

mine with accuracy; however, the observed ratio of counts incorporated into cell material to counts evolved as CO₂ was of the order of 90:1. This corresponds to a molar ratio of acetate assimilated to CO₂ evolved of approximately 0.02, some ten per cent of the value which can be inferred from Muller's data on acetate metabolism by purple sulfur bacteria. It is therefore evident that *Chlorobium* has a much more limited ability to form CO₂ from acetate than do the purple sulfur bacteria.

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ON THE ELECTRON-DONATING PROPERTIES OF INDOLES*

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It has been noted that indoles, such as 5-hydroxyl tryptamine (serotonin) form charge transfer complexes with riboflavin,¹ and that this charge transfer is a strong one, involving the formation of a primarily ionic complex and leading, under suitable conditions, to the formation of free radicals.² The orbital energies of indole, calculated by the Pullmans,^{3,4} do not predict such strong electron donor properties. They show indole to be only a fair electron donor. That this is the case was shown experimentally by E. Fujimori,[†] who measured the charge transfer spectrum of indole with various classical acceptors, capable of π - π interaction, like trinitrobenzene, benzoquinone, chloranil, and bromanil.

It seemed desirable to find the cause of this discrepancy since various biologically important molecules, like serotonin, tryptophan, and growth hormones, are indole derivatives.

The electronic structure of indole, calculated by the Pullmans,³ shows a high formal negative charge on carbon atom 3, and a smaller one on carbon 2. It seemed thus possible that the strong electron donor properties of indole were due to the high electron density on one or both of these C atoms. The π electrons of

these carbon atoms belong to the π electron pool of the highly conjugated molecule. So, it seemed possible that the high electron density makes these individual C atoms into good "local" donors, which could draw on the π electron pool of the whole molecule for the donated electron. As the work of Fujimori showed, this localized electron-donating property does not declare itself in a π - π interaction. However, it might declare itself in an interaction with a small, or "local," electron acceptor. Such an interaction would not demand the rigorous steric requirements on a donor-acceptor complex that a π - π interaction of two aromatic structures does. For this reason we have tested the interaction of indoles with iodine, the I_2 molecule being a relatively small molecule and a fair electron acceptor.

Charge transfer will occur only if there is overlap between the donor and the acceptor orbitals. At room temperature, in solution, thermal agitation may keep the two molecules apart. In order to study charge transfer, it may be necessary to force the two together, which can be done in various ways. One can, for instance, dissolve both substances in water and freeze the solution, whereby the solvent crystallizes out leaving the solutes in close proximity. In case of nonpolar substances, one can dissolve these separately in a volatile solvent like chloroform, then moisten a filter paper with the solution of the one, say the donor. After the solvent has evaporated, one can moisten the paper with the solution of the acceptor. After evaporation of the solvent, the two solutes will be left in close proximity. One can also evaporate a solution of the mixture of both components. In many cases it may suffice to lower the temperature of the mixture of the two substances in order to induce the desired complex formation. Another method consists of allowing a solution of the donor in question to dry on filter paper, then exposing the paper to the action of I_2 vapor.⁵

An 0.1 *M* solution of I_2 in chloroform is purple. If this solution is mixed with an 0.1 *M* solution of indole, the solution remains purple showing that little or no charge transfer complex is formed. However, if the solution is placed on filter paper and the solvent allowed to evaporate, a black spot is left behind. Similarly, if the solution is cooled in a dry-ice cooling mixture a voluminous black precipitate is formed which dissolves again on warming. The precipitate may be separated by centrifugation at -20°C . When the precipitate is pressed out between sheets of filter paper and quickly air dried, a black powder is obtained. Such a powder was placed into the cavity of an electron spin resonance apparatus and a high and narrow signal was obtained, showing 10^{19} spins per gram.[†] The signal showed no hyperfine structure and had a *g* value of 2.001, close to that of a free electron. This indicates that a strong charge transfer has occurred from indole to iodine. A similar black precipitate was obtained with skatole, indicating that the nitrogen atom was not involved in the reaction. Indole acetic acid and serotonin gave similar reactions. In these molecules the side chain does not appreciably alter the charge of the C atom at position 3 and 2. However, when the high negative formal charges of C2 and C3 were abolished by making these part of an additional conjugation, as is the case in carbazole, no black complex was formed, although the NH remained free. This supports the assumption that the C atoms 3, or 2, (or both), were involved in the formation of the charge transfer complex. Free pyrrole has a pronounced negative charge on both these C atoms and gives a similar reaction as in indole.[§]

These reactions deserve attention because it seems possible that the described high electron-donating activity in indole plays a role in the reactions of proteins, since the C3 atom is conjugated by a hyperconjugated CH_2 to the peptide link, which may form through the H-bridges a continuous electronic system with the other peptide links of the same molecule. It may play a role also in the biological activity of indoleacetic acid and serotonin.

The black indole- I_2 complex is, in all probability, in many ways analogous to the black iodine complexes formed by molecules with extensive conjugate systems, like perylene or violanthrene, which also give strong electron spin resonance signals.^{6, 7} This suggests that while extensive conjugated systems may tend to donate electrons to a local acceptor in any case, smaller and simpler molecules with conjugated systems can do so only if they contain highly charged carbon atoms acting as "local donors." Though "local" the donating atom may draw on the π pool of the molecule for the electrons to be donated.

Note added in proof: An elementary analysis of the indole-iodine complex has been made by the Schwarzkopf Microanalytical Laboratory and showed 27 N atoms for 23 I_2 molecules suggesting that the complex contained one I_2 per indole.

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† Fujimori, E., unpublished work, see quotation 4, page 63.

‡ The substance probably contained some moisture, due to the condensation of water vapor at the low temperature employed.

§ Due to the volatility of pyrrole, it is advisable to allow an 0.1 M solution in chloroform, mixed with equal or double volumes of 0.1 M I_2 in chloroform, to dry on filter paper instead of applying the two substances one by one.

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THE FUNCTION OF RNA IN T₂-INFECTED BACTERIA

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When *Escherichia coli* B are infected with virulent bacteriophages, no net synthesis of RNA can be demonstrated.¹⁻⁶ Nevertheless, an active metabolism of RNA has been shown to exist.⁶⁻⁸ Employing either P³²- or C¹⁴-labeled nucleic acid precursors, we determined^{6, 8, 9} that some of the host RNA is concomitantly synthesized and degraded (or utilized). This metabolic turnover of RNA is most extensive in the early stages after infection, exceeding the incorporation rate of isotope into DNA the first few minutes after infection.¹⁰ Unlike the RNA in