Comments on Photosensitivity in Leprosy

To THE EDITOR:

Lalit Bhutani in the last issue of the IJL (45:67) has noted the relative sparing of the underside of the chin and the circumocular region and the upper lip in lepromatous patients and the more severe involvement of the malar eminence, the forehead, and the chin. He attributes this variation to the possible effect of photosensitivity. Sabin (Arch. Neurol. 20 [March 1969]; and IJL 37 [1969]) describes the same pattern of involvement, measured by loss of sensation and showed that both on the face and also in the areas of the body that are ordinarily clothed the degree of sensory loss followed the pattern of local body temperature variations, the cooler areas being severely involved and the warmer areas being relatively spared, thus confirming the observations of Brand (IJL 27 [1959] 1-7).

I enclose a thermograph (cool areas dark, warm areas light) of a normal face (my own) which clearly shows the cool chin, nose, and malar area and the warmer underside of the chin, orbit and lips. The eyebrows are black because hair always shows black in a thermogram.

- Paul W. Brand, FRCS
Chief, Rehabilitation Branch
USPHS Hospital
Carville, Louisiana 70721

Comparative DNA Binding Studies with Clofazimine and B1912

To THE EDITOR:

B1912 is a phenazine quinonemine that was synthesized by Barry and co-workers (1) and subsequently recommended as an analog of clofazimine (B663) for possible screening of antileprosy activity in man (11-13). The compound was active against Mycobacterium leprae in the mouse foot pad assay (14) as well as inhibiting the growth of M. lepraemurium (15). Like clofazimine, B1912 will accumulate in the tissues of rodents (2-3) particularly in the skin, when added in powder form to the diet. It has been found in this laboratory that at a concentration of 0.05% by weight in the diet, rapid skin pigmentation will occur in the mouse in a three to four day period.

Following the observation that clofazimine will form complexes with DNA from various sources, including human (+*), it was of interest to compare the relative interaction of clofazimine versus B1912 with this important macromolecule. The degree of interaction was measured with a spectrophotometric assay (16) of the amount of upfield red shift that took place in the spectrum of clofazimine or B1912 following complexing to DNA. The upfield red shift resulted from the interaction of the heterocyclic phenazine quinonoidal ring with the nucleotide bases of the DNA strand. The assays were carried out in 0.01 M tris-HCl buffer, pH 7.0, at 22°C with DMSO added at 10% (v/v) to maintain the compounds in solution. Clofazimine or B1912 was added at a final concentration 2 × 10⁻⁶ M and dialyzed DNA was present at 250 µg per ml. The upfield red shifts were measured in a Cary 14 recording spectrophotometer (16).

It was found that, like clofazimine, B1912 interacted with DNA's from various sources with upfield red shifts and hypochromic spectral changes in the absorption peak at
482 nm. The degree of interaction was dependent upon the G + C content of the DNA strand. The upfield red shifts were significantly greater than those found with clofazimine. The extent of the increased interaction was calculated from the ratio B1912/B663 as measured from the upfield peak-to-peak shift in nanometers. For human DNA the B1912/B663 ratio was 4, for bovine DNA the B1912/B663 ratio was 5, and for M. leprae RNA the B1912/B663 ratio was 5.7. When the DNA was replaced by the synthetic polyribonucleotide strand, poly G, it was found that B1912 interacted to produce an upfield red shift of 50 nm and a calculated B1912/B663 ratio of 4.1. These data indicate that B1912 undergoes a 4- to 5.7-fold greater degree of spectrally identified interaction with DNA than does clofazimine.

A structural basis to this increased interaction has been found from the fact that B669, a derivative of B1912 in which the 7-position chloro substituent has been removed (2'), interacted with DNA to produce upfield red shifts that were comparable to those found with clofazimine. Evidently the 7-chloro substituent on the phenazine ring has an activating or enhancing effect on the binding of B1912 to DNA. By contrast when the chloro groups were located on phenyl and anilino substituent sites, such as in clofazimine, there was no significant enhancement of upfield red shifts. B1912 has also been found to interact with transfer RNA.

The interaction of B1912 with DNA would also account for the finding in this laboratory that B1912 has strong antimitotic effects in tissue culture. These effects were seen at 2 to 3 μg per ml concentrations in monolayer growth of human epithelial cells. Such concentrations represent levels that are attainable in human serum. These data thus have implications for the testing of B1912 in human volunteers.

—N. E. Morrison, Ph.D.
G. M. Marley, B.S.
Department of Pathobiology
School of Hygiene and Public Health
The Johns Hopkins University
Baltimore, Maryland, U.S.A.
21205

REFERENCES

Acknowledgments. Supported by BRS Grant FR-05445 from the Biomedical Research Support Branch, DFR, NIH. We thank R. S. McElhinney and M. L. Conalty for the supply of B1912.

Comments on Immuno-Epidemiology of Leprosy

To The Editor:

You kindly asked me to comment on your editorial "Immuno-Epidemiology of Leprosy" (JII, 43 [1975] 145-148), in relationship to the findings of our epidemiometric model (Lechat et al., Bull. WHO 51 361-373).

In fact, both your review of declining incidence trends in such countries as Norway and Hawaii, and the computer-simulated predictions we generated for South India, are approaches to the same end, using different methodologies. What the epidemiometric model tries to achieve is to predict the trends of incidence (that is new leprosy cases) under base line conditions over a long-term period on the basis of the number (prevalence) of infecting cases over the