two viruses have been found so frequently in close association and appear to have the same vectors and vertebrate hosts. There is some further support for the latter possibility in the following results of two experiments. (1) Laboratory mixtures of known strains of these two encephalitis viruses, although not interfering completely in mice, are maintained with great difficulty through two passages. The Western equine virus is greatly inhibited, and attempts to recover it beyond the third passage have all failed. (2) Guinea pigs are killed promptly with signs typical of the Western encephalitis by a suspension of the brains of mice which die after an inoculation with 100 LD_{50} (third mouse passage) of the mite virus mixed and incubated with an equal volume of undiluted hyperimmune Western equine serum. The same suspension of mouse brain which killed normal guinea pigs failed to kill Western equine immune pigs. Thus, a high dilution of a third mouse passage of the mite agent, mixed with a great theoretical excess of Western equine immune serum, still maintained through passage a large amount of the equine-like component.

Until more convincing evidence can be presented that this agent represents a "new" virus, however, it appears to be more appropriate merely to report that a virus, or mixture of viruses, has been isolated from wild bird mites which, after serial passage in the brains of mice, may be identified as a strain of St. Louis encephalitis virus, and that a factor identifiable as the virus of Western equine encephalomyelitis is obtained after the agent is passed serially through chick embryos.

Reference

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Periconia circinata, the Cause of Milo Disease

R. W. Leukel and A. G. Johnson

Division of Cereal Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering, U. S. Department of Agriculture, Beltsville, Maryland

The milo disease, which attacks the roots of susceptible milos and dwarfs and kills the plants, was first observed in Texas in 1924, and within a few years was found to occur also in other sorghum-growing states of the Southwest. For a time this disease threatened the growing of milo in certain areas of the Southwest. Resistant milos were developed, however, and now the severe losses formerly caused by the disease have been almost entirely eliminated.

For many years the exact cause of milo disease remained unsolved. The fact that susceptible milos grown in infested soil that had been sterilized by steam or chemicals remained free from the disease indicated that it was caused by a soil-borne organism. In 1937, Elliott, *et al.* (*1*) reported that the fungus *Pythium arrhenomanes* had been repeatedly isolated from the roots of susceptible milo plants grown in infested soil and affected with milo disease, and that this fungus was thought to be the causal organism. It had been previously found to be the cause of a common root rot of corn and sugarcane. It proved to be the cause also of a certain type of root rot in sorghum (*1*). It has never been demonstrated, however, that milo selections resistant to milo disease are resistant also to attack by *P. arrhenomanes*. The only logical conclusion, therefore, is that *P. arrhenomanes*, although able to attack the roots of sorghum, is not the fungus that causes milo disease.

Experiments have been in progress since 1942 at the Plant Industry Station, Beltsville, Maryland, to determine definitely the real cause of milo disease. Numerous isolations of fungi were made from the roots of susceptible milo plants grown in soil from infested fields and affected with typical milo disease. These fungi were grown on suitable media, and the cultures were used to inoculate separate portions of steam-sterilized soil. Selections of milo, resistant or susceptible to milo disease, were grown in these different lots of inoculated soil, and the growth of the plants was compared with the growth of these same selections in naturally infested soil and in steam-sterilized uninoculated soil. Some of these fungi reduced the percentage of emergence, caused damping-off, or produced other disease symptoms, but always with equal severity in selections resistant or susceptible to the milo disease. All isolates, except one, failed to produce results identical with those obtained in naturally infested soil. This one exception was a fungus that produced a mouse-gray mycelial growth on potato-dextrose agar. This growth later turned darker, due to the production of great numbers of large, black spores. The fungus was identified as *Periconia circinata* (Mang.) Sacc. When selections of milo susceptible or resistant to the milo disease were grown in the greenhouse in flats and beds of steam-sterilized soil inoculated with this fungus, the susceptible milo plants displayed symptoms identical with those shown by these same selections grown in naturally infested soil, while the resistant milos were not affected. Susceptible plants grown in lightly inoculated soil showed symptoms of milo disease similar to those seen in plants growing in lightly infested fields. Plants grown in heavily inoculated soil produced more severe symptoms similar to those seen in plants growing in infested fields that had been planted to milo for several years. Resistant and susceptible selections of each of 8 milo varieties yielded identical results when grown in flats of *Periconia*-inoculated soil and in flats of naturally infested soil. In each case the susceptible selections were killed, while the resistant selections were not affected. Typical milo disease was produced also in susceptible milo grown in an outdoor bed of steamed soil inoculated with *P. circinata*, while plants of resistant milo were not affected. Identical results were obtained in a similar adjacent bed containing naturally infested soil. In all these experiments in which susceptible milo was affected with typical milo disease when

grown in *Periconia*-inoculated soil or in naturally infested soil, the characteristic fungus, *P. circinata*, was readily recovered from the diseased roots.

In testing selections of milo for resistance to milo disease in the past, plant breeders were restricted in their studies to the use of soil naturally infested with the milo-disease pathogen. Such soils at times are rather heavily infested also with various other harmful fungi, whose effects on the plants under observation may be confusing. Now, however, with the use of steam-sterilized soil inoculated with the causal fungus, *Periconia circinata*, only, the resistance to milo disease will be more clearly indicated as effects of other fungi are eliminated. More complete details of these investigations will be published later.

Reference

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Nature and Spatial Relationship of the Prosthetic Chemical Groups Required for Maximal Muscarinic Action

Carl C. Pfeiffer

Department of Pharmacology,
University of Illinois College of Medicine

Dale, in his Dixon Memorial Lecture (*5*), states: "To the fundamental pharmacological problems, why a particular type of chemical structure, or more mysteriously, several apparently unrelated types, should be associated with a specific action on particular types of reactive cells, we have made no nearer approach." With this in mind we have attempted to correlate pharmacological action with certain prosthetic groups[1] which appear as a common denominator in all potent parasympathomimetic drugs. Attempts to correlate structure-activity-relationships (SAR) of chemical series of pharmacological importance have hitherto mainly considered activity as variations in chain length of aliphatic series as drawn in two dimensions. Occasionally theories have been centered around the well-known ring systems of organic chemistry. Greater correlation and understanding of SAR might be obtained by depicting formulas in three dimensions, with bond distances calculated as accurately as our present knowledge will allow. Schueler (*16*) has applied this approach successfully in relating estrogenic activity to chemical constitution. Clark (*1, 2*), from his studies and those of other workers, has amassed almost incontrovertible evidence that pharmacological action in many instances depends on the activation of receptors on the cellular surface by specific chemical prosthetic groups on the drug molecule. A cellular surface action as a possible mode of action of the muscarinic drugs is assumed in this study.

From inspection of the numerous drugs having parasympathomimetic stimulant action it becomes apparent that all contain a ketone oxygen group adjacent to an ether oxygen linkage with a methyl substituted nitrogen at a distance of two saturated carbon atoms. Assuming that these molecules have three prosthetic groups, the interprosthetic distances may be calculated for many of the known potent stimulants. The size of the acetylcholine molecule (approximately 9 A × 3 A), when compared to the size of the cell surface (20 × 1 μ for vascular smooth muscle), is infinitely small. With atomic models where 1 A equals 1 cm, the smooth muscle cell, if drawn to the same scale, would measure 2,000 × 100 m, or roughly 1 mile long and a city block in diameter. Thus, the surface of the cell containing receptors may be considered as the segment of an arc with an infinite radius, and the interprosthetic distances, for practical purposes, may be

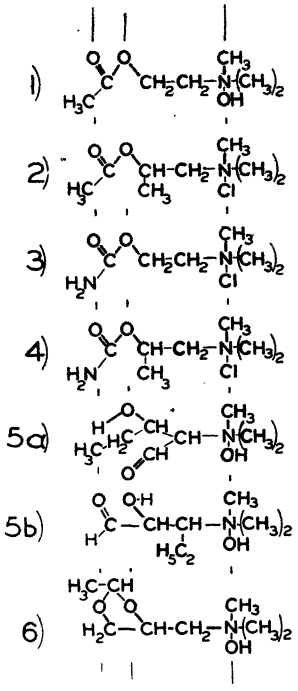

Fig. 1. Aliphatic parasympathetic stimulant drugs: (1) acetylcholine, (2) methacholine (Mecholyl), (3) carbaminoylcholine (Doryl), (4) urecholine, (5a) textbook formula for muscarine, (5b) Kögl's alternate formula for muscarine, (6) Bovet and Fourneau's acetal derivative.

regarded as linear distances and may be measured on Hirschfelder atomic models (*13*). The measured interprosthetic distances in acetylcholine are 7.0 A ketone oxygen to methyl, and 5.3 A ether oxygen to methyl (Fig. 1). Obviously, the interprosthetic distances for acetylcholine, methacholine, carbaminoylcholine, and urecholine are the same. Choline, which contains only two of the prosthetic group, has a parasympathomimetic stimulant action which is insignificant compared to that of acetylcholine. Compound 6 of Fig. 1, which has an acetal group, was

[1] The term "prosthetic group" as used in this study refers to simpler chemical complexes such as methyl groups or oxygen atoms, whereas enzymologists utilize this term for larger chemical complexes.