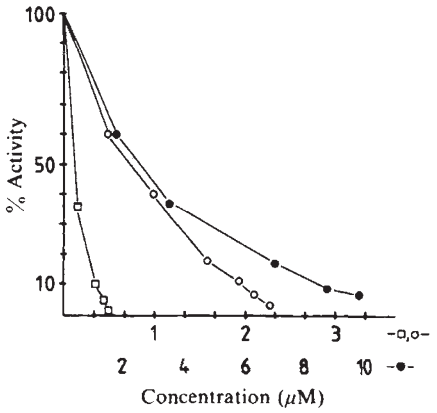

Fig. 3 Concentration dependency of pentalenolactone action. Assay conditions and enzyme concentrations were as detailed in Fig. 2, except for the non-phosphorylating glyceraldehyde-3-phosphate dehydrogenase (not shown, see text), when sodium arsenate was omitted. Incubation time before substrate addition was 15 min. □, Rabbit muscle enzyme; ○, spinach cytoplasmic enzyme; ●, spinach chloroplast enzyme. Note the change in scale on abscissa between glycolytic enzymes and chloroplast enzyme.

concentration of antibiotic needed to inhibit the chloroplast enzyme to the same degree is about 25 times those needed to inhibit the rabbit muscle enzyme. Thus, the chloroplast enzyme belongs in another group, as expected from its more variable cofactor dependency. To ensure that the NADP used in the chloroplast enzyme assay was not interfering with inhibition, some assays were done with NAD. The activity of a constant amount of chloroplast enzyme with NAD was about two-thirds of that obtained with NADP, but percentage inhibition with a controlled amount of pentalenolactone was the same. Thus, in a 2.5-μM assay solution the activity of the enzyme with NAD decreased 44% and the activity with NADP 50%. With a 3.5-μM solution 38% of activity was left in both cases. Furthermore, we confirmed that the protein-free plant extract contained nothing that interfered with the action of pentalenolactone. The third glyceraldehyde-3-phosphate converting dehydrogenase of higher plants, the nonphosphorylating enzyme, could not be inhibited to a significant degree by pentalenolactone concentrations up to 150 times that needed to inhibit completely the same amount of rabbit muscle enzyme.

Table 1 Relative sensitivities of different glyceraldehyde-3-phosphate dehydrogenases (GAPDH) to inhibition by pentalenolactone

Enzyme	Relative sensitivity
NAD-GAPDH, rabbit muscle	1*
NAD-GAPDH, *E. coli*	1.4*
NAD-GAPDH, yeast	3.2*
NAD-GAPDH, spinach	5.7
NAD(P)-GAPDH, spinach	24.5
NADP-GAPDH (nonphosphoryl. spinach)	>150

Sensitivities are expressed as concentration for 90% inhibition divided by concentration for 90% inhibition of muscle enzyme.
* Values from ref. 10.

Our results show that pentalenolactone is a universally applicable inhibitor of glycolysis. The difference in sensitivity to the antibiotic, shown by the three plant enzymes which function in different pathways, stresses the specificity of its action. The binding site of pentalenolactone to glyceraldehyde-3-phosphate dehydrogenase is not known, but Hartmann[10] has shown that the inhibitor strongly interferes with the binding of glyceraldehyde-3-phosphate to the rabbit muscle enzyme. Thus it seems possible that the substrate-binding part of the enzyme's active site is blocked by the inhibitor. The glycolytic enzymes from various organisms, having the same substrates and cofactors, and consequently a very similar active site, also show the same sensitivity to pentalenolactone. A minor change in the active site, indicated by the ability of the chloroplast enzyme to use NAD as well as NADP, is sufficient to decrease the enzymes' sensitivity to pentalenolactone considerably. Because of the epoxide structure of pentalenolactone E, its irreversible binding shown[10] for the rabbit muscle enzyme and also indicated for the spinach enzymes by the time-dependency of inhibition, and its interference with substrate binding, we conclude that it is covalently bound to the essential thiol group known to react reversibly with glyceraldehyde-3-phosphate during the catalytic cycle. The identification of the pentalenolactone binding site is in progress in our laboratory. We further speculate that the nonphosphorylating plant enzyme, which converts glyceraldehyde-3-phosphate to 3-phosphoglyceric acid, and is completely insensitive to inhibition by pentalenolactone, has no essential sulphydryl group in its active site.

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Erratum

In the article 'Tholins: organic chemistry of interstellar grains and gas' by C. Sagan and B. N. Khare, *Nature* **277**, 102–107, the following errors, generated at *Nature*, during editing, should be corrected: In paragraph 2 line 2, for H_2C read H_2S. In paragraph 3 line 11, for Sagan[8] read Sagan[10]. In paragraph 4 line 1, experimentally[9] should read experimentally[11]. In Table 2 the molecules listed, not the Table itself, are courtesy of Professor L. E. Snyder. The reference for the column labelled 'Gas phase spark' should be 37 not 30, and the reference for the column labelled 'Interstellar frost' should be 11 not 9. the last four lines of Table 2 should read:

	Molecule		
	Propionitrile		
	$CH_3CH_2C{\equiv}N$		
6.	Cyanodiacetylene	Pentenenitrile-H etc.	
	$CH{\equiv}C{-}C{\equiv}C{-}C{\equiv}N$		
8.	Cyanotriacetylene	Benzonitrile ring breakage-H	
	$CH{\equiv}C{-}C{\equiv}C{-}C{\equiv}C{-}C{\equiv}N$		
10.	Cyanoctatetrayne		
	$CH{\equiv}C{-}C{\equiv}C{-}C{\equiv}C{-}C{\equiv}C{-}C{\equiv}N$		

In paragraph 6 the penultimate sentence should read: 'The fit of four spectral features between 3 and 4 μm to an accuracy of 0.3 μm will occur by chance with a probability 10^{-8}.' In paragraph 9 line 13, for tholins[33] read tholins[41]. In paragraph 13 line 3, for spluttering read sputtering, and in the same paragraph line 5 should read 'positive ion-molecule reactions may produce the simplest inter-'. In paragraph 15 line 11, for induces[34] read induces[38]. In paragraph 16 line 6, the name of H. C. van de Hulst is misspelled. In reference 5 the authors of the paper in *Astrophys. J. Suppl.* are C. Sagan and E. E. Salpeter.

Corrigendum

In the letter 'Combined effects of adrenaline and insulin on active electrogenic Na^+–K^+ transport in rat soleus muscle' by J. A. Flatman and T. Clausen, *Nature* **281**, 580, in Table 3 the units for serum insulin concentration should read μU ml^{-1}.