blood chemistry has been validated in this laboratory by comparison with blood obtained by arterial puncture in both normal and polycythemic (induced by red cell transfusion) men at various degrees of oxygen saturation.

CONCLUSION

The direct transfusion of erythrocytes into normal young men appears to be a safe procedure and when the volumes transfused are sufficiently great, the resultant polycythemia appears to afford increased tolerance to hypoxia. The polycythemia persists approximately six weeks.

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CYTOPLASMIC DISEASES AND CANCER

RECENT articles by Dixon,[1] Potter[2] and others prompt us to review various data obtained in certain fields and to interpret these in the light of a theory we have proposed that links three groups of diseases: namely, virus diseases, variegational diseases and cancer. The theory is based on the similarity of plants and animals at cellular and subcellular levels. Realization of this similarity has repeatedly shown the way to new approaches in research. We only have to mention a few cases: (a) the laws of heredity, proven for plants and applied to animals; (b) studies on enzyme and respiration processes in yeast and organs of higher plants, often elucidating similar processes in animals; (c) studies on plant viruses, indicating the way for isolation and purification of animal viruses; (d) studies on lower plant and animal forms, demonstrating the similarity of these forms on a cellular level.

That the ultimate agent of cancer may reside in the cytoplasm was suggested by Warren Lewis,[3] among others, in 1936. However, he did not specify what component of the cytoplasm might be involved. In recent years there has been an increasing tendency[2,3,4] to consider many cancers as cytoplasmic diseases and to regard the nuclear abnormalities associated with them of a secondary nature. Without doubt certain diseases of the tumor type are induced by viruses: *e.g.*, the Shope papilloma and Rous sarcoma. Oberling[4] has suggested that all cancers may ultimately be shown to be virus diseases. However, in the great majority of cases it has not been possible to demonstrate a causal virus. Moreover, the induction of cancer by specific carcinogenic agents such as methylcholanthrene, x-rays, etc., can be explained on the basis of the pure virus theory only when a series of rather involved assumptions are made. Granting that some cancers may be cytoplasmic diseases, that viruses are for the most part typically associated with the cytoplasm and that at least some cancers are virus-induced, what evidence do we have for a theory which reconciles both the virus and the so-called non-virus theories of cancers?

There have been numerous theoretical discussions on this subject, but in practically all instances definite experimental or observational evidence has been lacking. After Bensley and Hoerr[5] in 1934 separated mitochondria from liver tissue, Claude[6] in 1940 isolated from tumor tissue macromolecular complexes containing ribose nucleoprotein. He subsequently suggested that these fractions might be identical with mitochondria and that they might have an etiologic significance. However, considering the rate of cell division of neoplastic tissues, the mere presence of mitochondria in such tissues is not of itself significant, because mitochondria are always abundant in dividing tissue. Claude could not show that the fraction differed qualitatively from a similar fraction obtained from normal tissues nor did he point to any analogous case in which plant or animal mitochondria function as pathogenic entities.

As a result of studies on the interrelations of plastid chromoprotein, tobacco mosaic protein and cell metabolism, Woods and duBuy suggested in 1941[7] that a fundamental relationship might exist between mitochondria (or plastids, which are specialized mitochondria *sensu* Guilliermond) and virus proteins. Later, in 1943,[8,9] they were able to show that plant variegations are diseases often of virus-like nature, which are caused by abnormal cytoplasmic particulates of hereditary character: the plastids or mitochondria. That plastids can be modified by a nuclear factor, and that this modification is transmitted thereafter by cytoplasmic heredity, was shown in maize by Rhoades in 1943.[10] Other cases of plastid inheritance were reported by Anderson, Demerec, Imai, Renner and others.[11] Woods and duBuy were able to establish a "spectrum of variegation" ranging from

[1] T. F. Dixon, *Nature*, 155: 596, 1945.
[2] Van R. Potter, SCIENCE, 101: 609, 1945.
[3] Warren H. Lewis, SCIENCE, 81: 545, 1935.
[4] Ch. Oberling, "The Riddle of Cancer," Yale University Press, 1944.
[5] R. R. Bensley and N. L. Hoerr, *Anat. Rec.*, 60: 251, 1934.
[6] A. Claude, SCIENCE, 91: 77, 1940.
[7] M. W. Woods and H. G. duBuy, *Phytopath.*, 31: 978, 1941; *id.*, 32: 288, 1942.
[8] M. W. Woods and H. G. duBuy, *Phytopath.*, 33: 637, 1943.
[9] H. G. duBuy and M. W. Woods, *Phytopath.*, 33: 766, 1943.
[10] M. M. Rhoades, *Proc. Nat. Acad. Sci.*, 29: 327, 1943.
[11] E. G. Anderson, *Bot. Gaz.*, 76: 411, 1923; M. Demerec, *Bot. Gaz.*, 84: 139, 1927; Y. Imai, *Genetics*, 13: 544, 1928; O. Renner, *Flora*, 30: 218, 1936.

the normal plastids through mutated mitochondria to typical viruses. These mutated mitochondria, of course, persist only when they are to a certain extent compatible with the genic constitution of the cell. Since Guilliermond and others[12] have shown that plastids are specialized mitochondria, the variegational plant diseases can be considered as mitochondrial diseases.

The authors further described numerous instances in which the presence of abnormal (mutated) plastids causes profound changes in cell metabolism; *e.g.*, disturbances in oxidizing enzyme systems, cell differentiation and growth, functions of the normal plastidome, etc. Thus factual evidence has been supplied for the validity of the suggestion made by A. F. Woods over forty years ago[13] that certain "non-infectious" variegations and certain viroses of the mosaic type are fundamentally the same. In the most extreme cases of cell disturbance these changes were so great that the cells remained more or less isodiametric or undifferentiated in form, thus paralleling some of the conditions found in cancer. More recently we have been able to demonstrate the influence of temperature on the relative dominance of either the normal or the pathogenic chondriosomal systems when both exist in the same cell.[14] From these observations it has been possible to offer by analogy definite suggestions as to what may be the underlying phenomena governing the latent period, initiation, development or regression of cancers, namely changes in chondriosomal equilibria.

We could demonstrate the presence of a ribose nucleoprotein in virus-free plastids, thus confirming the earlier report by Menke[15] of the presence of this type of protein in the plastid. Attention was called to the apparent relation between the ribose nucleoprotein components of the normal and the variegation-inducing plastids or mitochondria, and the ribose nucleoprotein constituting the pathogenic viruses. Considering the self-perpetuating nature of the abnormalities it was suggested that the mutational change in the plastids probably occurred in the ribose nucleoprotein component of the disease-causing entities, the chondriogene.

Since it has long been recognized, though sometimes forgotten, that plant and animal mitochondria are fundamentally alike, animal mitochondrial diseases might be expected to occur. Although the exact functions of animal mitochondria are not fully understood, there is increasing evidence that the mitochondria play an important role in controlling cell metabolism.[16] Hence, mitochondrial diseases in animals should be characterized by a disturbed cell metabolism in which the disturbing agent is self-perpetuating in the cytoplasm. Mitochondria, changed by a process of "retrograde" evolution, could be considered as the underlying cause not only of the plastid-controlled variegational diseases and ultimately of the viroses, but also, through a similar course of evolution, as the underlying causes of many cancers. As far as we are aware, the first experimental data to be presented pointing to such a fundamental relationship among these three types of diseases are given in our earlier papers.

On the basis of the data presented in these papers we can consider the viroses as infectious diseases caused by entities evolved from one-time normal cell components, namely, the ribose nucleoprotein components of the plastids or mitochondria. In the case of the variegational diseases, we deal with recently produced pathogenic cytoplasmic particulates, the variegation-inducing plastids or chondriosomes. Is it not highly plausible that similar conditions exist in animals? Thus we might expect to find cancer-inducing viruses at one end of a "cancer spectrum" and at the other localized cancer-inducing mitochondria, which should be demonstrable serologically and probably microchemically, corresponding to the spectrum of variegation-inducing plastids (mitochondria) of plants. In this connection it should be stressed that this theory does not hold that all agents now classed as viruses in animals are necessarily of mitochondrial origin. Agents such as those causing psittacosis, vaccinia, etc., may be more closely allied to known organisms such as the agents of pleuropneumonia, agalactia, etc., than to the true viruses. The detailed reasons for the acceptance of a biphyletic origin of the macroviruses on the one hand and of the true viruses on the other can not be elaborated here. The theory does not require that all the viruses which attack a given species, man for example, have necessarily arisen from chondriosomal proteins of the same species. In fact, the wide host range of some animal viruses suggests that chondriosomal nucleoproteins of many phyla may be involved.

Since our original publication of the foregoing concept relating viruses, pathogenic mitochondria (plastids) and hypothetical cancer-inducing mitochondria, Haddow[17] and Darlington[18] in England, and more recently Potter[2] in America, have broached somewhat similar concepts in theoretical reviews. In

[12] A. Guilliermond, G. Mangenot et L. Plantefol, *Traité de cytologie végétale*, Le Francois, Paris, 1933; A. Guilliermond, "The Cytoplasm of the Plant Cell," Chron. Bot. Co., Waltham, Mass., 1941.

[13] A. F. Woods, *Centralbl. f. Bakt. u. Par. Krankh.*, 5(II): 745, 1899.

[14] M. W. Woods and H. G. duBuy, *Phytopath.*, in press.

[15] W. Menke, *Ztschr. f. Physiol. Chem.*, 257: 43, 1938.

[16] A. Claude, *A.A.A.S. Research Conference on Cancer*, 4: 232, 1945.

[17] A. Haddow, *Nature*, 154: 194, 1944.

[18] C. D. Darlington, *Nature*, 154: 164, 1944; *id.*, 154: 489, 1944.

interpreting the paper of Darlington in particular, it is important to recognize that (a) there is an extensive experimental basis for the acceptance of a nuclear hereditary system; (b) there is increasing evidence for the existence of an extra-nuclear, mitochondrial hereditary system; (c) there is *no* evidence for a diffuse cytoplasmic hereditary system (Sonneborn's data[19] quoted by Darlington as evidence can be differently interpreted on the basis of experiments supplied by Marshak and Walker[20]); also, mitochondrial exchange has not been excluded in Sonneborn's experiments. Darlington does not even mention mitochondria, but only plastids of green plants); and (d) the development of simple non-hereditary cytoplasmic proteins into hereditary systems is entirely without experimental proof. A casual student might find illuminating and thought-provoking the postulate that a non-hereditary protein gradually develops into an hereditary protein, and this, via a particulate hereditary system, into a nuclear hereditary system. Such a view implies that any change in any part of the cell might be the cause of cancer and is, therefore, of little help to the person who has to make a choice among the many experimental approaches to the cancer problem.

It is possible that hereditary particulates of the cytoplasm, other than the mitochondria or plastids, may exist and may develop into pathogenic agents, but the important issue is that we do not have adequate evidence for their existence. On the other hand, we do have proof of the occurrence in nature of known cytoplasmic particulates that can become pathogenic and that can be connected with viruses by gradual steps in a "spectrum of variegation."

In the plastid-induced variegations of plants we have access to concrete facts which may well be applied to the solution of the cancer problem. We have at least been able to set up a theory which reconciles both the virus and so-called non-virus theories of cancer. In addition, it offers an explanation for such phenomena as latency, fluctuations in activity of tumor cells, etc. Also, the precise cellular structures which are changed at the induction of a cancerous condition are indicated. The data obtained from studies on cytoplasmic particulates of plants can offer leads for studies in the much more difficult field of animal cytoplasmic diseases.[21] They will help the workers in the field of cancer to make a choice among the great number of scientific approaches possible in their complex field.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

AN ANTIBIOTIC FROM A BEE PATHOGEN

THE fact that honyebee larvae dead of American foulbrood (scales) almost invariably contain pure cultures of *Bacillus larvae* suggested that this organism might produce an antibiotic. When such scales were placed on the surface of freshly seeded nutrient agar plates of a soil suspension, and of market milk, definite inhibition zones were noted. Since antibiosis against a wide bacterial flora was thus shown, qualitative studies were made employing cultures of Gram-positive, Gram-negative and acid-fast bacteria.

Inhibition zones were produced around scales placed on plates seeded with the following cultures: *Staphylococcus aureus, Staphylococcus albus, Streptococcus agalactiae, Escherichia coli, Aerobacter aerogenes, Brucella abortus, Brucella melitensis, Bacillus subtilis, Bacillus alvei, Mycobacterium tuberculosis* var. *hominis, Mycobacterium tuberculosis* var. *bovis.* No inhibition occurred with *Mycobacterium avium* or an unidentified acid-fast bacterium from a skin lesion. Secondary or "clearing" zones appeared progressively around the original inhibition zones with *B. subtilis* and *E. coli,* but not with *Staph. aureus.* Such zones produced no growth on subculture, even though, with *B. subtilis,* numerous spores were present.

The size of the inhibition zone around scales on agar plates seems to be a function of the diffusion properties of the antibiotic rather than a measure of its concentration or activity against a particular organism. Thus, approximately equal zones are produced around *E. coli* and *B. subtilis;* yet an aqueous scale extract which inhibited *B. subtilis* in nutrient broth at a dilution of 1:20,000 inhibited *Staph. aureus* at only 1:2,000. The cup assay method has not proved feasible.

What appears to be the identical antibiotic is produced in culture by *B. larvae,* but only, as has been shown for certain enzymes,[1] when sporulation occurs. Sporulation has not been attained in broth culture.

A peculiarity was noted when *B. larvae* was grown on serum-glucose-potato extract agar in the presence

[19] T. Sonneborn, *Proc. Nat. Acad. Sci.,* 29: 329, 1943; *id.,* 29: 338, 1943.
[20] A. Marshak and A. C. Walker, *Amer. Jour. Physiol.,* 143: 235, 1945.
[21] H. G. duBuy and M. W. Woods, *A.A.A.S. Research Conference on Cancer,* 4: 162, 1945.
[1] E. C. Holst and A. P. Sturtevant, *Jour. Bact.,* 40: 723–731, 1940.