

nucleotide degradation fragment (3) readily demonstrates inhibition, with a measured potency of $3.2 \pm 0.5 \times 10^8$ agrocin units per mol.

Thus, it can be seen that the nucleotide fragment (3) without the N^6 -D-glucofuranosyloxyporphoramidate substituent shows the nonspecific growth inhibition of a simple antibiotic, but only agrocin 84 (1) with its unusual N^6 substitution shows the strain specificity of a bacteriocin for the pathogenic strain 57A.

Over similar concentration ranges, neither strain was inhibited by nucleotides (4) and (9) or the nucleoside (7). The absence of detectable activity using either indicator strain with the N^6 -substituted nucleotide (9) shows that although this is a necessary condition for selectivity, it is not a sufficient requirement for antibiotic activity. For detectable antibiotic activity in the bioassay, using initial well concentrations of less than 10^{-3} M, a phosphoramidate linkage to the amphiphilic 2,3-dihydroxy-4-methylpentanamide (6) seems to be mandatory.

The pathogenic *in planta* transconjugant strain 57A was originally obtained² using the non-pathogenic strain 57 as the recipient and the pathogenic strain 27 as donor. Examination by agarose gel electrophoresis¹¹ of the isolated plasmid DNA¹² from strains 57, 57A and 27 confirmed that strain 57A differed from strain 57 solely by the presence of a band corresponding in mobility to the 1.5×10^8 , molecular weight¹³ donor strain 27 tumor-inducing plasmid (pTi27). Murphy and Roberts¹⁴ have used this subtle difference in plasmid makeup to show that the virulence plasmid (pTi27) encodes an energy-dependent transport mechanism which selectively transports intact agrocin 84 into strain 57A. The absence of the virulence plasmid (pTi27) in strain 57 leads to exclusion of agrocin 84, and it is noteworthy

that agrocin 84 does not bind irreversibly to the cell surface of susceptible strains. The analogy of a molecular 'Trojan Horse' seems apt.

A cytosine 'nucleotide bacteriocin', agrocin 108 is also under investigation (Elvin and M. E. T. unpublished) and we suggest that these cell-specific nucleotides may be more abundant in nature than is generally realised.

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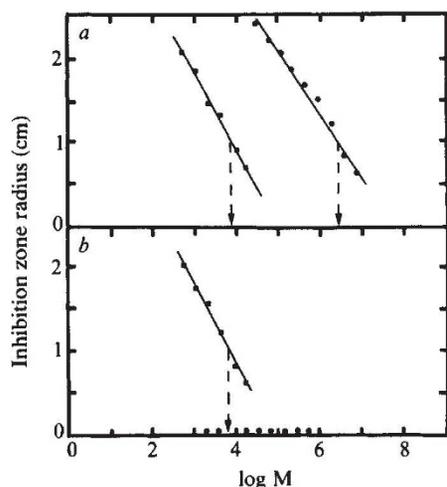


Fig. 2 Least squares fit for the negative logarithm of centre well ($20 \mu\text{l}$) molarity (initial) against inhibition zone radius using the pathogenic *Agrobacterium radiobacter* var. *tumefaciens*, strain 57A. Comparison of agrocin 84 (●; Fig. 1(1)) with the nucleoside 5' phosphoramidate of D-threo-2,3-dihydroxy-4-methylpentanamide (■; Fig. 1(3)). *b*, As for *a*, but with the non-pathogenic *Agrobacterium radiobacter* strain 57 as the bacterial indicator lawn in the Stonier¹⁰ bioassay. Dashed ordinates indicate the negative log molarity (initial) for an arbitrary agrocin unit of potency. Note that whereas the nucleoside 5' phosphoramidate (Fig. 1(3)) shows antibiotic activity towards both strains, only agrocin 84 (Fig. 1(1)) with an N^6 -D-glucofuranosyloxyporphoramidate substitution shows specific growth inhibition of the pathogenic strain 57A. Molar absorptivities of 19,860 at 264 nm for agrocin 84 (ref. 6) and 15,400 at 260 nm for the nucleoside 5' phosphoramidate were used for initial concentration measurements on electrophoretically and chromatographically homogeneous samples.

Erratum

In the letter 'Inhomogeneous distribution of filipin-sterol complexes in smooth muscle cell plasma membrane' by R. Montesano, *Nature* **280**, 328-329, Fig. 2 on page 329 was printed with poor contrast. The figure is reprinted below.

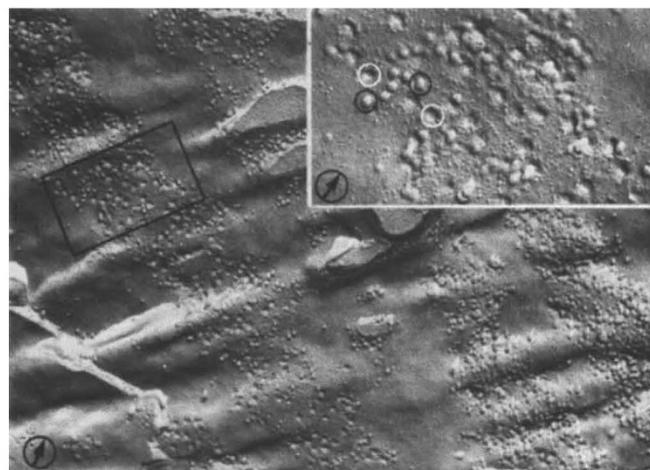


Fig. 2 Freeze-fracture replica of a smooth muscle cell fixed in glutaraldehyde-filipin mixture. The detail of a typical band of invaginations (black rectangle) is shown at high magnification in the inset. Numerous filipin-sterol complexes (black circles) are interspersed among the microinvaginations (white circles), whereas they are virtually absent outside the invaginated membrane regions. Filipin-sterol complexes appear as 25-30 nm protuberances which can be distinguished from the contiguous microinvaginations by the opposite polarity of platinum deposition (the direction of the shadowing is indicated by the encircled arrow). $\times 22,000$. Inset: $\times 44,000$.