

paths. The same is true when South America is included in the analysis. The inference is quite strongly that Africa and South America behaved as a unit for the whole of the Precambrian, and indeed the authors bring forward some rather less compelling evidence that North America belonged to this unit until about 1,000 million years ago.

There are two fairly simple conclusions to be brought out of this: first, that orogenic belts do not necessarily form only during continent/continent collisions, and, second, that for much of the geological past a layer fraction of the continent of the world was not subject to rifting. This raises the intriguing possibility that it might be feasible to date (give or take a few hundred million years) the start of plate tectonics as it is known. Before that time one would have to assume that either plates as thin boundary layers did not exist or that somehow rifting was not a normal occurrence in tectonics. An amusing twist to uniformitarianism.

D. D.

HAEMOGLOBIN

Quaternary T, Tertiary r

from a Correspondent

INTEREST in the details of the structural differences between unligated high-spin deoxyhaemoglobin in the T (tense) state and the ligated oxy and met derivatives in the R (relaxed) state continues to stimulate further investigations. Anderson (*J. molec. Biol.*, **79**, 495; 1973) now describes a technique for obtaining crystalline methaemoglobin in the deoxy quaternary conformation; that is, with the tertiary structure in the met or r state, and with the quaternary structure in the deoxy or T state (the upper and lower case letters here denote quaternary and tertiary states respectively). As Anderson points out, a better stereochemical understanding of the pathway(s) by which conformational changes take place requires crystallographic studies of stable intermediates. Mutant human haemoglobins of the M type, in which the quaternary structure is locked because two of the four haems remain permanently in the met (r) state, have been useful in studies of the effect of ligand binding on the α or β subunits in the absence of a change in quaternary structure. Similarly BME-Hb, in which a bifunctional reagent has formed a covalent cross-link between the Cys(93) and His(97) residues of each β chain, is locked permanently into the oxy (R) state.

Anderson's new approach is to intro-

duce an acrylamide monomer into the water space of the human adult deoxy-Hb crystal by co-crystallisation, followed by photochemical polymerisation of the monomer inside the crystal, in the hope that the polymer would clamp the molecule in its original T quaternary structure, while allowing small, tertiary movements on ligand binding. Air oxidation of these deoxy-acryl crystals does indeed give met-acryl (T; t \rightarrow r) crystals suitable for X-ray diffraction studies, in striking contrast to the breaking up of Hb-A crystals which normally follows changes in the ligation state.

X-ray analysis of these crystals to 3.5 Å resolution by the difference Fourier technique revealed marked changes in tertiary structure in the region of the haem pockets and the $\alpha_1\beta_2$ contacts between the subunits. The iron atom moves towards the plane of the porphyrin, though probably not the full distance of 0.45 Å which it moves in the deoxy (T) to met (R) shift. The even larger movement (nearly 0.75 Å) of the iron atom, almost into the haem plane, found for CO (R) haemoglobin cannot be observed in this constrained T state system because the crystals break up, presumably because of a transition to the R state, despite the presence of the polymer in the crystal lattice. Movement of the iron atom causes a change in tilt of the haem. This acts as a lever which initiates stereochemical changes weakening several hydrogen bonds, both within and between subunits, which determine the stability of the tertiary and quaternary deoxy structures. The later ligand of the r state ferric subunits causes a slight opening of the haem pocket in the α subunit and a much larger one in the β subunit.

The structural changes seen by Anderson in changing from the tertiary deoxy (t) to the aquomet (r) state within the quaternary T structure are similar but opposite to those seen by earlier workers in changing from the aquomet (r) state to the deoxy (t) state in the quaternary R structure of BME haemoglobin. Thus changes in tertiary structure associated either with addition of ligand to the T structure or the removal of ligand from the R structure are complementary. The electron density maps show that the α haems autoxidise more readily than the β haems, just as the β haems were reduced more easily than the α haems in BME haemoglobin.

Anderson's results support many of Perutz's earlier ideas about the mechanism of haem-haem interaction. They confirm that changes in tertiary structure on ligation are a consequence of the contraction of the iron-nitrogen bonds and also, especially in the β subunits, of the steric effect of the ligand itself. The differences in the roles of the two types of subunit contact are also evident. The $\alpha_1\beta_1$ interface, although showing

large collective movements, seems to be an essentially passive contact, as there is no evident set of movements linking α and β haems across it. By contrast, in the $\alpha_1\beta_2$ interface the smaller number of relative movements are concentrated on individual side chains. A series of interactions can be seen involving residues in van der Waals contact with α and β haems and mediated between subunits by a specific rearrangement of hydrogen bonds. Although the conclusions drawn from the X-ray study of the met-acryl (T, r) crystal must be regarded as tentative for several reasons, Anderson suggests that the major role of the $\alpha_1\beta_2$ interface in determining quaternary structure now seems to be well established.

BACTERIA

Computer Identification

from a Correspondent

TAXONOMY may be divided principally into classification, nomenclature and identification. Computerised classification based on the results of a large number of characteristics of individual bacterial strains has been developed, initially by Sneath. Taxonomic groups (taxa) are then chosen which contain bacteria with the greatest similarity of characters. Names are then assigned, according to an International Code, to the taxa. An unknown strain may then be identified by comparison with known, named taxa using a few diagnostic characters, often in the form of a key.

This identification stage has now been automated by the Computer Trials Department at the Central Public Health Laboratory, Colindale (*J. gen. Microbiol.*, **77**, 273; 1973). The department has been building up this system since 1967 and it has based its evidence on trials of 1,595 bacterial strains, 1,079 of which were reference strains and the remainder mostly strains sent for identification by the Public Health Laboratory Service. These were strains which the PHLS laboratories had had difficulty in identifying, and which a large centralized laboratory could more easily deal with.

The groups chosen as the known taxa in the system were made up from those bacteria known to be found in medical specimens, those strains considered pathogenic and a few other strains. The bacteria were all Gram-negative and rod-shaped. The forty-eight characters or tests chosen for the survey were those which were considered the most reproducible and easiest to prepare, carry out and read. The range of the tests chosen included many of the objective, biochemical tests used in medical bacteriology laboratories.

The method used for identification was polythetic—that is, organisms shar-