indicate changes not revealed by optical methods. From resonance peak areas as a function of denaturant concentration, it seems that the order in which the histidine residues participate in the unfolding process is 12, 119, 48, 105. This order can be rationalised in terms of the known tertiary structure of RNase, and is therefore also relevant to the folding process.

The possible methods for revealing the existence of intermediate partially unfolded states were noted by R. H. Pain who pointed out that although some earlier equilibrium and kinetic studies on, for example, lysozyme unfolding in guanidine hydrochloride, indicate a 2-state process with first-order kinetics up to about 90% reaction, this situation is not general. The analysis of bimodal kinetics can reveal the presence of intermediate states, or of alternative states off the main folding-unfolding path. Pain reported his investigation of the pencillinase of *S. aureus*, a protein with no sulphydryl or disulphide groups, which was specifically selected for unfolding studies, and summarised the evidence for the existence of a highly expanded state, with the same α-helix content as the native protein, formed by a cooperative transition at low denaturant concentrations.

L. W. Nichol (Australian National University, Canberra) discussed ligand binding in associating protein systems, and showed how the form of the binding curve could be discussed in terms of regions of sigmoidality defined by the interaction parameters. He described how ultracentrifugal sedimentation equilibrium measurements can be used to evaluate these parameters. G. A. Gilbert (University of Birmingham) emphasised the central role of protein–protein interactions in the conformational control of protein function mediated by allostery, and pointed out that in the so-called non-equilibrium methods (for example, chromatography, electrophoresis, velocity centrifugation) the progress of the moving boundary reflects the equilibrium situation in the following plateau region, as well as the non-equilibrium situation within it.

The topical subject of membrane-bound proteins and protein–lipid interactions was discussed by C. Tanford (Duke University). Experimental data on protein–detergent associations (much used as a contemporary model) were described. The behaviour of cytochrome *b*5 and a phage protein in detergent systems as consistent with the idea that membrane proteins possess distinct regions around which true micellar structures can organise. The discussion included a brisk exchange on the problems of measuring protein molecular weights by ultracentrifugation in the presence of detergents.

Proton NMR studies on histones were discussed by E. M. Bradbury (Portsmouth Polytechnic). With increasing ionic strength structure is induced through interchain interactions between regions of apolar residues, and the basic segments interact with DNA, leading to the formation of chromosome superstructure. The lysine-rich F1 histone, however, seems to be involved specifically in salt-induced contraction of chromatin gel, and in chromosome condensation through a phosphorylation process.

The programme included two parallel sessions of shorter papers on various aspects of protein unfolding and folding processes, ribosome reconstitution, RNA-protein complexes, and mucoprotein. Noteworthy was the study by Creighton, Dyckes and Sheppard (MRC Laboratory of Molecular Biology, Cambridge) on the refolding of reduced pancreatic trypsin inhibitor, in which the different species existing at the 1-disulphide bond stage were detected and characterised. The refolding of this protein proceeds with eventual complete recovery of native structure, and the pathway for refolding indicated by this work is consistent with the view that folding is directed by segments which have acquired the correct secondary structure.

Chlorophyll and primary productivity

from our Plant Ecology Correspondent

SINCE chlorophyll is essential for photosynthetic energy conversion, it is not unreasonable to suppose that it might provide a rough estimate of the potential gross primary productivity of an ecosystem. Early in this century the work of German plant physiologists suggested a close relationship between chlorophyll and photosynthetic rate. Later it became apparent that the chlorophyll density of vegetation expressed as g m^{-2} of ground area was remarkably similar in a variety of ecosystems and this led Odum (*Fundamentals of Ecology*, Saunders, 1959) to propose that there is a community homeostasis in which chlorophyll levels are maximised within the limiting physical constraints of the environment, leading to similar chlorophyll densities in most ecosystems.

Such a tenuous hope was shattered by Bray (*Can. J. Bot.*, **38**, 313; 1960) who concluded that chlorophyll densities in a wide variety of vegetation types might vary by as much as twenty times in natural ecosystems. He found that chlorophyll densities were higher in climax ecosystems than in seral ones and were greatest in highly stratified systems such as forests. These conclusions were largely confirmed by Ovington and Lawrence (*Ecology*, **48**, 515; 1967) who made detailed studies of four contrasting ecosystems—a prairie grassland, a savanna, an oakwood and a maize field.

Although Odum's simple homeostasis proposal was no longer tenable, there remained the possibility that chlorophyll might provide an index of gross primary productivity. Ovington and Lawrence's data in no way contradicted such a proposal. The underlying presupposition, however, is that all of the chlorophyll present must be active in photosynthesis whenever light levels permit; physiologists have always regarded this as extremely unlikely.

One rather startling observation which Ovington and Lawrence made was that a large proportion of the total chlorophyll content of their oak woodland was contained in the branches of the trees. In the month of August the trees themselves contained 29×10^3 g ha^{-1} chlorophyll, of which 12×10^3 g ha^{-1} (41%) was contained in the older branches. The bulk of this branch chlorophyll remained after leaf fall in September/October. Immediately the question must be raised as to whether this large reserve of chlorophyll is active and, perhaps even more important, whether it could result in winter photosynthesis in these deciduous trees.

Although only relatively simple experiments were required to settle this question, they have been conducted only very recently. Much is known about the winter photosynthesis of evergreens (see, for example, Kozlowski and Keller, *Bot. Rev.*, **32**, 293; 1966) but very little work has been done on deciduous trees. Keller (*Photosynthetica*, **7**, 320; 1973) has now examined rates of photosynthesis and respiration in the bark of ash (*Fraxinus americana*), aspen (*Populus tremula*) and larch (*Larix decidua*) during the winter. In his experiments plants were taken from their growing sites where they had been exposed to normal winter temperatures, they were kept at 16°C for 16-20 h and their carbon dioxide exchange rates were then measured by infrared gas analysis at 40, 10 and 0 klx respectively. In all species at all light intensities the carbon dioxide emission exceeded absorption; in other words, respiration exceeded photosynthesis. Thus, despite the presence of abundant chlorophyll, these species are likely to be experiencing a negative carbon balance through the winter months.

These experiments should serve as a salutary warning to production ecologists of the dangers inherent in the use of chlorophyll level as a productivity index, despite the general correlation between chlorophyll density and the gross primary productivity of ecosystems.