tidis, and *Sporotrichum schenckii*) incubated on the horse serum agar (pH 7.0) at 37° C also were inhibited by 1.0-mg/ml concentrations of di-phenyl-pyraline and pyribenzamine. The growth of *C. albicans* was not affected by either drug.

Results obtained from these preliminary studies suggest that both di-phenyl-pyraline and pyribenzamine possess properties which are inhibitory to pathogenic fungus species and that their activities, therefore, are not limited to the alleviation of allergic manifestations incited by these organisms. The results also suggest that further clinical study is imperative to determine the effectiveness of these drugs *in vivo*.

References

1. CAMPBELL, C. C. and SASLAW, S. *Proc. Soc. exp. Biol. Med.,* 1949, **70**, 562.
2. HILLEGAS, A. B. and CAMP, E. *J. Invest. Derm.,* 1945, **6**, 217.
3. OSTER, K. A. and GOLDEN, M. J. *Amer. J. Pharm.,* 1949, **121**, 375.
4. PATIALA, R. *Ann. Med. exp. Biol. fenn.,* 1949, **27**, 145.

Triphenyltetrazolium Chloride in Tissue Culture[1]

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A number of reports have recently appeared in the literature describing a tetrazolium salt (2,3,5-triphenyltetrazolium chloride) as a possible indicator of viability of plant and animal tissues. Lakon (*3*), Cottrell (*1*), and Porter, Durell, and Romm (*4*) have used this compound to test the viability of seeds and wood cuttings, and Straus, Cheronis, and Straus (*6*) employed this salt to demonstrate reducing enzyme systems in living normal and neoplastic mammalian tissues.

In studies of the nucleic acids and of the effect of folic acid and its analogues on tissue cultures, it is occasionally important to determine cytologically the number of living cells present in a particular culture. Since the cytoplasm and nuclei of living cells do not stain with any of the vital dyes, and dead cells stain only diffusely, the use of tetrazolium as an indicator of cellular viability suggested itself. It is the purpose of this paper to report the failure of 2,3,5-triphenyltetrazolium chloride as an indicator of cell viability of tissue grown *in vitro*.

The method employed in testing this compound consisted of growing chick-heart fibroblasts under perforated cellophane (*2*) in a medium composed of equal parts of human fetal (umbilical cord) serum and Simms salt-diluted ultrafiltrate (*5*). After 72 hr, when sufficient migration and outgrowth of fibroblasts had occurred, the cultures were placed in a 1% solution of the tetrazolium salt in 0.9% NaCl. They were then examined grossly and microscopically, every 20 min for 6 hr, for evidence of reduction of the colorless tetrazolium to the red insoluble formazan. In none of the 16 cultures tested was there any observed reduction of the salt. Cultures left in this solution for 48 hr also gave negative results.

The failure of whole, uninjured, rapidly growing embryonal cells to reduce tetrazolium chloride indicates that this compound is not necessarily a measure of cellular viability. The action of tetrazolium chloride depends on combination with dehydrogenases which cause a reduction of the dye to its colored form. Apparently the failure of the compound to penetrate through the living cellular membranes is responsible for the negative results obtained.

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References

1. COTTRELL, H. J. *Nature,* Lond. 1947, **159**, 748.
2. EVANS, V. J. and EARLE, W. R. *J. nat. Cancer Inst.,* 1947, **8**, 103.
3. LAKON, G. *Ber. Dtsch. Bot. Ges.,* 1942, **60**, 299.
4. PORTER, R. H., DURELL, M., and ROMM, H. J. *Plant Physiol.,* 1947, **22**, 149.
5. SIMMS, H. S. and STILLMAN, N. P. *Arch. Path.,* 1937, **23**, 316.
6. STRAUS, F. H., CHERONIS, N. D., and STRAUS, E. *Science,* 1948, **108**, 113.

A Modification of the Hardy-Weinberg Law

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The Hardy-Weinberg law, as it is customarily specified or understood, applies to populations which are indefinitely large, which are panmictic, and in which gene frequencies are the same in both sexes. If, in addition to these conditions, gene frequencies are constant, genotype frequencies will also be constant through succeeding generations (*2–6*).

Whether gene frequencies are constant or not, the primary distribution (*primary* being used in the same sense as applied to sex ratio) of mean genotype frequencies for two alleles A and a, with respective frequencies p and q ($q = 1 - p$), is written $p^2AA:2pqAa:q^2aa$ when A and a are autosomal and ♀♀ ($p^2AA:2pqAa:q^2aa$) + ♂♂ ($pAY:qaY$) when they are sex-linked. But the frequency of one of the two alleles, say q, may be very small, and when it is sufficiently small it is evident that generations will occur in which homozygous recessives will not be produced; regardless of the smallness of q, the term q^2 then represents the probability of occurrence of genotype aa, but not its frequency, which of course is 0. Under this condition the genotypic distribution for a pair of autosomal genes A, a becomes $(p-q)AA:2qAa$, which provisionally may be called a limiting distribution. This can readily be seen, since under panmixis the proportions of pA and qa that recombine will be equal and will