Preface

Living organisms resist the changes imposed on them by the environment—in the words of geneticists "they exhibit homeostasis". They so maintain themselves against outside influences that this property can be used as a criterion to separate living from non-living matter, and yet, paradoxically, the triumph of biochemistry has been, and continues to be, the analysis of organisms and their metabolism in terms of the same chemical and physical processes that operate on inanimate matter. One may justifiably ask how then can life be separated from non-living things, at what level of complexity or organization does life begin? Alternatively, what are the characteristics of the reactions carried out by living organisms which inanimate reactions lack? The reader is advised that clear answers to these questions are not to be found in this book. What he will find is a survey of the current state of our knowledge of the diverse mechanisms used by plants to regulate their biosynthetic metabolism and growth.

During the last fifty years the major metabolic pathways have been largely worked out in sufficient detail to show that the overall reactions do not proceed exactly as predicted from measurements of the kinetics of the individual reactions. Even quite small sequences of reactions that produce cellular products show an unexpected degree of regulation and the more complex growth processes are obviously subject to intricate control.

Although some of the individual steps of many reaction sequences and the enzymes that catalyse them are still undiscovered, a new insight has been obtained into the behaviour of the overall processes during the last fifteen to twenty, and particularly during the last five, years. This new knowledge has begun to give an understanding of how the reactions are controlled within the cell. An earlier volume in this series ("Biosynthetic Pathways in Higher Plants", J. B. Pridham and T. Swain, eds) dealt with the biosynthetic pathways used by plants; in 1972, at the meeting in Canterbury, we returned to the same topic but our discussions had a different emphasis. Obviously, knowledge of this subject is still far from complete but enough is now known of the different kinds of mechanism that regulate growth and metabolism for one to be able to review the subject of control mechanisms which operate in plants.

In the first lecture of the symposium Professor Davies brought out an exciting new concept of metabolic control, arguing that the pH of cells is regulated by the mutually antagonistic activities of carboxylating and decarboxylating enzymes being adjusted as the pH of the cellular environment moved towards or away from their respective optima. He also discusses the other broad concepts of metabolic regulation "energy charge", the NAD/NADH ratio, the pyradoxamine phosphate/pyridoxal phosphate ratio and the

balance and co-ordination of the enzymes of nitrogen and carbohydrate metabolism.

Bacteria, honorary plants for the meeting, regulate their enzymatic activities principally by making new enzyme molecules and the intricacies of the process are described with clarity by Mr Tristram so that the methods of control, described by Dr Miflin, which occur in higher plants (barley roots) are shown in counterpoint. The biosynthesis of amino acids is perhaps better defined than that of any other comparable cellular constituent; consequently, this basic knowledge provides the best framework for investigating regulatory mechanisms. Mr Tristram's review is illustrated by his work using naturally-occurring amino acid analogues (isolated from various higher plants by Professor Fowden and co-workers) as novel inhibitors to probe the capacities of the bacterial biosynthetic system. Similarly, Dr Miflin describes some of his own work to illustrate the interrelationships between the separate pathways by which the amino acids are formed.

The contrasts between the methods of regulation employed by the two kinds of organism are profound: the control of enzyme production used by bacteria can hardly be demonstrated in barley roots where allosteric effects and feedback inhibition are all important and allow the cells to respond more rapidly than if they depended on synthesis and destruction of enzyme protein.

Dr Ingle describes the intricate factors concerned in the production of large quantities of ribosomal RNA in cells, the devices used by plants to make sufficient copies of it and the variety of biochemical, genetic, cytological and analytical techniques that are used to investigate the problem.

Professor Street describes the behaviour of plant cells in tissue culture and shows the potential of the technique for investigating cellular metabolism without the complications of permeation into a cell mass and variations in cell types that bedevil experiments with isolated organs, or even separated tissues. The manipulation of free cells in a chemostat promises to provide a useful addition to the techniques available for biosynthetic and genetic investigations.

In the paper by Dr Daphne Osborne, the molecular events are followed through to their visible, morphological conclusions. She records the effects of ethylene on the favoured test object—the plumular hook of pea seedlings, and describes some of the results. Ethylene is a regulator of plant growth that has some of the attributes of an insect pheromone but its complex action is intimately concerned with the cell wall and its constituent hydroxyproline-rich proteins.

The other paper on the subject of plant hormones was given by Professor West, who reviews gibberellin biosynthesis and what is known of the regulatory processes that operate on it.

In his review Professor Cornforth discusses the influence the isotope effect has on the choice of hydrogen atom removed from a methyl group by iso-

pentenylpyrophosphate isomerase and relates the selection to the strict stereospecificity of enzyme reactions. The stereospecificity can be considered as a "labour-saving device"—in this case it is simpler to be highly specific. However, the same picture of integration and control is described in other reviews at higher and higher levels of complexity; at the atomic, molecular, enzymatic, pathway, organelle, cell and organ levels of organization there is precision and regulation.

Dr Sedgwick describes what is known of the biosynthesis of fatty acids in plants and how the processes are controlled. The reactions have been more thoroughly studied in animals and several of the regulatory mechanisms are different although recent results obtained by Dr Sedgwick, using chirally-labelled acetate, have shown that the stereochemistry of the reactions is the same.

The papers by Dr Briggs, who discusses the changes which occur during the germination of barley, and Professor Bradbeer, who describes the synthesis of enzymes and other materials which occurs during chloroplast formation, both show that these complex reactions are under precise control. As yet the way the control is operated remains largely unknown.

Professor Smith deals with current research on the regulation of flavonoid synthesis and concentrates in particular on the role of phenylalanine ammonia lyase (PAL). It is a branch-point enzyme for several different families of secondary products and its activity in extracts is affected by the previous illumination of the plant material from which it is extracted. The present evidence is insufficient to allow any definitive conclusions to be drawn concerning either the enzyme involved with the light-operated switch or even whether the change in overall activity depends on the synthesis of more enzyme protein or on adjustment of the activity of pre-existing molecules.

The final contribution of the Symposium, by Professor Fowden, provides a paradoxical contrast to the foregoing papers which all show evidence for the rigorous control of all aspects of biosynthesis. The non-protein amino acid, azetidine-2-carboxylic acid, occurs widely in the Liliaceae and can be used as a taxonomic marker as it occurs in a very few other species. Recently, as the result of a fortuitous set of circumstances, azetidine-2-carboxylic acid has been identified in sugarbeet where it occurs at very low concentrations. So low, in fact, that it would be undetectable were it not for the special processing which the juice undergoes.

The intriguing possibility suggested by this discovery is that azetidine-2-carboxylic acid occurs widely in plants as a metabolic mistake—as the result of imprecise selection of a substrate by an enzyme. It might be that azetidine-2-carboxylic acid is an example of "metabolic noise".

Physiological sciences have advanced to their present state of knowledge, largely by the use of a simple mechanistic outlook as expressed by the law of mass action. The "law of the minimum" or "limiting factor hypothesis" was

the first, and until recently, the major exception to this way of regarding complex processes but the self-regulating and interlocking pathways of metabolism are now coming to provide an even more serious exception. The investigation of self-regulating systems in living organisms by the "cause and proportional effect" design of experiment is inadequate because the integrated system may not respond at all to a stimulus, or respond in the reverse manner to that expected on the basis of a simple logic.

From the relative simplicity of the feedback loop, through the repressor/inducer/operator system of bacterial protein synthesis to the incompletely understood complexity of hormonal action, life processes show the capacity for self-regulation. By gathering together in one volume examples of the different kinds of control mechanism we can appreciate more fully the intricacy of metabolism and perhaps become more aware of the dangers of trusting a hypothesis when it accommodates the facts. As Whitehead said "Seek simplicity, then distrust it". Perhaps this book will help to make us more cautious in our interpretation of physiological data.

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