

## Concerning the Structure of $\alpha$ -Mycolic Acids

By A. H. ETEMADI†

[*Institut de Chimie des Substances Naturelles, Gif-sur-Yvette (Essonne), France.*]

RECENT papers by Minnikin and Polgar<sup>1,2</sup> contain some criticism of our previous papers on the structure of  $\alpha$ -mycolic acids, to which we wish to reply.

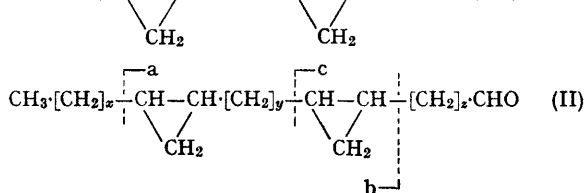
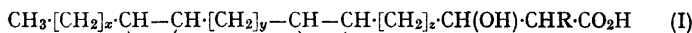
In 1964 we proposed the structure (I) for a type of mycolic acid,<sup>3-5</sup> which was later accepted by Minnikin and Polgar.<sup>6†</sup>

Having ascertained that the mass spectra of  $\alpha$ -branched,  $\beta$ -hydroxylated acids [for instance (I)] show among other peaks those of the spectra of the "meroaldehydes" [for example (II)]:<sup>7</sup>

we proposed values for  $x$ ,  $y$ ,  $z$  deduced from the fragmentation following (a) and (b); this gave in the case of the  $\alpha$ -mycolic acids of *M. tuberculosis*:  $y = 16$  and  $z = 11$ .

Then we found that these values were in disagreement with biogenetic considerations;<sup>8</sup> a re-examination of the mass spectra showed that the peaks due to fission (b) are accompanied by peaks at  $m/e \pm 14$ ; this fact vitiating the significance attributed to the former.

We noticed also the presence of peaks which we



† Present address: Milstead Laboratory of Chemical Enzymology, Broad Oak Road, Sittingbourne, Kent.

‡ Concerning the stereochemistry of the asymmetric centres at 2 and 3 positions of mycolic acids see C. Asselineau and J. Asselineau, *Bull. Soc. chim. France*, 1966, 1992; A. H. Etemadi, *Chem. Phys. Lipids*, 1967, 1, 165; A. H. Etemadi, J. Markovits, and F. Pinte, *Compt. rend.*, 1966, 263, D, 835; D. E. Minnikin and N. Polgar, *Chem. Comm.*, 1966, 648.

attributed to fragmentation following (c); the consideration of these led us to values  $y = 14$  and  $z = 13$ .<sup>9-11</sup> Our recent work, including degradation studies, supports the reality of the fragmentation (a) and (c).<sup>12</sup>

Minnikin and Polgar wrote: "recently Etemadi§ has mentioned a revised structure with  $y = 14$  and  $z = 13$  for the  $\alpha$ -mycolate (Test) based on a biogenetic analogy with the mycolic acids of *M. smegmatis* but evidence for this is not yet available". However, in the article they mention<sup>11</sup> we also added: "... et un nouvel examen des spectres de masse des  $\alpha$ -mycolates de méthyle Test¶ nous ont amené à proposer que  $y = 14$  et  $z = 13$ ".

Minnikin and Polgar posed three questions based on the mass spectra of these compounds.

The first question deals with the significance of doublet peaks of the series ... 487 ... The authors now accept that the oxygenated components originate from aldehydes (II): although the weakness of the intensity of the hydrocarbon components appeared not a general fact in our experiments with compounds of the type (I), nevertheless, we did not consider them as significant for structure determination<sup>9-11</sup> for the reasons given.

The second question deals with our choice of the peak of meroaldehyde at  $m/e = 740$  rather than that of its homologue at  $m/e = 768$  for a calculation on the basis of the fragmentation following (a) and (b). We gave the reasons for this choice [see peaks at  $m/e = 1132 (M - 18)$ ,  $m/e = 1100 = M - 50 (18 + 32)$  for the parent product, and the peak of meromycolate at  $m/e = 770$ ]; the over-oxidation the authors observed in their experiments did not occur in ours, otherwise we should have observed, by analogy, an important peak at  $m/e = 770 - 28$  (see Figure 5d in ref. 4). This is only a

question of the relative importance of two homologues in a mixture. We consider this to be of secondary importance; indeed the authors found, in agreement with our assignments, that the peak at  $m/e = 740$  is more important than its homologous peak.

In the third question the authors criticize our interpretation of the fragmentation of cyclopropane containing long-chain aldehydes. This criticism is, however, not valid as we had already stated that the corresponding methyl ester do not cleave in the same way as the aldehydes.<sup>9</sup> Moreover the difference in the behaviour of long-chain cyclopropanealdehydes (meromycolals) with specific fragmentation and the corresponding esters is implicit in our papers on mycolic acids of type (I).<sup>3-5</sup>

Another aspect questioned by Minnikin<sup>2</sup> concerning the n.m.r. spectra of mycolic acids of the type (I); in discussing the obvious unequal intensities of the signals at 35 and -18c./sec. (see for example the n.m.r. spectra we reported<sup>4</sup>) the author attributes to us the intention to assign to them an equal intensity, because we wrote, when we proposed structure (II) for  $\alpha$ -kansamycolic acids:<sup>3</sup> "Les signaux centrés à 35 et à -18 c./sec. peuvent être attribués à des protons de cycles

propaniques (a et b):  $\begin{array}{c} \text{b} \quad \text{b} \\ \diagdown \quad \diagup \\ \text{CH}-\text{CH}- \\ \diagup \quad \diagdown \\ \text{CH}_2\text{a} \end{array}$ ". We did not

wish to imply respective attribution of the signal at 35 c./sec. to protons of methylene and that at -18 to methine protons, but only that these signals indicate the presence of cyclopropane units; this is made apparent in our subsequent articles.<sup>4, 5, 13</sup>

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§ They refer to the reference 11.

¶ We mentioned here the references 9 and 10, the first of which appeared January, 1967.

<sup>1</sup> D. E. Minnikin and N. Polgar, *Chem. Comm.*, 1967, 312.

<sup>2</sup> D. E. Minnikin, *Chem. and Ind.*, 1966, 2167.

<sup>3</sup> A. H. Etemadi, A. M. Miquel, E. Lederer, and M. Barber, *Bull. Soc. chim. France*, 1964, 3274.

<sup>4</sup> A. H. Etemadi and E. Lederer, *Bull. Soc. chim. France*, 1965, 2640.

<sup>5</sup> A. H. Etemadi, Ph.D. Thesis, 1965.

<sup>6</sup> D. E. Minnikin and N. Polgar, *Tetrahedron Letters*, 1966, 2643.

<sup>7</sup> A. H. Etemadi, *Bull. Soc. chim. France*, 1964, 1537.

<sup>8</sup> A. H. Etemadi and F. Pinti, *Compt. rend.*, 1966, **262**, D, 1151; A. H. Etemadi, F. Pinti, and J. Markovits, *Compt. rend.*, 1966, **262**, C, 1343; A. H. Etemadi, "Colloque sur les aspects biochimiques de la phylogénèse", Montpellier, October 1966. *Bull. Soc. Chim. biol.*, in the press. A. H. Etemadi, M.D. Thesis, Paris, 1967.

<sup>9</sup> A. H. Etemadi, F. Pinte, and J. Markovits, *Bull. Soc. chim. France*, 1967, 195.

<sup>10</sup> A. H. Etemadi, "Colloque sur la chromatographie pyrolytique et la chromatochimie", October 1966, *J. Gas Chromatog.*, 1967, in the press.

<sup>11</sup> A. H. Etemadi, *Compt. rend.*, 1966., **263**, C, 1257.

<sup>12</sup> G. Lamonica and A. H. Etemadi, *Bull. Soc. chim. France*, 1967, in the press; *Compt. rend.*, 1967, **264**, C, 1711.

<sup>13</sup> E. Walczak and A. H. Etemadi, *Compt. rend.*, 1965, **261**, 2771.