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Introductory Remarks

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Introductory remarks

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The three aspects of the enzymes of glycolysis – structure, activity and evolution – are, of course, closely interlinked, because catalytic activity, as well as regulatory properties, depend on structure. As for evolution, a relevant general principle of evolution by natural selection states that the chances of survival in a competitive environment are greatest if optimal use is made of resources. From this principle arises the question: are the structures of enzymes optimal or can more effective enzymes be visualized? In attempting to answer this question one must bear in mind that the main physiological significance of glycolysis is to provide energy under anaerobic conditions.

To make clear what I have in mind about the relations between evolution, structure and function, I should like to illustrate my point by a recent experience in a neighbouring field, that of aerobic energy-providing processes.

I have often asked myself for the reasons why metabolic cycles have evolved. Why, for example, is acetate oxidized by the apparently complex pathway of the citric acid cycle rather than by a 'direct' pathway via glycollate, glyoxylate, formaldehyde and formate? What are the advantages, if any, of the citric acid cycle? The answer to this long-standing question emerged only a few months ago in discussions that I had with Professor Jack Baldwin of Oxford. It is the function of this cycle to provide energy in a form that can be utilized by living cells, i.e. in the form of ATP. Hence the optimal mechanism of acetate oxidation is that which gives the highest yield of ATP. In evaluating the potential yield of ATP, it has to be borne in mind that the bulk of the synthesis of ATP is not directly connected with the degradation of foodstuff but with the transport of hydrogen atoms from NADH to molecular oxygen. Thus it is a prerequisite of ATP synthesis that the primary process of degradation is a reduction of the coenzymes (NAD, FAD).

If we look at the 'direct' pathway of oxidation of acetate from this point of view, we realize that this pathway is grossly disadvantageous because, as Baldwin pointed out to me, acetate cannot be dehydrogenated and therefore cannot donate electrons to the mitochondrial electron transport chain. Acetate can be oxidized by molecular oxygen, but only through monooxygenase. This enzyme system is not linked to the synthesis of ATP. It is not located in the mitochondria but in the microsomal fraction, and it catalyses the insertion of one oxygen atom of molecular oxygen into the organic substrate while the second oxygen atom is reduced to water. This reduction requires a second substrate that donates the electrons. In most cases, the second substrate is $NADPH_2$. The general equation of the oxygenase reaction is

$$\underbrace{R.CH_3 + O_2 + NADPH + H^+}_{\downarrow}$$
$$R.CH_2OH + H_2O + NADP^+,$$

where $R.CH_3$ stands for any methyl group that, because of the nature of R, cannot be dehydrogenated.

Not only is oxygenation of acetate wasteful because it cannot be coupled to ATP synthesis but, in addition, the consumption of one $NADPH_2$ for the reduction of the second O atom of O_2 robs the cell of a potential synthesis of 3 ATP molecules. By first approximation, half the energy set free in the oxidation of acetate by oxygenation is lost as heat.

This concept also offers an explanation of why the alternative to a 'direct' pathway must be a cyclic one. The principle is this: when a metabolic sequence of reactions cannot occur efficiently by a 'direct' route but necessitates a primary attachment of a substrate to another substance of low molecular mass (acetate to oxaloacetate), then this other substance (oxaloacetate) must be regenerated when the metabolic process of the substrate (acetate) has been completed. This is necessary, because were it not so, the other substance of low molecular mass would have to be produced afresh and this would be wasteful, if not impossible.

Thus a cyclic process is to be postulated as the only rational and economic mechanism for the organization of certain metabolic processes.

This concept explains also the evolution of other metabolic cycles, e.g. the ornithine cycle of urea synthesis, and I should like to suggest that it is always fruitful when studying structure, or reaction mechanisms, or metabolic pathways to examine the question of whether or why what has evolved in living organisms is optimally efficient. Usually this amounts to the question of whether something more efficient can be conceived on the basis of general chemical knowledge. I believe that the answer to this question will very likely be in the negative. But this does not mean that it is not worth while asking the question. On the positive side it may help in the understanding of biochemical and biological mechanisms.

I wonder, in particular, whether analogous questions should be raised for the enzymes of glycolysis. Would it be useful to ask whether improvements in structure (in relation to function) are feasible. Or have the enzymes, like many other features of living organisms evolved to perfection? Or, furthermore, can we detect differences in perfection achieved by evolution between the lowest and the highest forms of life?

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