Gibberellic acid, abscisic acid or cytokinins could not substitute for the indol-3-ylacetic acid. The efficacy of authentic exogenous glutamine and indol-3-ylacetic acid in substituting for the embryo in inducing both lipase activity and triglyceride metabolism in the storage tissues of wheat grain indicates that these substrates might also be responsible for the induction of triglyceride mobilization *in vivo* during germination. The proof of this point must await the isolation of indol-3-ylacetic acid from germinating wheat. It is known, however, that wheat produces considerable quantities of glutamine during germination (Chibnall, 1939), and, perhaps significantly, indol-3-ylacetic acid has been detected in the grain of *Zea mays* (Powell, 1964).

The metabolism of the triglycerides in the bran of the wheat was investigated further by incubating the bran in the absence of the starchy endosperm. Under these conditions no triglyceride metabolism occurred, but the addition to the medium of either an aqueous extract of the starchy endosperm or authentic cytokinins at hormonal concentrations induced the metabolism of 22% of the bran triglyceride in a pattern identical with that in the bran of incubated endosperm halves. These results suggest that a cytokinin from the endosperm is responsible for the mobilization *in vivo* of a part of the triglyceride reserves in the bran. The suggestion is supported by the known occurrence of a cytokinin in the endosperm of *Zea mays* (Letham & Miller, 1965), but the hormone has not yet been isolated from wheat.

With regard to the nature of the induction processes, the inductions of lipase activities by glutamine and indol-3-ylacetic acid were strongly inhibited by proflavin, actinomycin D, cycloheximide and puromycin. This indicates that both RNA and protein synthesis are required for the inductions. The induction by cytokinin of the non-lipase triglyceride metabolism in the bran was strongly inhibited by proflavin, cycloheximide and puromycin, but not by actinomycin D, indicating a requirement for protein synthesis but possibly not for RNA synthesis.

In summary, it is proposed that the induction of triglyceride mobilization in the reserve tissues of the germinating wheat grain is controlled by the embryo. The hormonal induction processes involved are, however, distinct from that which induces the mobilization of reserve carbohydrates in the same species.

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An Examination of the Evidence for Stable Messenger Ribonucleic Acid in Seed

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It has been suggested that, in the early stages of the germination of certain seeds, protein synthesis is directed by a long-lived mRNA. The question whether such a long-lived mRNA is present in seed is important in considering the mechanism of the initiation of germination. The identification of mRNA presents a number of difficulties and it is the purpose of this communication to examine in detail the evidence for long-lived mRNA in seed.

Actinomycin D, which is known to interfere with transcription (Reich, Franklin, Shatkin & Tatum, 1962), has been reported to be without effect on protein synthesis in some seed (Dure & Waters, 1965; Waters & Dure, 1966; Marcus & Feeley, 1964a; Marcus, 1969; Chen, Sarid & Katchalski, 1968a), but there is some disagreement on the effects of this drug on enzyme development (Gientka-Rychter & Cherry, 1968). The conclusion that the absence of effect on protein synthesis is due to the process being independent of transcription is open to the criticism that the drug may fail to be taken up by the tissue, as was observed by Neumann (1964) and Abdul-Baki (1969). In the experiments of Chen *et al.* (1968a) it was shown that actinomycin D is taken up by wheat seed, but the possibility remains that it does not penetrate to the site of RNA synthesis. Doubt is also cast on the conclusion by the possibility, suggested by Chrispeels & Varner (1967), that synthesis of RNA may take place before the drug becomes effective.

The presence of polyribosomes in ungerminated seed, which would suggest the existence of stable mRNA, has not been established unambiguously (Marcus & Feeley, 1965; Jachymczyk & Cherry, 1968; Barker & Rieber, 1967) and, although it is well established (Marcus, 1969) that polyribosomes are formed in the early stages of germination, it has not been demonstrated whether they are formed from

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newly synthesized or pre-existing RNA. Thus whereas early experiments failed to reveal the occurrence of RNA synthesis in the initial phase of the germination of peas (Barker & Hollinshead, 1964), the use of higher extents of radioactive labelling enables synthesis to be recognized at an earlier stage (G. R. Barker, C. M. Bray, M. A. Detlefsen & C. J. Kemp, unpublished work). Moreover, it is significant that RNA synthesis is not completely abolished by actinomycin D in cotton seed (Dure & Waters, 1965; Waters & Dure, 1966), in barley seed (Chrispeels & Varner, 1967) and in peanuts (van Huystee & Cherry, 1966).

Attempts have been made to recognize mRNA by hybridization with DNA (van Huystee & Cherry, 1966), but this technique, which requires labelled mRNA, is difficult to apply to long-lived messenger. Chen, Sarid & Katchalski (1968b) tried to circumvent this difficulty in studies on wheat embryo. The assumption was made that the mRNA of the dry embryo is the same as that labelled by ^{32}P between 24 and 48 h of germination. This RNA was hybridized with DNA and was found to be displaced from the complex equally by unlabelled RNA from seed germinated for 24 or 48 h and by RNA from ungerminated seed. It was concluded that mRNA is conserved in the dry seed. However, not only is the correctness of this conclusion dependent on the reliability of the above assumption, but also there is no certainty that mRNA forms a duplex with DNA under the conditions of the experiment (Waddington, 1969; Melli & Bishop, 1970). Even if DNA becomes saturated with mRNA under the conditions of hybridization, the results of Chen *et al.* (1968b) indicate that saturation of DNA with total RNA from wheat embryos requires 50 times more RNA for a given weight of DNA than is the case with either rRNA or tRNA from the same source. This suggests that RNA species other than rRNA and tRNA represent a very small proportion of the whole. Thus it is questionable whether the presence or the absence of mRNA would be detectable in the competition experiments described.

Incorporation of amino acids into protein in cell-free systems from peanuts, which is stimulated by polyuridylic acid and by certain fractions of RNA from germinated seed (Marcus & Feeley, 1964b; Jachymczyk & Cherry, 1968), is unaffected by fractions of RNA from dry seed. On the other hand, incorporation of amino acids into protein by cell-free systems from wheat seed is stimulated by RNA prepared from dry embryo (Chen *et al.* 1968b; Weeks & Marcus, 1970, 1971), and such RNA also reacts *in vitro* to convert monoribosomes into polyribosomes. Thus the situation may well be different in these two types of seed.

It is concluded that, in general, the evidence for

long-lived mRNA in seed is unreliable, but that there is a *prima facie* case for its existence in wheat seed. However, apart from the possibility of messenger-independent incorporation of amino acids into protein (Novelli, 1969), it has been emphasized (Boulter, 1970) that characterization of natural mRNA requires recognition of the protein synthesized. The possibility of the presence of long-lived mRNA in ungerminated seed must be regarded as tentative.

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Preformed Messenger of Quiescent Wheat Embryos

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Initiation of protein synthesis in the early phases of seed germination is correlated with the rapid conversion of a considerable part of the monoribo-