neurotic tendencies (60 percentile and slightly below) had gained less than a 40 D.M.F. score, while those who were above these limits had, with but five exceptions, gained a D.M.F. score higher than this. The results obtained on the introversion-extroversion scoring showed that all but six of the subjects who were well within the normal limits had gained fewer than 40 points, and all but four above this limit had scored higher than 40.

The correlation coefficient for neurotic tendency percentile and D.M.F. points in men was + 0.446, and for introversion-extroversion and D.M.F. points the coefficient was + 0.405. These correlations are significant at the 5% level of confidence.

The coefficient of correlation between neurotic tendency percentile and D.M.F. points in women was + 0.463, and

TYPE OF SUBJECTS	NUMBER OF SUBJECTS	NEUROTIC TENDENCIES	INTROVERSION EXTROVERSION
MEN	25	7.8	5.3
WOMEN	25	11.9	11.9
MEN AND WOMEN	50	19.8	10.9

SIGNIFICANCE
$\chi^2 > 3.84$

FIG. 4. Chi square values.

the introversion-extroversion coefficient was + 0.447 which is also significant at the 5% level.

The combined correlation coefficients for the total 50 subjects were n_c + 0.474 and $i\text{-}e_c$ + 0.443. These are significant at the 1% level.

Another method of testing the realtionship just shown is the chi square test, which was applied with the following tabulated results.

Since all of these results are well above the required 3.84, the data obviously have statistical significance. The mean results indicate that there is less than 1% possibility that the results might be owing to chance distribution.

The Bernreuter Personality Inventory is not necessarily an exact measurement of personality, but is today the best and most valid test of its kind to measure traits of personality as divorced from intelligence. Furthermore, the important fact is not so much that specific personality traits are measured, but rather that a correlation between some traits and oral conditions exists. The D.M.F. scales which we are forced to use are also not the final answer to the measurement of caries incidence, but again are the most valid we have at our command. Added to this, we have a good indication that the value of the correlation coefficient will prove to be above + 0.40 if further studies are undertaken. This is surprisingly high if one realizes that the best correlations between medical disorders and psychic factors are rarely higher than + 0.50. It appears that the correlation between psychological factors and oral conditions merits further investigation with different types and larger numbers of subjects.

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A Preliminary Report on Histochemography[1, 2]

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During the summer of 1948, it was observed that normal rat bone marrow diluted with clear dog serum blackened an Eastman NTB photographic plate when smeared directly on the emulsion surface and stored at approximately − 15° C for several days. This observation was made on control experiments during an attempt to obtain single bone marrow cell autoradiographs.

Fig. 1 is a dark-field photomicrograph showing individual silver grains after development. The grains are

[1] A histochemograph is defined as a gross picture on a photographic plate or, at high magnification, a pattern of silver grains produced by the chemical action of a histological specimen in direct contact with the emulsion of the plate.

[2] This paper is based on work performed under contract with the U. S. Atomic Energy Commission at the University of Rochester Atomic Energy Project, Rochester, New York, and supported in part by the National Advisory Cancer Council of the U. S. Public Health Service.

grouped beneath cells[3] in some cases, but other groups are unassociated with cells. Examination of the gelatin of the emulsion indicated that the spots unassociated with

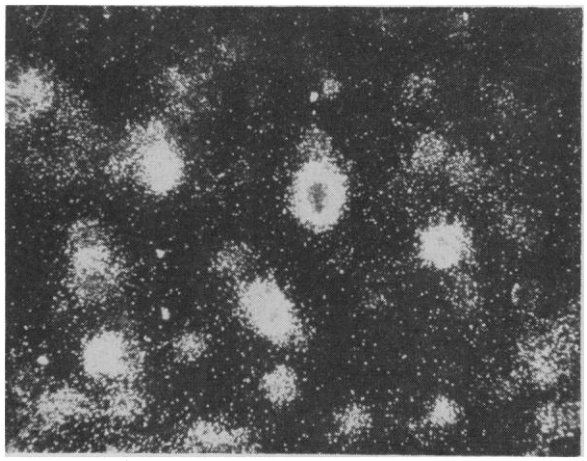

FIG. 1. A dark-field photomicrograph of a chemograph produced when rat bone marrow, diluted with dog serum, was smeared on an Eastman NTB photographic plate. Magnification approximately 666 ×. Some groups of silver grains are in the emulsion beneath a single cell and extending for several μ in all directions beyond the edge of the cell. This is illustrated by a bright spot (black spot in the original) with a dark spot in the center, the dark spot being the cell which has absorbed the scattered light from the grains beneath. Other bright spots show no cells present.

cells did not represent sites where cells had been washed away in the photographic fixing process, as was first suspected. The cause of the spots is unknown.

Peripheral blood from the same rat diluted with the same dog serum did not blacken the plate. The exposures continued for as long as 67 days. All plates developed serially showed that the bone marrow blackening increased in intensity with time while the peripheral blood smears showed no blackening, as can be seen from the background of the photographs published earlier in *Science* (*1*).

This emphasized the long-suspected likelihood of histochemical fogging and possible misinterpretation of autoradiographs from unfixed tissues. It was considered prudent to publish the results of preliminary experiments to bring this danger to the attention of those making autoradiographs.

A series of NTB plates were exposed to peripheral blood, spleen, kidney, liver, heart, and lung of a normal rat, sacrificed in the darkroom under a red light, Wratten Series I. A drop of blood was released from the jugular vein onto the emulsion and was not smeared. Slices of fresh organs were cut with a razor blade at 3–5-mm thickness and placed in direct contact with the emulsion. The tissues were on the emulsion at room temperature for 10–40 min, after which time they were stored at approximately −15° C to prevent further autolysis.

[3] Because of the difficulty in properly staining the cells after passage through photographic developer and fixative, various observers could not agree on the identification of the cells; but it seems probable that those showing the greatest blackening were the earlier forms of the erythrocyte series.

The plates were removed serially in time. After warming the plates to room temperature and removing the organ slices with soaking in water, we developed them in dilute Eastman Kodak D19 (1 part D19 to 3 parts water) for 25 min. Photographic densitometer measurements were made. In general, the density increased with time. It is interesting that the blood, placed as a drop on the emulsion, showed a small amount of blackening in about 2 weeks, whereas the blood of the first rat, in the summer of 1948, when diluted with dog serum and smeared on the emulsion, showed no blackening after 67 days.

Some of the exposures showed patterns which, it was felt, corresponded to the architecture of the section surface. Fig. 2, a chemograph of a kidney cross section, shows striations and other patterns, which may have been produced by tubules or blood vessels in the cortex, and shows a reticular pattern in the medulla, although not evident in the illustration. This reticular pattern is composed of discrete lines 4–8 μ wide, intersecting to form tiny unexposed areas of varying size. These were observed to be coincident in position with the interstitial connective tissue or basement membrane of the medullary epithelium. In the cross section of the liver, the lobule parenchyma was indicated by heavy blackening and portal areas by lighter blackening of the emulsion.

The experiment was repeated with two variations: (1) the age of the rat was approximately 250 days as contrasted with 90 days for the rat of the first experiment; and (2) immediately upon removal of the organ from the rat, it was placed on dry ice and remained frozen during sectioning, placing on the emulsion, and storage until removal for development. The same patterns of

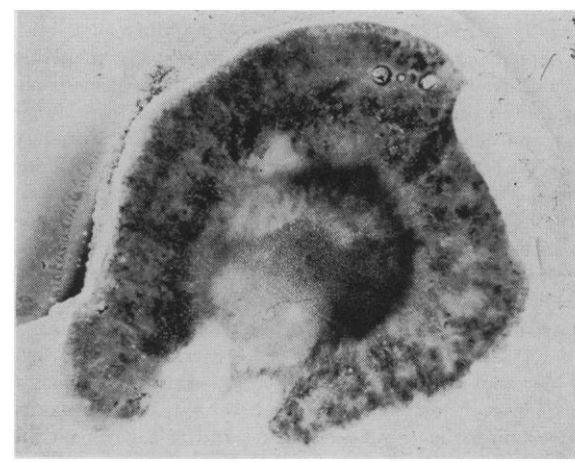

FIG. 2. A chemograph of a rat liver section placed in direct contact with an Eastman NTB plate. Magnification approximately 5 ×. The cortex and medulla are clearly distinguishable.

blackening were obtained as in the first experiment, with the exception of the high reticular resolution in the kidney.

Recently, a desensitizing effect was observed when the spinal cord of a cat was placed in contact with an NTB emulsion surface for less than 30 sec, and the entire slide

exposed to light from an overhead incandescent lamp for less than 1 sec. The entire plate except that part in contact with the cord was black after development. The area in contact with the cord was composed of two parts. One was completely transparent, indicating complete desensitization of the emulsion to light, and was structurally associated with white matter. The other was very slightly blackened and was structurally associated with gray matter.

While we felt the blackening had been produced by direct chemical action, the possibility of blackening by

TABLE 1

RESULTS OF A TEST FOR PHOTONS PRODUCED BY A TISSUE AS DETECTED BY AN EASTMAN NTB EMULSION

	Tissue and emulsion separated by quartz slide		Tissue and emulsion in contact	
	Control not exposed	Exposed to ultraviolet light 5 min	Control not exposed	Exposed to ultraviolet light 5 min
2 hr	No image	No image	Image present, photographic density .07	Image present, photographic density .06
24 hr	No image	No image	Image present, photographic density .14	Image present, photographic density .13

photons produced in the fresh tissue could not be overlooked. It is well known that histological tissues produce photons by fluorescence after exposure to ultraviolet light. Usually fluorescence is produced only by ultraviolet light, is short-lived, and the number of photons is relatively large. In principle, however, there could be some compounds in the tissues which might be raised to a higher energy level by any portion of the spectrum and produce a delayed emission of a small number of photons. If this were so the photographic plate would integrate the effect over a long exposure, making observable a phenomenon unobservable instantaneously by the eye. There was also the possibility of a small number of photons from oxidative processes. Even though these possibilities seemed remote they were investigated in a preliminary manner.

Two series of experiments were carried out in which kidney and liver slices were separated from the photographic emulsion by (1) a glass cover slip and (2) quartz plates approximately 1 mm thick. In this experiment the animal, a rat, was dissected in the lighted room. After a 2-week exposure of the slices in the deep freeze, no image was observed.

In a repeat experiment using the kidney, a guinea pig was sacrificed in total darkness. Tissue slices exposed to ultraviolet light for 5 min and slices left in darkness were placed side by side on a quartz plate, which in turn was placed on an NTB plate. A similar set was placed in direct contact with emulsion of an NTB plate. The time from the end of ultraviolet exposure to placing on the photographic plates was about 3 min. The quartz plate was 1 mm thick. The results are given in Table 1.

After both 2- and 24-hr exposures at approximately −15° C, no image was found under the sections on the quartz plate. This indicates that the kidney picture shown in Fig. 2 was not produced by photons of fluorescence, oxidative luminescence, or any form of biochemical photon production in the range of wavelengths transmitted by the quartz and recorded by the NTB emulsion.

The experiments on direct contact, showing approximately equal densities for the tissues exposed and unexposed to ultraviolet light, for both 2 and 24 hr, indicate that there was no delayed emission of photons produced in the companion experiment and filtered out by the quartz. The interesting well-known observation of yellow fluorescence was seen on irradiation, but died out within a few seconds after the ultraviolet source was turned off.

As many of the autoradiographs made in this laboratory are fixed in Bouin's solution, experiments were run to test the possibility of artifacts from this source. Paraffin sections of rat lung, heart, kidney, and liver, which had been fixed in Bouin's solution, were placed in direct contact with an NTB plate. No blackening was observed after several weeks of exposure at approximately −15° C.

These preliminary experiments indicate that Figs. 1 and 2 were produced by direct chemical action. It should not be inferred that these observations prove the nonexistence of biochemically produced photons. Emulsions of different sensitivity or biological tissues under different conditions might show the production of a small amount of photons.

These results on NTB emulsions can be taken only as a warning when working with other emulsions. It does not follow that lantern slides or x-ray emulsions will show the same pattern or intensities. They may give the same or different patterns and may be more or less intense. For example, it is well known that hydrogen peroxide will fog many photographic emulsions (*3*), but it does not fog NTB emulsions. Also, the effect of histological fixative solutions and stains should be checked until proved harmless.

One advantage of the NTB plate, especially the NTB 2, or electron track plate, is that autographs can be differentiated from chemical fogging by virtue of the tracks for autographs and random grains for chemical fogging.

These observations suggest a new histochemical tool. The NTB plate with its low background fog and small grain size giving high resolution, is probably the best for this purpose, though other plates have not yet been tested.

It should not be inferred that this artifact makes autoradiography impossible. It does, however, emphasize the necessity of running controls on all solutions and tissues coming in contact with the emulsion. Another means of eliminating the artifact is by interposing a thin impermeable film between the subject of study and the emulsion. Stripping film is one type of emulsion designed for this purpose (*2*).

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