2.2 Protein and Nucleic Acid Metabolism during Germination

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1. Introduction

We have reviewed studies on the metabolism of nucleic acids and proteins which occurs during germination in order to compare the ways in which protein synthesis is modulated in a wide range of fungal spores parasitic on higher plants. In this way we hope to provide an insight into the mechanisms by which protein synthesis is controlled during spore germination. These comparisons have also provided a view of the different degrees of dormancy encountered in the many types of spores. Most spores represent a quiescent phase of the life cycle, and we would expect ribosomes extracted from spores to be less active than ribosomes extracted from rapidly growing mycelia. These expectations have been largely borne out by experience. Newer studies have sought to determine how ribosome activity is shut down during sporogenesis and how the ribosome is activated during germination. Most research has not progressed to the point where a rigorous statement of the control mechanisms can be made, so we have attempted to present summary of the work under way.

2. Facultative Parasites

2.1 Germination of Conidia

2.1.1 General

Conidia are dikaryotic or diploid vegetative spores. Typically when wetted, they consume water, swell, begin to increase in dry weight, then produce a germ tube. Yanagita (1957) has divided conidial germination in Aspergillus niger into three morphologically recognizable phases as endogenous swelling, exogenous swelling and sprouting. The first swelling phase, in contrast to the second phase, is not influenced by severe environmental factors, especially by the concentration of carbon dioxide. The timing of these phases varies with the species of conidium and the environmental conditions used to induce germination, but there seems to be general agreement that conidial germination can be described adequately by these three phases. The increase in conidial dry weight occurs during exogenous swelling which precedes germ-tube emission.

In studies on intact conidia, it must be borne in mind that lack of effect of an inhibitor may mean that the inhibitor has not penetrated. To assume that a metabolic function is inactive because an inhibitor failed to act is to ignore results
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as recent as those of TisDALE and DeBUSK (1972). These authors demonstrated that conidia of N. crassa are impermeable to actinomycin D and α-aminitin.

2.1.2 Changes in Macromolecules Related to Protein Synthesis during Germination

YANAGITA (1957) was one of the first to study the effects of germination on conidial synthesis of proteins and nucleic acids. In his studies on A. niger conidia, he and his coworkers found that the spores imbibed water to increase their volume about 2.5 times. The earliest cellular synthesis detected was the synthesis of nucleic acid which accompanied exogenous swelling. At the end of this phase, accumulation of amino acids was observed followed by a synthesis of proteins. A conspicuous increase in oxygen consumption commenced in parallel with the active synthesis of protein, and this was followed by the emission of germ tubes.

Recently, BAINBRIDGE (1971) extended these studies using conidia of Aspergillus nidulans (Erdam) Winter. He reported that upon inoculation of spores into a liquid medium, the spores consumed water and synthesized RNA within 30 min, together with an increase in Kjeldahl nitrogen. Protein synthesis was detected within 150 min and synthesis of DNA within 180 min. Germ tubes were first detected by 210 min, and almost all of the spores produced germ tubes within 360 min. About half of all of the germ tubes was produced between 240 and 300 min, indicating that at least a partial synchrony of germination was achieved. Spores containing 2, 4 and 8 nuclei were first detected at 270, 390 and 480 min, respectively.

HORIKOSHI and IKEDA (1969) studied protein synthesis in germinating conidia of A. oryzae. They also found that incorporation of a mixture of amino acids into protein was biphasic with an early incorporation occurring in the first 30 min followed by a very rapid upturn thereafter. Germ tubes appear after 3 h in this species (TOKORO and YANAGITA, 1966).

Alanine and adenine have been shown to be essential for complete germination of conidia of A. oryzae (TsAY et al., 1965). In an examination of this effect, TsAY and HANAOKA (1972) found that adenine was required for the initiation of vegetative growth, while alanine was required for initiation of germination. Adenine must be available from 40 to 90 min after germination begins to be effective. During this period the rate of RNA synthesis rises rapidly, and adenine synthesis is apparently not adequate to provide sufficient nucleotides for this accelerated synthesis.

Protein synthesis was also retarded if adenine was omitted during this important period, possibly from a lack of ATP. TsAY and HANAOKA (1972) showed that synthesis of DNA, which occurs 140 min after initiation of germination, was inhibited almost completely if protein synthesis was inhibited by addition of cycloheximide at 60 min. Thus the 40 to 90 min transient period was a highly critical time for the synthesis of proteins important for the initiation of vegetative growth. The work focuses attention on the fact that there are two separate phases of protein synthesis in these conidia—one phase involved in spore germination and the other in vegetative growth.

In a comparative study of several saprophytic and parasitic fungi, HOLLOMON (1970) reported that macroconidia of Neurospora crassa readily incorporated 3H-uridine into RNA. An analysis of nucleic acids by electrophoresis on polyacrylamide gels showed that all classes of RNA had been synthesized. At first, uridine was