some way, but we have no reason to believe that the enzymes initially present in the egg have been subjected to a significantly reduced possibility of error. Various solutions seem possible; for example, selection in the growing embryo may be strong enough or the process of embryogenesis may demand such a high accuracy that its successful completion guarantees the necessary accuracy of protein synthesis. One can also conceive of special mechanisms of quality control; for example, special proteins might be synthesized which are converted by a certain class of errors into lethal polypeptides. This would guarantee that the frequency of this class of errors in viable cells is kept low. At present there is no evidence available which enables one to select among these possibilities.

I wish to make it quite clear that I am not proposing here that the accumulation of protein transcription errors is "the mechanism of ageing." My object is the more modest one of pointing out one source of progressive deterioration of cells and cell lines. Since I am unable to estimate the time scale of this process, I can only suggest experiments which should show where, if anywhere, it contributes to the ageing process in higher organisms.

I am indebted to Professor H. C. Longuet-Higgins and Dr. F. H. C. Crick for valuable criticisms of my original manuscript.

3 Benzer, S., and S. P. Champe, these PROCEEDINGS, 47, 1025 (1961); 48, 532 (1962).
4 Note that in an exponentially growing culture there is a strong selection for cells with a short division time.

DYNAMIC ROLE OF TRIPLET STATES IN PHOTOSYNTHESIS*

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The purpose of this communication is to point out some attractive features of primary photosynthetic steps based, partially at least, on energy transfer and energy multiplication mechanisms which involve the triplet state of chlorophyll. A more critical comparison of triplet state mechanisms, singlet state mechanisms, and photoionization mechanisms will be deferred until a later time. Here we strive only to correct some misunderstandings concerning the possible role of triplet states in photosynthesis and to speculate upon certain mechanisms often excluded from past thinking because of the misunderstandings.

Population of Triplets.—There seems little doubt that efficient singlet \( \rightarrow \) triplet radiationless transitions (intersystem crossing) can occur in molecular aggregates, providing processes such as radiation or chemical quenching which limit the lifetime of the singlet state are not too fast. In the case where excitation transfer is
slow compared with the singlet-triplet radiationless transition time, the process is not fundamentally different from that which occurs for a molecule in an inert solvent\textsuperscript{1--3} such as a hydrocarbon "glass." When the excitation transfer time is fast compared with the time for intersystem crossing, the probability of transfer divides itself among a number of molecules, but the over-all rate remains the same as for the localized case, providing the molecular vibrations are not affected by the fast excitation transfer. In the particular strong coupling limit\textsuperscript{4} in the singlet where the vibrations of the aggregate in the excited state are different than they are for localized excitation, the singlet-triplet transition probability is different by virtue of its dependence on vibrational factors\textsuperscript{5,6} in the transition matrix elements. However, these factors and the associated transition probabilities are probably larger than they are when both excitations are localized, since the magnitude of vibrational factors between two nondegenerate electronic states often increases with increasing nonorthogonality of the vibrational eigenfunctions in the two states. Furthermore, the decreased energy separation between singlet and triplet states of the aggregate as compared to that in the isolated molecule should also enhance intersystem crossing.\textsuperscript{5} Thus, we conclude that in almost all cases, except the unreasonable one where both singlet and triplet exciton interactions are in the strong coupling limit, the radiationless intersystem crossing process in a molecular aggregate should have the same order or higher transition probability than that normally found in rigid glass experiments. The arguments in the weak and intermediate coupling limits are particularly applicable to chlorophyll \textit{in vivo} where absorption and fluorescence spectra imply that the intermolecular coupling is weaker than in crystalline chlorophyll.\textsuperscript{6}

The expectation of efficient intersystem crossing in aggregates is borne out in mixed crystal experiments\textsuperscript{7} with organic molecules. More important, a certain amount of information about intersystem crossing \textit{in vivo} is implied from existing fluorescence quantum yield measurements. It is known that monomeric chlorophyll-\textit{a} in ether or methanol solution under 430--436 m$\mu$ excitation shows a fluorescence quantum efficiency of about 33 per cent.\textsuperscript{8} Similarly, recent measurements by Goedheer\textsuperscript{9} using $\sim$615 m$\mu$ excitation of \textit{monomeric} chlorophyll-\textit{a} both in $10^{-4}M$ methanol solution and in postetiolated bean leaves show nearly identical fluorescence quantum efficiencies, the \textit{in vivo} to \textit{in vitro} ratio running between 0.6--1.2 for a series of experiments. Such experiments indicate that the rate of intersystem crossing in monomeric chlorophyll-\textit{a} \textit{in vivo} or \textit{in vitro} may be up to twice that for the purely radiative process from the singlet state. Even though the 67 per cent upper limit to the efficiency can be achieved only in the absence of other nonradiative processes, it appears that intersystem crossing might be an important path by which excited singlet states are depleted in monomeric chlorophyll-\textit{a}.

In \textit{aggregated} chlorophyll \textit{in vivo}, efficient photosynthetic processes further reduce the lifetime of the excited singlet. Such an effect would be anticipated from the over-all high quantum efficiency of photosynthesis and is evidenced by the greatly reduced fluorescence quantum efficiency in mature plants. Taking 2 per cent to be the fluorescence quantum efficiency for chlorophyll aggregates \textit{in vivo}\textsuperscript{8,9} and assuming \textit{no change in the rate of intersystem crossing in the aggregate}, it would appear that at most only about 4 per cent of the singlets undergo intersystem cross-
ing to the triplet state. Such a low efficiency would hardly be important. If it actually existed in the photosynthetic system, the arguments presented in this paper would not be pertinent. On the other hand a 10-fold increase in the rate of intersystem crossing in the aggregate would enable triplet state population to be important even in the face of strong "chemical quenching" of the singlet state. As suggested in the first paragraph of this section, an increase of such magnitude may not be unreasonable.

Some evidence for an increased efficiency of intersystem crossing in \textit{in vivo} chlorophyll aggregates does in fact seem to exist. Experiments\textsuperscript{10} have shown that in living cells the normal 2 per cent fluorescence efficiency rises to 4 per cent upon inhibition of photosynthesis. Assuming that the radiationless transition from the excited singlet state directly back to the ground state is unimportant in the aggregate, this result implies that under inhibition of photosynthesis, where the excited states can do no photochemistry, 96 per cent of the excited molecules do indeed undergo intersystem crossing to the triplet state. The rate of intersystem crossing in the \textit{in vivo} aggregate therefore appears to be about 24 times that for fluorescence radiation. In the case where photochemistry is not quenched by an inhibitor, use of the 2 per cent fluorescence efficiency leads one to expect quantum efficiencies of 48 per cent for intersystem crossing and 50 per cent for photochemistry from the excited singlet state.\textsuperscript{11} These estimates are relatively crude because it has been assumed that inhibition changes only the efficiencies, but not the rates of the various processes which take place in the \textit{in vivo} system. More important, this simple picture would be severely complicated by the presence of two quantum processes (\textit{vide infra}) involving singlet or triplet states. Nevertheless, one is still tempted to say that a good fraction of the excited \textit{in vivo} chlorophyll-a molecules at least reach the triplet state. In order for the triplets to contribute to photochemistry in the plant, they must do something useful during their $\lesssim 10^{-3}$ sec lifetime.\textsuperscript{12, 13} Later in the paper we will point out that the triplets should indeed be able to do something very useful during this period of time.

One may at first think that the population of triplet states in a photosynthetic system should be accompanied by some phosphorescence (triplet-ground state emission) and might conclude that, since none has ever been observed, the existing evidence is therefore against triplet state population. There are three reasons why phosphorescence is not expected from \textit{in vivo} aggregates of chlorophyll molecules, even though the triplets do get populated. The first reason has to do with the nature of monomeric chlorophyll in particular, the second concerns the behavior of triplet states in molecular aggregates in general, and the third has to do with the fact that, if efficient photochemistry from the triplet state is to occur, the triplets have little time to phosphoresce. The first two points will be discussed separately later in the paper under the headings of \textit{Quenching of Triplets} and \textit{Triplet-Triplet Annihilation and Energy Multiplication}. It develops that a low phosphorescence quantum yield need in no way be related to the efficiency of intersystem crossing to the triplet state.

\textit{Triplet Energy Transfer}.—It has often been remarked that "the transfer of triplet energy in molecular aggregates is slow because the required overlap of electronic eigenfunctions in the intermolecular region must be small." This statement only partly hits the mark. The interaction energy $\beta$\textsuperscript{14} for triplet excitation transfer in
hydrocarbons does depend primarily upon electron exchange\textsuperscript{16} which, in turn, depends on intermolecular overlap of the wave functions. The interaction energy, however, need not be excessively small, being of the same order of magnitude as that for lowest singlet excitation transfer in certain $\pi$-electron molecules.\textsuperscript{7-16} The principal reason for this is that electronic eigenfunctions for a many-electron system fall off considerably less rapidly than $\exp \left(-Z_{\text{eff}}r/n\right)$ by virtue of the decrease toward unity of the effective nuclear charge $Z_{\text{eff}}$ at large electronic distances $r$.\textsuperscript{17} A direct experimental determination of the nearest neighbor $\beta$ for crystalline benzene gave\textsuperscript{16} $0.0015$ $e$-$\nu$ (12 cm$^{-1}$) corresponding to a time for one excitation transfer event of about $4 \times 10^{-13}$ sec! Such a fast process can easily overwhelm other radiative and nonradiative processes from the triplet state.

Theoretically, the exchange interaction can be shown to be dependent upon the mutual orientation of the molecules,\textsuperscript{16} the interaction of greatest magnitude occurring when the $\pi$-orbitals on each molecule are pointing directly toward one another. It is important to point out that the triplet interaction, unlike that for certain singlets,\textsuperscript{18} has nothing whatsoever to do with the strength or polarization direction of the transition from the ground state to the triplet state. It would truly be coincidental if the optimum orientation for greatest heat of aggregation in the ground state, optimum orientation for singlet transfer, and that for triplet transfer were all the same. It should be realized, however, that even when the orientations are not optimum, as in crystalline benzene, energy transfer may still occur with high efficiency.

The relatively large triplet interaction energy is not the only reason that triplet energy transfer in molecular aggregates is important. The lifetime of the triplet state, being much longer than that of the singlet, allows ample time for triplet energy transfer even in the absence of large interaction energies. Thus, the two points together, moderate interaction energies and intrinsically long triplet state lifetimes, cause triplet energy transfer in many kinds of organic aggregates to be as important as singlet transfer, or more so.\textsuperscript{19} There is therefore no obvious reason why the triplet state should not provide an efficient means of transferring energy through chlorophyll aggregates.

Another statement often made is that triplet energy transfer, being a short range interaction, dies off exponentially with distance, so that at most only nearest-neighbor transfer is ever expected. Again, this statement is only partially true. Certainly, for two interacting molecules in a vacuum, the exchange term for fixed $Z_{\text{eff}}$ does drop off exponentially with distance at large distances. In real molecules the exchange term diminishes considerably more slowly than this, because $Z_{\text{eff}}$ is itself decreasing. More important, in a molecular aggregate the transition probability for excitation transfer does not depend solely upon the direct (vacuum) interaction between two molecules. Any interactions which can occur by virtue of the fact that the molecules are both coupled to the whole system, and are thus indirectly coupled to one another, will contribute to the transition probability. In many cases the indirect interactions are expected to outweigh the direct ones by far.

Indirect interactions have been found to be responsible for long-range tunneling of triplet excitation between energy traps (sinks) in a host crystal when the triplet states of the host are nearby. The mean time for this transfer in the case where the aggregate is one dimensional is given by\textsuperscript{20}
\[ \tau \approx \hbar \Delta E N \beta' - (N-1) \beta^{-2} \]

for both \( \beta \) and \( \beta' \ll \Delta E \), where \( \Delta E \) is the energy difference between the triplet states of the guest and the host, \( \beta \) is the nearest-neighbor guest-host vibrational-electronic interaction energy, and \( \beta' \) is the nearest-neighbor host-host interaction. \( N \) is the number of intervening host molecules. The time \( \tau \) is expected to be concentration-dependent because of the exponential factor \( N \), and is expected to be temperature-dependent because of the fact that \( \Delta E \) effectively decreases with increasing temperature. This mechanism provides a good way by which triplet states can undergo long-range energy transfer in molecular aggregates containing energy traps. In the instance where the energy traps are shallow or where there are no traps, the simple formula above does not apply. Nevertheless, in such cases an analogous type of long-range transfer of energy occurs. Thus, it is possible for triplet excitation to migrate efficiently either through pure aggregates or through aggregates containing impurity or defect traps.

Quenching of Triplets.—Radiationless processes (intersystem crossing) between a singlet state and a triplet state have been shown to depend upon the strength of the spin-orbit coupling between the two states and upon the magnitude of the vibrational Franck-Condon factors associated with the two states.\(^2\)\(^,\)\(^3\)\ The Franck-Condon factors range from zero to unity, and, for the latter value, singlet-triplet radiationless transitions between \( \pi \)-electron states of hydrocarbons take place in roughly \( 10^{-10} \) sec.\(^2\)\(^1\) Such high rates would easily dominate fluorescence which occurs in only about \( 10^{-8} \) sec, and would even be able to compete successfully with the faster \textit{in vivo} singlet state mechanisms in the photosynthetic system.

The Franck-Condon factors are very much smaller than unity for singlet and triplet states of widely different electronic energy, but approach unity as the two electronic states get close together. This point is especially pertinent in the process analogous to intersystem crossing which we call triplet state quenching, that is, the radiationless transition between the first excited triplet state and the ground state. One sees that triplet states lying relatively close to the ground singlet are expected to be quenched rapidly by a radiationless transition to the ground state. The over-all triplet state lifetime \( \tau_T \), which is the reciprocal of the sum of the triplet radiative and nonradiative rates, therefore decreases markedly for decreasing triplet state energy \( E_T \). Furthermore, of the molecules reaching the triplet state, the fraction \( f_T \)\(^2\)\(^2\) which phosphoresce is expected to decrease rapidly with decreasing \( E_T \). The series anthracene\(^2\)\(^3\) (\( E_T = 1.82 \) e-v, \( \tau_T < 0.1 \) sec, \( f_T = \) very low), naphthalene\(^2\)\(^4\) (\( E_T = 2.64 \) e-v, \( \tau_T = 2.6 \) sec, \( f_T = 0.16 \)), and benzene\(^2\)\(^5\)\(^,\)\(^2\)\(^6\) (\( E_T = 3.66 \) e-v, \( \tau_T = 7.0 \) sec, \( f_T = 0.25 \)), illustrates this point for simple hydrocarbons. In chlorophyll-\(b\) Becker and Kasha\(^1\)\(^3\) have found that the triplet state lies near 1.43 e-v (865 m\( \mu \)). Such a low-lying triplet state is expected to be subject to rapid quenching. Similarly, the triplet state of chlorophyll-\(a\) lies very close to the ground state (1.40 e-v, 885 m\( \mu \)).\(^2\)\(^6\) The \( 10^{-3} \) sec triplet state lifetime\(^1\)\(^2\)\(^,\)\(^1\)\(^3\) of monomeric chlorophyll is therefore doubtless caused primarily by nonradiative transitions from the triplet state back to the ground state, and it is not unexpected that the quantum yield for phosphorescence emission is low. There is no reason to believe that, associated with chlorophyll in aggregates, there is not a similar quenching mechanism.

A low phosphorescence quantum efficiency for the monomeric chlorophylls has
been indicated in the experiments of Becker and Kasha, a result which should not be construed to mean that the triplet state is not populated efficiently, but instead that the quenching rate as expected for the low-lying triplet state is high.

**Triplet-Triplet Annihilation and Energy Multiplication.**—In a molecular aggregate, phosphorescence and nonradiative quenching directly back to the ground state are not the only ways by which triplet states can disappear. *Triplet excitations on two different molecules can mutually annihilate one another, so that one of the two molecules goes back to the ground state while the other is promoted to a higher electronic state having up to twice the energy of the triplet state!* Spin selection rules allow this higher state to be a singlet, a triplet, or a quintet, where, respectively, zero, two, or four electrons are unpaired.

The interaction energy for the triplet annihilation process, like the triplet transfer process, depends upon electron exchange. Long-range triplet annihilation interactions through a vacuum are therefore not very strong. However, just as for long-range trap-to-trap triplet energy transfer in a molecular aggregate, the annihilation process itself can occur at long range, providing host states are relatively low-lying. The existence of excitation residing on nearest neighbors is therefore not an important requirement for efficient annihilation in such cases. Furthermore, it can also be shown that, either in pure molecular aggregates or in aggregates containing a low concentration of shallow traps or a high concentration of deep traps, the triplet energy transfer rate, as well as the annihilation rate, can both far exceed the rate at which radiation or simple quenching of the triplet state back to the ground state occurs. In other words, by virtue of the relatively long triplet state lifetime for radiation plus quenching, an efficient mutual annihilation of triplets in molecular aggregates can occur which allows triplets to be converted into high energy quanta. Thus, an important *energy multiplication* process can take place by way of the triplets. In pure organic crystals the annihilation process may serve only to quench phosphorescence rapidly; in mixed organic crystals, long-range annihilation between traps can partially quench phosphorescence to produce higher singlets and lead to delayed fluorescence; in a photosynthetic system, the mutual annihilation of triplets can lead to useful high energy photochemistry.

It must be borne in mind here that even though annihilation is a two-quantum process whose rate depends upon the square of the concentration of excited states, the dependency of the annihilation rate upon light intensity remains linear in the limit of fast annihilation. This fact follows from an examination of the over-all kinetics. Physically it is an expected result since, in the limit where the major cause of triplet disappearance is bimolecular annihilation rather than unimolecular quenching, the stationary triplet concentration depends on the square root of the rate at which triplets are produced, i.e., on the square root of the light intensity; and the rate of triplet-triplet annihilation as well as the rates of processes which depend upon it, such as delayed fluorescence and high energy photochemistry, depend linearly on light intensity. The well-known linear variation of photosynthetic yield with light intensity in the range of low intensities up to saturation intensities is therefore not at variance with the presence of a fast triplet-triplet annihilation step in the photosynthetic mechanism.
Summary.—In this communication the possible importance to photosynthesis of triplet state quenching, triplet energy transfer, triplet–triplet annihilation, and triplet energy multiplication has been pointed out. The more restricted role of triplets in photosynthesis discussed by Franck and others can now be broadened to include these more efficient processes. The presence of triplet mechanisms in the photosynthetic apparatus would not, of course, rule out quenching, energy transfer, annihilation, and energy multiplication involving singlet states; nor does it rule out annihilation and energy multiplication steps via either the triplet or singlet states to produce charge separation with further energy migration or chemical processes involving the charge carriers. It would not be surprising if two or more of these types of energy transfer mechanisms combine in the primary photochemical steps. Such might be anticipated from Emerson enhancement effects which indicate that chemical cooperation on a relatively long time scale (~5 sec) takes place between the products of primary light absorption at different wavelengths. Thus, two-quantum physical mechanisms may combine in a "two-quantum" chemical process in over-all photosynthesis.

The presence of shallow energy sinks in the photosynthetic apparatus seems assured by the existence of electronically similar but slightly different pigments. These energy sinks are chlorophyll-a molecules which appear to be in very special chemical environments or in very special states of aggregation. One might speculate that at room temperature efficient energy transfer and annihilation may occur between the energy sinks themselves (according to what was said earlier in the paper about long-range effects), provided the energy sinks are appropriately coupled through the nearby states of their own accessory pigment system. However, at fairly low temperatures the transfer of energy may be retarded. At such temperatures where triplet energy transfer is relatively slow and where photochemistry has been quenched, triplet–triplet annihilation may reveal itself as delayed fluorescence having a nonexponential decay of similar half life as the triplet state. That low temperature emission of this kind has already been in fact observed perhaps may be direct evidence for a dynamic rather than a static role of triplet states in photosynthesis.

* Supported in part by the U.S. Atomic Energy Commission, the U.S. Army Research Office, and the National Science Foundation.
† Alfred P. Sloan Fellow.
‡ Contribution No. 2943.
3 Ibid., in press.
11 Franck, J., J. L. Rosenberg, and C. Weiss, Jr., in Luminescence of Organic and Inorganic
14 Becker, R. S., and M. Kasha, J. Am. Chem. Soc., 77, 3669 (1955); see also ref. 26 of the present paper.

The interaction energy referred to here is the transition matrix element for a radiationless transition between an initial state \( \phi_{A\uparrow} \) and a final state \( \phi_{A\downarrow} \), where the dagger represents electronic singlet or triplet excitation and the A and B represent two different molecules. The characteristic time for such a transition is given roughly by \( \hbar^2 \) where \( \hbar \) is Planck’s constant divided by \( 2\pi \).

\[ \text{Slater, J. C., Phys. Rev., 42, 33 (1932).} \]
\[ \text{Foerster, Th., Discussions Faraday Soc., 27, 7 (1959).} \]

Assume as an upper limit that the singlet interaction in crystalline chlorophyll is given by the energy difference between the crystal absorption maximum at 720 m\( \mu \) and the absorption maximum of the monomer at 660 m\( \mu \). This energy is at most only 100 times that for the triplet interaction and yet the lifetime of the singlet is certainly more than 100 times shorter than the triplet lifetime.

\[ \text{Corresponding to a } \beta \text{ of about } 10^{-4} \text{ e- } \text{v (0.08 cm-1); spin-orbit interactions between states surely fall within two orders of magnitude of this value.} \]
\[ \text{In the absence of quenching of the lowest singlet, } \beta = \frac{Q_{F}}{1-Q_{F}}, \text{ where } Q_{F} \text{ and } Q_{F} \text{ are the quantum efficiencies for phosphorescence and fluorescence. In general } \beta = \frac{\tau_{I}}{\tau_{F}}, \text{ where } \tau_{I} \text{ is the radiative triplet state lifetime, and } \tau_{F} \text{ is, as before, the over-all triplet state lifetime.} \]
\[ \text{Lim, E. C., J. Chem. Phys., 36, 2497 (1962).} \]

Quenching back to the ground state in a way described in the last section is, of course, but one of many ways that quenching can occur. Quenching by impurities and by chemical reactions are two other ways. Triplet-triplet annihilation described in this section is another more complicated way by which depletion of triplets can occur.

The lowest quintet state of chlorophyll should not be overlooked. According to estimates of the same type made by Gouterman,29 the lowest quintet state of chlorophyll should lie roughly in the range 2.34–3.27 e-\text{v}, an energy region about double that of the lowest triplet. Therefore (vide infra), the lowest quintet state appears to be accessible as a final state for triplet-triplet annihilation. Since the lowest quintet state is expected to have a lifetime (for intersystem crossing to lower triplets) possibly as long as \( 10^{-4} \text{ sec, relatively slow chemical steps or further annihilation steps can occur from the quintet state. Thus, while singlets or triplets having the same energy may not be able to carry out efficient photochemistry or annihilation because of their short lifetime, the quintet by living longer might be able efficiently to carry out chemistry or annihilation at roughly double the energy of the lowest triplet. The author wishes to acknowledge a helpful discussion with George S. Hammond concerning quintet state chemistry.} \]
\[ \text{Gouterman, M., J. Chem. Phys., 33, 1525 (1960).} \]

Shallow and deep compare the electronic energy difference \( \Delta E \) between guest and host with the interaction energy \( \beta \) for the radiationless transition. A deep trap is one where \( \Delta E \gg \beta \), while a shallow one is where \( \Delta E \lesssim \beta \).

\[ \text{See, for example, French, C. S. in Light and Life, ed. W. D. McElroy and B. Glass (Baltimore: Johns Hopkins University Press, 1961), pp. 463–464.} \]
\[ \text{Rabinowitch, E., J. Phys. Chem., 61, 870 (1957); Kasha, M., Rev. Mod. Phys., 31, 162 (1959).} \]
S. A. Rice, Phys. Rev. Letters, 8, 410 (1962) and J. Chem. Phys., in press. However, annihilation of singlets can produce only ionization or short-lived singlet states but no triplets or quintets.


35 Emerson, R., R. F. Chalmers, and C. Cederstrand, these Proceedings, 43, 133 (1957).


37 For example, a long wavelength form (\(\sim 700 \text{ nm}\)) of chlorophyll-\(a\) probably acts as an energy sink for its own "accessory" pigments \(C_a\) 683 and \(C_a\) 695.\(^{38}\) The energy difference here between the "sink" and the "accessory" pigments is sufficiently small that the "sinks" act only as very shallow traps for singlet excitation but probably as somewhat deeper traps\(^{39}\) for triplet excitation. Efficient long-range transfer and annihilation effects at room temperature may occur between the sinks in both instances. The excitation, however, would be mostly localized near the sink pigments rather than spread equally throughout the photosynthetic unit, an advantage from a photochemical point of view. For the very shallow sinks which are presented to the singlet excitation, the room temperature fluorescence from the system is expected to originate primarily from the accessory pigments which are present in excess, while at low temperatures, the fluorescence should arise mostly from the energy traps. Certain recent experiments\(^{40}\) can be interpreted in this way.


1 See, for example, Tollin, G., E. Fujimori, and M. Calvin, these Proceedings, 44, 1035 (1958). Near room temperature there persists a very long-lived fluorescence which presumably is caused by partial reversal of enzymatic steps.\(^{38}\) This very slow component cannot be caused directly by triplet annihilation, but the shorter-lived components can be.


THE SPECIES VARIATION OF RNA METHYLASE*

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The presence of the methylated purines and pyrimidines in soluble RNA presents two questions: their origin and function. Assuming that all nucleic acid sequences are predetermined by the bases in DNA, the origin of nucleotide chains bearing methylated bases from a sequence of DNA which is devoid of them, appeared problematical. This anomaly was recently resolved by the demonstration of the existence in E. coli of an enzyme system (RNA methylase) which methylates s-RNA at the polynucleotide level\(^{1,2}\) using S-adenosyl-methionine as the methyl donor\(^3\) for all bases, including thymine.\(^4\)

A variety of functions has been ascribed to the methylated bases of s-RNA, ranging from their being mere disposal systems for surplus methyl groups to a pivotal role in the coding for amino acids. These hypotheses have been based on studies of the levels of the bases in the RNA of different species.