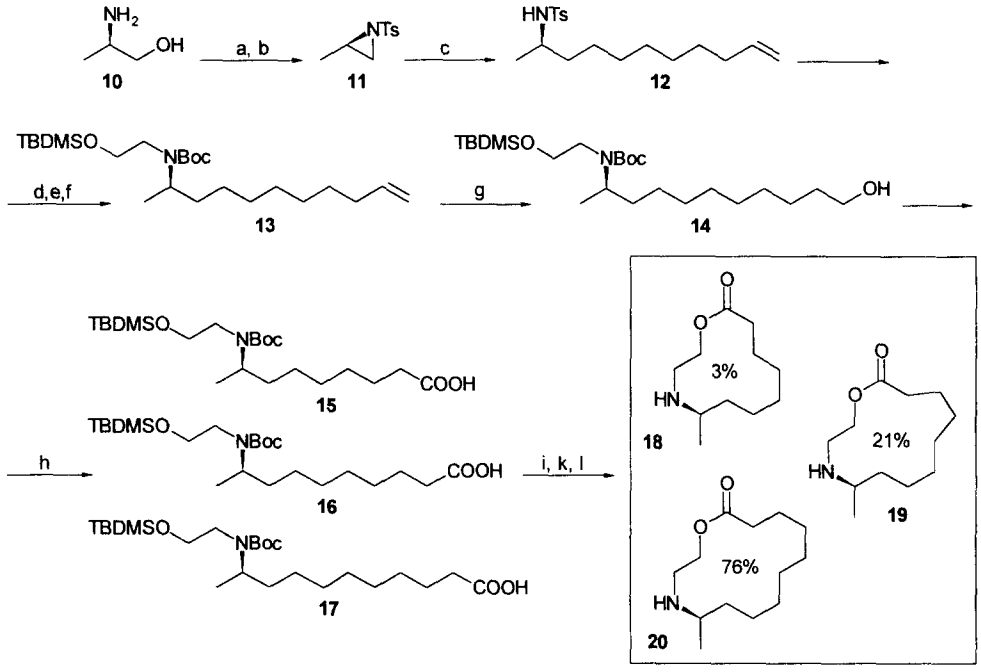
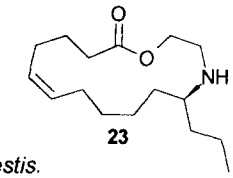



**Fig. 2** Synthesis of the (*R*)-enatiomers of the lactones **18**, **19**, and **20** from D-alaninol. - a: TsCl, pyridine. - b: $K_2CO_3$, acetone. - c: *bis*-(7-octenyl)magnesium, CuI, ether. - d: NaH, $Br(CH_2)_2OTBDMS$. - e: sodium naphthalide, DME. - f: $Boc_2O$, THF. - g: $BH_3$·THF. - h: PCC, acetic anhydride, ethyl acetate.[5] - i: $(C_4H_9)_4NF$, THF. - k: 2-chloro-1-methylpyridinium iodide, triethylamine, $CH_2Cl_2$, high dilution.[6] - l: $CF_3COOH$.

The conversion of the natural mixture of PAML's into the monomeric lactones, **18-20**, required us to perform the synthetic transformations shown in Fig. 3 on a very small scale.[9] Methanolysis of the PAML's afforded a mixture of the methyl esters **21**, containing about 20 μg of the methyl ester of the least abundant, lowest homologue **7**.[1] Following protection of the nitrogen with Boc anhydride and hydrolysis of the methyl ester, the mixture of N-protected hydroxy acids **22** was lactonized. Deprotection with trifluoroacetic acid yielded a mixture of the lactones **18**, **19**, and **20**, which were converted into the corresponding MTPA-derivatives.[7]




**23**

**Fig. 5** (*S*)-Epilachnene (**23**) from *E. varivestis*.


## ACKNOWLEDGMENT

The partial support of this research by National Institutes of Health grants no. GM53830 and AI2908, and by Deutsche Forschungsgemeinschaft grant no. Schr609/1-1 is gratefully acknowledged.


## REFERENCES AND NOTES


1. Schröder, F. C.; Farmer, J. J.; Attygalle, A. B.; Smedley, S. R.; Eisner, T.; Meinwald, J. *Science* **1998**, in press.
2. For recent reviews of the diverse defensive chemistry of coccinellid beetles, see: Daloze, D.; Braekman, J.-C.; Pasteels, J. M. *Chemoecology* **1995**, *5/6*, 173-183; King, A. G.; Meinwald, J. *Chem. Rev.* **1996**, *96*, 1105-1122.
3. The pupal secretion of *E. borealis* contains trace amounts of cyclic oligomers consisting of up to 20 apparently randomly selected units of **7**, **8**, and **9**.
4. Pure (11*R*)-11-(2-hydroxyethylamino)undecanoic acid (**9**) can be synthesized more conveniently via a slightly different route as described in reference 1.
5. Corey, E. J.; Schmidt, G. *Tetrahedron Lett.* **1979**, *20*, 399-400.
6. Mukaiyama, T.; Narasaka, K.; Kikuchi, K. *Chem. Lett.* **1977**, 441.
7. Dale, J. A.; Dull, D. L.; Mosher, H. S. *J. Org. Chem.* **1969**, *34*, 2543.
8. Farmer, J. J.; Attygalle, A. B.; Smedley, S. R.; Eisner, T.; Meinwald, J. *Tetrahedron Lett.* **1997**, *38*, 2787-2790.
9. *E. borealis* pupae (3-5 days old) were washed with 0.2 ml of $CH_2Cl_2$. The combined washings of 50 pupae were evaporated *in vacuo*. Subsequently, the oily colorless residue (about 45 μg per pupa) was dissolved in 0.5 ml of 2 N NaOMe in methanol. After stirring for 10 h at 0 °C, 0.1 ml of acetic acid was added. The resulting mixture was evaporated *in vacuo*, and the residue was dissolved in a mixture of sat. aqueous $K_2CO_3$ solution (1 ml) and ether (1 ml). The organic layer was separated, dried over $K_2CO_3$ and evaporated. The residue was treated with Boc-anhydride (1 mg, 5 μmol) in THF (0.1 ml) for 2 h at 25 °C. After evaporation, the Boc-protected ester mixture **21** was hydrolyzed by stirring with 1 N aq. KOH (1 ml) and methanol (1 ml) for 6 h at 25 °C. After evaporation of most of the methanol and acidification with acetic acid (0.1 ml), the mixture was extracted with ether (2 × 1 ml). The combined extracts were evaporated, the residue was redissolved in acetonitrile (1 ml), and a large excess of triethylamine (17 mg, 187 μmol) was added. This mixture was then added *via* syringe pump to a refluxing solution of 2-chloro-1-methylpyridinium iodide (24 mg, 94 μmol) in acetonitrile (5 ml) over a period of 2 h under argon. After the addition was complete, the mixture was refluxed for an additional 30 min. The mixture was then evaporated, and the residue redissolved in ether (2 ml) and water (2 ml). The organic layer was separated, filtered through a plug of silica, and evaporated. The residue was then treated with $CF_3COOH$ (50 μl) in $CH_2Cl_2$ (0.5 ml) for 2 h at 0 °C. After the addition of sat. aq. $K_2CO_3$ solution, the organic layer was separated and evaporated. The residue was derivatized as described in reference 7.