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EFFECT OF POLARIZED LIGHT ON THE PHARMACOLOGICAL PROPERTIES OF SOME DRUGS[1]

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Introduction

While the effect of light on chemical reactions has been known for a long time, and photochemistry has been receiving adequate attention for many years, the influence of light and other radiations on the pharmacological properties of drugs has not been studied until comparatively the last few years.

In the last decade or two, attention has been directed to the important role played by ultraviolet rays on the toxicity of a number of dyes, and very much more recently the effect of such radiations has come into prominence in connection with the marvelous strides in our knowledge of the vitamins. The earlier work on the dyes is summarized by Tappeiner and Jodelbauer[2] who have reviewed the previous work on the subject and contributed some original observations. These authors have found that eosin, while relatively non-toxic for paramecia in the dark, is very destructive to the same animalcules when irradiated by ultraviolet rays. Straub[3] analyzed this photo-dynamic effect further and obtained some evidence pertaining to oxygen being activated in the course of this phenomenon. Noguchi[4] and others have studied effects of light on the bactericidal properties of some dyes. Amsler and Pick[5] and Kolm and Pick,[6] studying the effects of eosin solutions on isolated organs, found that that compound was much more active when irradiated by ultraviolet rays than in the dark. Macht and Teagarden[7] studied the effect of ultraviolet irradiations on the fluorescent solutions of quinine and quinidine sulfates and found that these solutions were more active when so irradiated. Recently Macht[8] noted that the toxicity of sodium benzoate for yeast is much greater when exposed to sunlight than in the dark, and the remarkable experiments of Hess

[1] Read before a joint session of the Organic and Biological Divisions of the American Chemical Society at the Richmond meeting, April 12, 1927. A preliminary note on part of the subject was published by David I. Macht and J. C. Krantz, Jr. [*J. Am. Pharm. Assoc.*, **16**, 106 (1927)].

[2] Tappeiner and Jodelbaurer, *Ergebnisse Physiol.*, **8**, 698 (1909).

[3] Straub, *Arch. exptl. Path. Pharmakol.*, **51**, 583 (1904).

[4] Noguchi, *J. Exptl. Med.*, **8**, 30 (1908).

[5] Amsler and Pick, *Arch. exptl. Path. Pharmakol.*, **82**, 88 (1918).

[6] Kolm and Pick, *ibid.*, **86**, 1 (1920).

[7] Macht and Teagarden, *J. Pharmacol.*, **22**, 1 (1923).

[8] Macht, *Proc. Soc. Exptl. Biol. Med.*, **23**, 638 (1926).

and other investigators on the irradiation of codliver oil and cholesterol are well known to everyone.

The above observations on the effect of light on pharmacological reactions have hitherto been confined almost exclusively to a study of the ultraviolet and the visible rays of the solar spectrum. The senior author of the present paper has since 1921–1922 been interested in the effect of light on the action of various drugs, a subject which we may term "photopharmacology." In connection with a study of the keeping qualities of digitalis and cocaine, experiments have been made by Macht and Krantz[9] on the effect of ultraviolet as well as other radiations on solutions of these drugs. It was found that not only ultraviolet rays markedly produced deterioration in their potency but also that x-rays and radium emanations on the one hand and the much longer invisible infra-red rays of the spectrum produced changes in their pharmacodynamic properties. In this connection a study was undertaken in regard to possible effects of *polarized light* on the potency of the above drugs. The results obtained were most remarkable, and in the present paper it is proposed to describe in detail the findings in connection with three drugs, digitalis, cocaine and quinine. The pharmacological effects of polarized light hitherto have never been the object of any investigation. Indeed, no biological effects of any kind produced by polarized light have ever been described, with one important exception. While the above experiments were being carried on by the authors Miss E. S. Semmens, working in the Laboratory of Professor E. C. C. Baly, at Liverpool, and collaborating with him, discovered that the hydrolysis of starch by diastase into sugar is accelerated to a high degree by exposure to polarized light. Semmens and Baly[10] have made a short report of their work and deserve the credit of being pioneers in the study of the biological effects of polarized light. One of the present authors has repeated and corroborated their work with starch.[11] The present investigation on polarized light was begun by the authors before the English work on starch and diastase was published, and deals with an entirely new aspect of the subject, namely, the effect of polarized light on pharmacological reactions, this being the first study of the kind on record.

Methods of Irradiation

In the experiments to be reported two classical methods of obtaining polarized light were employed: polarization by means of Nicol prisms in some experiments, and polarization by piles of glass plates in other experiments. In the experiments with Nicol prisms, prisms of different sizes were utilized. In the earlier experiments performed in Baltimore, a large Nicol prism 3.5 cm. in diameter and 12 cm. long was utilized. We are

[9] Macht and Krantz, *Proc. Soc. Exptl. Biol. Med.*, **23**, 340 (1926).

[10] Semmens and Baly, *Proc. Roy. Soc.*, **97**, 250 (1924).

[11] Macht, *Proc. Soc. Exptl. Biol. Med.*, **22**, 473 (1924).

indebted to Professor Pfund, of Johns Hopkins University, for his courtesy in loaning this instrument. For a source of light with this large prism Mazda tungsten nitrogen electric bulbs of various sizes were employed, their power ranging from 5 to 75 watts. Where a more powerful source of light was desired for use with this prism, a Krohmayer mercury-vapor lamp (Hanovia Company) was used with a collar adjustment made so as to fit the aperture of the prism. Spectroscopic examination of the rays from the Krohmayer lamp which passed through the Nicol prism indicated, as was expected, that all but the longer ultraviolet rays were cut out by the prism.

In most of the experiments, all of the ultraviolet rays from the Krohmayer lamp were cut out by the use of glass containers or filters so that the drugs which were irradiated received rays only from the visible spectrum and the Krohmayer lamp served merely as a source of light of great intensity. Control experiments were made in each case with non-polarized light from the same source, the intensity of light being either exactly the same as that of the polarized rays employed, or intentionally made slightly greater in order to perform control experiments of the most crucial kind.

Smaller Nicol prisms, as well as the above large one, were used in the experiments performed in Newark and also in the later experiments performed in Baltimore.

The following scheme or arrangement was employed in irradiating the drug samples at the Hanovia Research Laboratory, in the spring of 1926.

The light source used was a 75-watt tungsten electric lamp operated on alternating current with series resistance.

In front of the bulb was supported a quartz water cell containing distilled water. The cell was placed in front of the Nicol prism in such a manner that all the light passed through the cell before going through the prism. The prism was placed in front of a slit in a black box and was blocked off with heavy black paper so that only the light going through the prism entered the box.

The intensity of the light passing through the Nicol prism was measured by the thermopile. A diamond-shaped slit of the same size as the prism was made in a piece of black paper and the intensity of the light passing through was measured by the thermopile. The resistance in series with the bulb was regulated so that the thermopile reading indicated the same intensity as with the Nicol prism.

The tubes containing the drugs were placed in the box and in the path of the beam going through the prism. The distance of the tube from the prism was regulated so that the entire tube was radiated. The prism was placed with the line cut across the black cork perpendicular to the table, and the filament of the bulb also perpendicular to the table.

The drugs were divided into two parts, one radiated with polarized and one with non-polarized light. In radiating the tubes with non-polarized

light the black diamond-shaped slit was substituted for the prism as described above.

Inasmuch as the authors have found, as will be shown further on, that irradiation, even with weak light, when polarized produces definite changes in drugs, another and slightly modified type of apparatus was employed in a different series of experiments. This set-up or arrangement was used in some of the most carefully and accurately performed experiments carried out by the authors, and consisted of the following. A wooden box with a detachable top is blackened inside and outside. Inside the box at one end is an attachment in which is fixed a rectangular quartz cell for the solutions of drugs. At the other end of the black box directly opposite the quartz cell is a circular aperture into which can be fitted snugly a Nicol prism, 2.5 cm. in diameter and 6 cm. long. In obtaining polarized light, the tube containing the Nicol prism is placed in the aperture so that its long axis is parallel to the table. In front of the Nicol prism is placed a small Spencer microscope lamp provided with a daylight filter. This lamp consists of a small Mazda lamp of 15 watts, enclosed in a dark cell with a window of glass which allows the light to diffuse through. The light from this lamp passing through the Nicol prism is polarized so that the drug contained in the quartz cell inside the black box is irradiated with polarized light. The amount of such polarized light and its intensity were very carefully determined by means of a thermopile. The exact wave lengths of which the light consisted were also determined by the spectrograph. In order to obtain a control light of exactly the same intensity and quality, with the exception of its not being polarized, the lamp is moved to a position from the aperture of the black box and the Nicol prism is removed. With the lamp in this position, and the Nicol prism taken out, the light entering the box is so calibrated as to be the exact control of the polarized light which was used. By means of this apparatus, which we may call for brevity the M.-A. (Macht-Anderson) apparatus, a drug can be radiated with polarized and non-polarized light for any given length of time so as to have the two forms of light of exactly the same intensity and wave length. Examples of experiments performed with the above apparatus will be given in the following section.

In order to obtain polarized light by the method of glass plates a different kind of apparatus was constructed. This apparatus was designed carefully by one of the authors and Professor A. H. Pfund, of the Department of Physics, Johns Hopkins University, and was constructed under Professor Pfund's supervision and calibrated by him. The plan of this apparatus is as follows. A box or cell in the form of a truncated pyramid 80 cm. high was constructed with a lower base 60 $\times$ 45 cm. and upper end about 25 cm. square, the back of the wall of the cell being perpendicular to the base, and the front wall and door slanting. At the upper

or small end of the cell a socket is fixed, into which is inserted a large, round, Mazda tungsten nitrogen electric bulb, of 500 watts power, which serves as source of light. The lower part of the apparatus or cell is divided into two compartments, completely separated from each other by a blackened partition. The light of the Mazda lamp is allowed to penetrate into the chamber on one side of the apparatus after first passing through a dozen plates of smooth glass, placed at the "polarizing " angle, so that this chamber is illuminated with highly polarized light. The light from the same Mazda lamp, on the other hand, is allowed to penetrate into the second or neighboring chamber after first passing through a pile of smooth plates of glass placed perpendicular to the line of propagation of the light, so that this second chamber is illuminated with non-polarized light. The number of glass plates in this second pile was adjusted so that the intensity of the non-polarized light was just equal to the intensity of the polarized light in the first chamber. By boring apertures in the floors of the two respective chambers and taking spectrophotographs of the two transmitted lights, it was found that the spectral range of light waves in the polarizing and non-polarizing chambers was the same, the shortest waves transmitted being about 3650 Å. The temperatures in the two chambers were practically the same, not deviating from each other by more than a fraction of a degree.

The source of light was an electric bulb of 700 candle power. This intensity, of course, was cut down by passage through the piles of plates, but the intensity of the transmitted light in each chamber was made the same by photometric calibration in the Physics Laboratory, performed by Professor A. H. Pfund. The intensities in the two chambers were compared by Professor Pfund by reflecting the lights passing through the two sets of glass plates, from a white surface, and allowing the rays to pass through a Lummer tube. The light from the two chambers was thus reflected diffusely, and hence was depolarized before the comparison was made. Thus, while the eye was used in comparing, it could not be argued that there might be a difference in the physiological effects of polarized and non-polarized lights on the eye. Of course such an objection would be purely hypothetical as, so far as is known, no difference in the effects on the eye between polarized and non-polarized lights has ever been noted, and if such a difference should be experimentally demonstrated, it would be a fundamental physiological discovery.

In order to make sure that small variations in intensity of the control did not affect the results, a number of experiments were made with the non-polarized light of a slightly greater or slightly lesser intensity than the polarized light (by changing the number of plates in the control chamber). Such variations did not appreciably change the marked effect of the polarized light.

The temperatures in the two chambers were the same to within a fraction of a degree, as indicated by thermometer readings and also by thermographic tracings. Here again a number of experiments were made in which the temperature in the control chamber was purposely made a little higher or a little lower than in the polarizing chamber, respectively, and the results obtained still showed a marked difference between the effects produced by polarized and non-polarized light.[12]

Methods of Pharmacological Evaluation

The drugs studied intensively in the present paper were a galenical preparation of tincture of digitalis and the pure crystalline drug cocaine, either in the form of the free base or of the hydrochloride. The hydrochloride of cocaine was used in most cases because of the very poor solubility of the alkaloid itself. In addition to these two drugs, the authors have studied also a number of others and more particularly quinine and cinchonidine. Extensive work is in progress on several important alkaloidal and other drugs, which will be reported in a later paper.

It is a well-known fact that physiological test objects are often so sensitive to the pharmacological effects of certain chemicals or drugs that by their means one can detect changes in those drugs much more easily than by any quantitative chemical methods. Indeed it is a fact that some of the most potent and important medicinal substances employed by pharmacologists and physicians can be standardized or assayed only by biological methods. Thus, for instance, the uterus of the virgin guinea pig will respond to such dilute concentrations of histamine as could not be quantitatively estimated by even the more refined microchemical methods and the very highly potent chemical body "A" obtained by Professor Abel[13] from the posterior lobe of the pituitary gland, is some 40 or even more times more powerful in this respect, as shown by tests on the isolated uterus and on certain preparations of the urinary bladder.[14]

Digitalis, undoubtedly one of the most important and useful drugs in the Pharmacopeia, is a drug the chemistry of which is still but incompletely known, and the only methods by which the therapeutic efficiency or potency of digitalis preparations can be estimated and compared are biological. While cocaine is a well-known chemical entity and its chemical structure is very well understood, a dilute solution of it can be assayed chemically only with the greatest difficulty or almost not at all, and even then the results are not sufficient for determining its pharmacological activity. The same is true of the majority of alkaloidal drugs employed by pharmacologists and physicians, so that in order to evaluate various samples of

[12] Macht, *J. General Physiol.*, **10**, 41 (1926).

[13] Abel, Rouiller and Geiling, *J. Pharmacol.*, **22**, 4, 289 (1923).

[14] Macht, *J. Pharmacol.*, **27**, 389 (1926).

the same drug biological or pharmacological methods of assay are quite indispensable and indeed are absolutely essential.

In the present work where the purpose of the irradiations was to detect even the smallest changes in the pharmacological properties of certain drugs, chemical methods could be employed only to a limited extent; the greatest amount of information had to be obtained and the greater reliance placed on pharmacodynamic tests and analyses.

Tincture of digitalis was used almost exclusively in the study of digitalis. The methods of assay employed in this case were of different kinds. In the first place the standard cat method of Hatcher and Brody was employed,[15] the exact technique being a slight modification introduced by Rowntree and Macht.[16] This method consists in the regular injection of tincture of digitalis diluted 1:10 with physiological saline solution into the femoral vein of a cat under light ether anesthesia. The injections are continued as described by the authors until the arrest of the heart in systole. An average of three experiments is usually made for an assay. The cat method has recently been shown by Lind Van Wijngarden, on the basis of a very large number of animal experiments, to yield quite reliable results in experienced hands.[17] The figures from which averages were made lay well within the experimental variation allowed by the formula of Wijngarden, that is, the mean of the percentage deviations from the average value of the N estimations made was less than $6.67\sqrt{N-1}$.

A still more sensitive and reliable method, especially for comparative examinations of different samples, has been described by one of the authors in collaboration with Krantz.[18] In contradistinction to the cat method which is a zoöpharmacological method, this method is phytopharmacological, employing living seedlings or plants as the biological test objects. These two methods, the cat method and the plant method, were used almost exclusively by the authors in the study of tincture of digitalis. In addition to these a comparison of digitalis tinctures exposed to polarized and non-polarized light was also made by other methods. Thus, in some cases, the goldfish method of Pittenger and Vanderkleed was used.[19] In still other experiments the comparative potency of the specimens treated with different forms of light was tested on frogs' hearts and finally some comparative tests were made by very interesting and effective methods described by Hanzlik and Shoemaker, in which the emetic effect of digitalis on pigeons is used as a criterion.[20] It may be stated at once that the results obtained by *all* of the above methods of experimenta-

[15] Hatcher and Brody, *J. Am. Pharm.*, **82**, 360 (1910).

[16] Rowntree and Macht, *J. Am. Med. Assocn.*, **66**, 870 (1916).

[17] Van Wijngarden, C. de Lind, *Arch. exptl. Path. Pharm.*, **12**, 252 (1926).

[18] Macht and Krantz, *J. Am. Pharm. Assocn.*, **16**, 210 (1927).

[19] Pittenger and Vanderkleed, *ibid.*, **24**, 427 (1925).

[20] Hanzlik and Shoemaker, *Proc. Soc. Exptl. Biol. Med.*, **23**, 298 (1926).

tion with digitalis were quite concordant and in most cases at least two methods, namely, the cat and the plant methods, were used in examining each sample, so the results were doubly reliable.

The evaluation of cocaine in weak solutions, such as are generally employed in surgical practice and which are known to be easily decomposed by heat and other physical agents, is very difficult by chemical means, and moreover gives little information as to the therapeutic or pharmacological efficiency of the drug. In order to detect deterioration in cocaine solutions more effectively, two new and very delicate methods have been elaborated by one of the authors, one of these being a zoöpharmacological method and the other being phytopharmacological. In the *zoöpharmacological* method the effect of cocaine solutions was studied on goldfish, *Carasseus aureus*. It was found that when fish are placed in even very dilute solutions of cocaine hydrochloride, for instance 1:10,000, they gradually become anesthetized and lose their sense of equilibrium. The fish "keel" over and finally float on their sides. A longer exposure leads to paralysis and death. To compare two specimens of cocaine, solutions are diluted to the same concentration in two vessels, small goldfish are immersed in the same and their behavior is studied. There is usually a preliminary stage of excitement which is followed by an anesthetic and a paralytic stage, and by noting the rapidity of onset of these phenomena it is easy to determine which solution is the more active pharmacologically. The more active solution in these cases is the one which produces anesthesia and paralysis more quickly.

The *phytopharmacological* method employed is even more delicate than the goldfish method. This method is based on the remarkable observations of Macht and Livingston obtained in a comparative study of the effects of cocaine on animal and plant protoplasm.[21] It was found by those authors that whereas the cocaine molecule itself is not very toxic for living seedlings of *Lupinus albus*, requiring sometimes as much as 6% or more of the solution of cocaine hydrochloride to inhibit growth completely, and not affecting growth at all in concentrations of 0.17%, the decomposition products of the cocaine molecule behaved very differently. When the cocaine molecule is hydrolyzed or broken down it yields one molecule each of the base ecgonine, methyl alcohol and benzoic acid according to the following equation:

$$\underset{\text{Cocaine}}{C_{17}H_{21}NO_4} + 2H_2O = \underset{\text{Ecgonine}}{C_9H_{15}NO_3} + \underset{\text{Benzoic acid}}{C_6H_5CO_2H} + \underset{\text{Methyl alcohol}}{CH_3OH}$$

Of these decomposition products benzoic acid, when tested on animal preparations (in the form of sodium benzoate) is practically non-toxic, but when tested on plant preparations sodium benzoate is extremely toxic. It was found that as little as 0.0007 of 1% inhibited growth of *Lupinus*

[21] Macht and Livingston, *J. General Physiol.*, **4**, 573 (1922).

albus completely, and 0.004% produced an appreciable inhibition of growth of the roots. This method suggested a delicate means of determining the decomposition of cocaine solutions. The greater the decomposition, the greater the toxicity of the solutions becomes for the growth of living seedlings. In the present investigation this method yielded very striking results, as will be seen from Table I. In most of the experiments solutions of cocaine hydrochloride were used, but in some weak solutions of the alkaloid cocaine itself were also studied. Slight changes in the hydrogen-ion concentration of the solutions were found to produce no appreciable effect on the growth of the seedlings, so that the inhibition of growth was due actually to the decomposition products and more particularly to benzoic acid.

In addition to the above zoöpharmacological and phytopharmacological methods of evaluating cocaine solutions, samples of cocaine hydrochloride which were exposed to polarized and non-polarized light were also tested in other ways. The comparative *anesthetic* effect of the two solutions was studied on the cornea of the rabbit. The comparative anesthetic effect was also studied on the living frog skin. Finally, the anesthetic effect on human beings was studied on some subjects by direct application to the mucous membrane of the cheeks. The difference between solutions of cocaine exposed to polarized and non-polarized light was also tested by physical chemical methods, namely, the determination of their hydrogen-ion concentration. We are indebted to Mr. W. C. Harden for his kind assistance in this work.

In evaluating the potency of quinine and cinchonidine sulfates and other salts, the authors again made use of the phytopharmacological method. Several years ago one of the authors described the effect of solutions of cinchona alkaloids on the growth of plants and found that they were quite toxic for them.[22] A comparative study of the toxicity of solutions of such alkaloids, therefore, was made by noting the growth of seedlings of *Lupinus albus* in definite percentage solutions of the drugs studied.

A few determinations were made also of the optical activity of quinine. In this case a quinine tartrate solution was employed which has been shown by Martinotti to be useful in the study of the keeping qualities of the drug, by means of observations on its optical rotation.[23]

Experiments with Digitalis

Irradiation of digitalis tincture was performed by all of the methods described above. Tinctures of digitalis were taken and in each case a given specimen was divided into two parts. One of these was irradiated with

[22] Macht, *Proc. Soc. Exptl. Biol. Med.*, **20**, 1971 (1922).

[23] Martinotti and Martinotti, *Notiz. chim. ind.*, **1**, 182 (1926).

polarized light; the other was irradiated with non-polarized light of the same intensity as a control. Occasionally the control with non-polarized light was irradiated with a stronger intensity of light or for a longer period of time. In all experiments the authors found that irradiation with polarized light produced a *change in the potency* of the tincture. Table I gives ample illustration of the findings.

In Expt. 101 irradiation was through a Nicol prism for one hour. The tincture of digitalis was in a glass test-tube, and the source of light was a Krohmayer lamp. In Expt. 106 irradiation was performed by the Macht-Pfund apparatus. The assay of the digitalis in these two experiments is given by the phytopharmacological method. Expts. 10 and 16 give the results obtained with irradiation by means of the Nicol prism and the Macht-Pfund apparatus, respectively, as indicated by assay experiments on cats. It will be noted that in each of these experiments irradiation with polarized light produced a deterioration of the tincture as indicated by a weaker pharmacological action of the same. It should be noted also that in all of these experiments the light employed consisted of rays belonging only to the visible spectrum plus some of the infra-red rays.

Expt. No. 200 was performed with the Macht-Anderson apparatus described above and in order to indicate the care exercised in controlling all the conditions in the experimentation, the physical data are here given in full. The apparatus used was the same as described above, the source of light being a microscope lamp with a daylight filter. The time of irradiation for each specimen, polarized and non-polarized, was exactly two hours. The radiation intensity for both polarized and non-polarized light was 70 ergs/sec./mm.^{2} The light from the microscope lamp as determined by spectroscopic examination consisted of the visible rays and infra-red rays. All of the measurements were made with the greatest care by means of the thermopile. The average light intensity at the surface of the irradiated solution inside the blackened box was 5.538 ergs/sec./mm. the measurements in this case being made through water so that infra-red rays of more than 14,000 Å. were eliminated. The time of irradiation as stated above was two hours, or exactly 7200 seconds. The average light intensity measured here is the value obtained when the total intensity falling on the area is divided by area. This is necessitated by the fact that the light transmitted by the prism is not disposed uniformly. These very careful measurements established beyond a shadow of a doubt that any difference between the specimens obtained after irradiation must be due to polarization of the light because this was the only variable, the intensity, time of irradiation, the volume, surface and concentration of the drug irradiated being in all instances identical. It will be noted from the protocols of Expt. 200 that the assay of the polarized and non-polarized specimens by the cat method, by the plant method and by the pigeon

method all indicated that the polarized specimen was much more deteriorated than a non-polarized one.

Expt. 210 gives the results obtained by irradiation with the Krohmayer lamp. The tincture of digitalis here, as in previous experiments, was exposed in a quartz cell. In this case the rays transmitted were the visible rays with the exception of the extreme red and the ultraviolet rays ranging from 2500 to 4000 Å. In this experiment the polarized and non-polarized specimens were irradiated for 30 minutes each because the actual visible light intensity with this lamp was much higher than in the instance of the irradiation by the microscopic lamp. However, the intensity was lower because the microscope lamp produced an abundance of heat which was

TABLE I

TINCTURE DIGITALIS

Effect of a 0.5% soln. on growth of *Lupinus albus*

Expt. 101	Index of growth, %	Expt. 106	Index of growth, %
Original non-radiated tincture	54	Original non-radiated tincture	70
Irradiated through Nicol K. lamp, 1 hr.	69	Polarized in Macht-Pfund apparatus, 4 hrs.	84
Control, non-polarized	57	Control, non-polarized	71

1:10 assay on cats, average of three

Expt. 10	Lethal dose, cc./kg.	Expt. 16	Lethal dose cc./kg.
Original non-radiated tincture	9.2	Original non-radiated tincture	9.5
Irradiated through Nicol K. lamp, 1 hr.	13.0	Polarized in Macht-Pfund apparatus; temp. 26°	14.9
Control, non-polarized	10.0	Control, non-polarized, 6 hrs.; temp., 26°	12.6

Expt. 200, Macht-Anderson apparatus; exposure, 2 hrs.; 2 cm. Nicol; microscope lamp; visible and infra-red rays

Expt. 210, 3.5 cm. Nicol; Krohmayer lamp; 30 min.; visible rays except red; ultraviolet, 2500–4000 Å.; Corning filter G986A

	Lethal dose for cats, cc./kg.		Growth of seedlings		Effect of 0.1 cc. on pigeons	
	Expt. 200	Expt. 210	Expt. 200	Expt. 210	Expt. 200	Expt. 210
Polarized	13.3	9.4	67	51	No vomiting in 20 min.	Vomits in 5 min.
Non-polarized	10.8	12.5	62	67	Vomits in 5 min.	No vomiting in 20 min.
Original, non-radiated	10.6	10.6	60	62	Vomits in 5 min.	

Expt. 30, May 14, 1926
Effect of soln. on cats; average of three

Visible and infra-red rays

	Lethal dose, cc./kg.
Polarized for 6 hrs.	11.5
Non-polarized control	9.0

Expt. 40, June 15, 1926
Effect of soln. on growth of *Lupinus albus*

	Index of growth, %
Normal specimen	51
Specimen polarized for 6 hrs.	68
Non-polarized control, 6 hrs.	52

not present from the light from the water-cooled Krohmayer lamp. The radiation density in this case, for both the polarized and non-polarized lights, was exactly the same, namely, 43 ergs/sec./mm.^{2} The average light intensity at the surface of the irradiated source was in this case 42.66 ergs/sec./mm.^{2} The intensity of this Krohmayer at the surface of the radiated specimen was 7.7 times that of the microscope lamp used above, while the time of irradiation with the Krohmayer was only one-fourth that used for the microscope lamp, or exactly 1800 seconds. Consequently, the samples irradiated by the Krohmayer rays received approximately twice as much light as those irradiated by the bulb. It was found that by irradiating digitalis tincture with ultraviolet light, a *further change or decomposition* in its constituents was produced. The tincture became actually more poisonous as indicated by the experiments on cats, pigeons and plants. Tincture of digitalis is a complex of a large number of active principles, the chemical nature of which is still not definitely known. It is, therefore, not at all surprising that progressive changes may be produced in it by irradiation with powerful rays for prolonged periods of time. The authors noticed in performing experiments on cats with such tinctures that the emetic properties of the tincture were markedly increased. This again indicated some change produced in the digitalis which is not ordinarily produced by the visible rays of light with which the other experiments were performed. *It is important to note, however, that here also it was the polarized light which produced a photodynamic effect, whereas the non-polarized control was relatively unaffected.*

Expts. 30 and 40 were performed with a Nicol prism and a Mazda nitrogen electric bulb of 75 watts.

Experiments with Cocaine

Table II exemplifies the effect of light on cocaine. In Expts. 120 and 50 the solutions were irradiated through a large Nicol prism. In Expts. 125 and 51 they were irradiated in the Macht-Pfund apparatus. It will be noted that in each case cocaine exposed to polarized light was markedly changed from the normal solution on the one hand and the one exposed to non-polarized light on the other hand. This was indicated by its lesser toxicity for animals (goldfish) and greater toxicity for seedlings, as explained above. Hydrogen-ion determinations also showed a change in the polarized specimens.

Expt. 220 shows the effect of polarized light of different wave lengths on solutions of cocaine hydrochloride. In this case irradiations with visible and infra-red rays were performed by the Macht-Anderson apparatus and microscope lamp under exactly the same physical conditions as described in the digitalis experiments. Irradiation with visible and ultraviolet rays was performed with the Krohmayer lamp with the same

physical constants as described under digitalis. Finally, ultraviolet rays alone were used in the third part of the experiment, by allowing the rays from the Krohmayer lamp to filter through a Corning filter G986A which allowed the rays from 2500–4000 Å. only to filter through. It will be noted in the case of cocaine that irradiations with polarized light of both short and long wave lengths produced a deterioration of the drug solution. This was tested out in every conceivable way; on living seedlings, on goldfish, on the rabbit's cornea, on the living frog's skin and on human mucosa.

TABLE II

COCAINE HYDROCHLORIDE

	*P*H		*Lupinus albus*,[d] index of growth, %		Time for paralysis of goldfish, min. (1:5000 soln.)[f]	
	Expt. 120[a]	Expt. 125[b]	Expt. 120[a]	Expt. 125[b]	Expt. 120[a]	Expt. 125[b]
Original soln.	5.1	5.1[c]	95[e]	90		
Polarized specimen	4.9	4.9	82[e]	80	16	No change after 25 min.
Non-polarized specimen	5.1	5.1	95[e]	88	6	6

Growth of *Lupinus albus*, %

	Expt. 50[g]	Expt. 51[h]
Original soln.	105	100
Polarized specimen	91	75
Non-polarized specimen	106	100

Expt. 220

1:10,000 Soln. of cocaine hydrochloride

	Visible and infra-red rays, no ultraviolet, 2 hrs. Effect on plants, %	Visible and ultraviolet, no red or infra-red, 30 min. Effect on plants, %	Ultraviolet alone, 30 min. Effect on plants, %
Polarized	83	80	84
Non-polarized	98	95	93
Normal	100		

[a] A fresh solution of cocaine hydrochloride 1:1000 was made in distilled water. Twelve cc. of soln. was irradiated with a small Mazda electric bulb of 15 watts for 45 min. Another portion was irradiated with non-polarized light of the same intensity for 50 min.

[b] Freshly prepared solutions of cocaine hydrochloride 1:1000 in distilled water. Two portions were irradiated for one hour in Macht-Pfund apparatus, one in polarized light, the other in non-polarized light of the same intensity, temperature of both being 26°.

[c] 1:1000 solution.

[d] The more cocaine decomposed, the greater the toxicity for plants.

[e] 1:10,000 solution.

[f] The more cocaine decomposed, the less the toxicity for animals.

[g] December 5, 1925, irradiated one hour with Nicol prism.

[h] December 8, 1925, irradiated three hours in Macht-Pfund apparatus.

In all three cases, the non-polarized specimens were more toxic on fish, and more anesthetic on rabbit's cornea, frog's skin and human mucosa.

Experiments with Quinine and Cinchonidine

In experiments with quinine and cinchonidine, solutions of quinine and cinchonidine sulfates, respectively, were used in most of the experiments. In some of the experiments a solution of quinine tartrate was examined. Irradiation of these solutions with polarized and non-polarized light of the visible spectrum by means of either a Nicol prism or the Macht-Pfund apparatus showed that such solutions became less toxic. This was easy to demonstrate quantitatively by means of phytopharmacological experiments. Expts. 60 and 70 will serve to illustrate the findings. In the case of quinine tartrate a few determinations with a polariscope were made which are indicated in Table III. Further polariscopic examinations are in progress.

TABLE III

PLANT TESTS, *Lupinus albus*

	Index of growth, %[a,b] Expt. 70	Index of growth, %[c] Expt. 60	Polariscope reading
Original soln.	77	56	—8°40′
Polarized specimen	87	72	—8°25′
Non-polarized specimen	77	64	—8°42′

[a] October 7, 1926. Solutions of quinine tartrate 1%, especially prepared by Mr. Fitzgerald Dunning. Two portions of this are irradiated for two hours in the two chambers of Macht-Pfund apparatus, at 26°; dilution, 1:100,000.

[b] The more deterioration in quinine, the less the toxicity for plants.

[c] January 20, 1926. Quinine sulfate irradiated for two hours in Macht-Pfund apparatus; 1:100,000 soln.

Experiments with Circularly-Polarized Light

In all of the above experiments with polarized light, *plane*-polarized light was obtained. The question naturally arises, what may be the effect of circularly-polarized light on the pharmacological properties of the above drugs? Such an investigation is at present in progress. It may be well, however, to state the results of a few preliminary experiments on the subject. The authors obtained circularly-polarized light by the use of the Macht-Anderson apparatus together with certain quartz plates. It has been found by physicists that if a ray of light passes through a plate of crystal quartz which has been cut perpendicularly to the axis, it is divided into two rays moving with different velocities, the vibrations in each ray, however, instead of being rectilinear and at right angles to one another, like plane-polarized light, are circular and in opposite directions. The ray of light is said to be circularly polarized. Ordinary complex light passing through such a plate will be circularly polarized, but the net result will be little, if any, different from that of the ordinary rays which were already vibrating in all directions. If the ray of light which passes through the quartz plate be already plane-polarized, for example, by means of a Nicol prism, the two beams emitted by the quartz plate will be circularly

polarized. Experiments were, therefore, performed by taking a quartz plate cut perpendicular to the axis of the crystal and allowing the plane-polarized light obtained by passing light from a microscope lamp through the Nicol prism to pass through this plate. Control experiments for the same were made by allowing the plane-polarized light to pass through another quartz plate cut parallel to the axis of the crystal. Table IV will give an idea of the results obtained with tincture of digitalis. The experiments indicate that circularly-polarized light produced even greater deterioration of the tincture than did the plane-polarized light. Further work on the subject is to be carried on.

TABLE IV

TINCTURE DIGITALIS

Macht-Anderson apparatus; 2cm. Nicol; microscope lamp; visible and infra-red rays; exposure, 2 hrs.

Irradiated by	Lethal dose for cats, cc./kg.	Effect on seedlings, index of growth, %	Effect of 0.15 cc. on pigeons, vomiting in
(Normal, non-radiated)	11.2	59	3 min.
Non-polarized light	11.4	63	3 min.
Plane-polarized light	13.1	68	4 min.
Circularly-polarized light	15.1	73	6 min.

The authors are also planning to study the effects of polarized and non-polarized monochromatic lights.

Discussion

The experiments described above make it evident that polarized light may produce a profound change in the pharmacological properties of certain drugs. Perhaps the most remarkable fact in this connection is that such changes *are produced by polarized light of the ordinary or visible spectrum.* This is not only of scientific interest but of practical importance, as may be concluded from the following considerations. Polarized light is not at all as rare or uncommon as might be supposed on first thought. Wherever light is reflected from smooth surfaces some polarization occurs. Again, whenever light passes through certain transparent media such as glass at certain angles a little polarization takes place. It is known to physicists and physical chemists that transparent sheets of cellulose polarize light to a certain extent. In other words, thin sheets of paper which are more or less transparent produce considerable polarization of light that passes through them. Bearing this in mind the authors performed the following experiment. A tincture of digitalis was divided into two parts and corked tightly in two flint glass bottles of exactly the same shape and quality. One of the bottles was wrapped in thin tissue paper such as is often used by drug manufacturers for wrapping various vials of medicaments. The two bottles were left standing in the sunlight for several weeks and then the tinctures were assayed carefully both by the

plant method and by the cat method. It was found that while the quantity of light, as determined by physical measurements, that passed through the paper into the wrapped specimen of digitalis was less than one-sixth the quantity of light which passed through the uncovered glass bottle, nevertheless, the deterioration of the digitalis specimen in the paper-covered specimen was actually *greater* than in the uncovered specimen. Thus the index of growth for plant seedlings of *Lupinus albus* given by the uncovered specimen was 66%, whereas the index given by the paper-covered specimen was 71%. The lethal dose for cats of the uncovered specimen, was 9.2 cc. per kg., whereas the lethal dose of the paper-covered specimen was 10.6 cc. per kg., a difference of nearly 15%. Bearing in mind that the amount of light to which the covered specimen was exposed was less than one-sixth that of the uncovered specimen, these findings are truly remarkable. Similar results were obtained by exposing specimens of digitalis in test-tubes wrapped in thin tissue paper to the light from a large electric bulb. As another example of the practical importance of polarized light we may quote from a recent article on some interesting studies concerning the follicular hormone of the ovary, by Jordan and Doisy,[24] in the Proceedings of the Society of Experimental Biology and Medicine, December, 1926. We read, "In an earlier publication Allen and Ellis call attention to the destruction of the hormone by ultraviolet light. Some data, which confirm and extend theirs, have been obtained, but our most striking observations are upon the effect of diffuse daylight from north windows upon the activity of the hormone." To the present authors these findings have undoubtedly a significance in relation to polarized light, as may be made clearer by another citation. We quote from the pioneer work of Baly and Semmens on the hydrolysis of starch,[25] "In this connection it is interesting to note that in the living leaf the starch synthesized during the day undergoes hydrolysis to sugars in the early evening when, as is well known, the light from the sky is polarized. The results obtained would seem to show that plane-polarized light exerts very definitely a selective photochemical effect compared with ordinary light." Again, on page 253, we read "These experiments have been repeated more than twenty times and similar results were obtained in all. In the early observations some hydrolysis was sometimes observed on the slide exposed to ordinary light, and this effect varied in a remarkable way. It was found, however, that this was due to stray light reflected from the bench top and from the legs of the microscope stand, and also from the glass slide, and hence partially polarized. When this stray light was excluded, the abnormal effect vanished and constant results were obtained."

In the opinion of the authors the above phenomena can be explained

[24] Jordan and Doisy, *Proc. Soc. Exptl. Biol. Med.*, **24**, 216 (1926).

[25] Baly and Semmens, *Proc. Roy. Soc.*, **97B**, 250 (1924).

satisfactorily on the basis of the photochemical and the photopharmacological effects of polarized light.

While the phenomena and experiments just described are rather unusual, on more careful reflection they do not seem to be as unexpected as one might be inclined to regard them. They are no more striking than the fact that solutions of organic chemicals possessing an asymmetric carbon atom have the power of turning the plane of polarized light to the right or to the left. If "for every action there is a reaction" we may look at this phenomenon from the other end and argue that if a given solution can turn the plane of polarized light to the right or to the left, then *vice versa* we may expect polarized light also to react on the solution, and tend to produce some rearrangement of its molecules. This is, however, only by way of speculation. The authors may state in this place that they are continuing further work on the effects of polarized light on a considerable number of other drugs and have already sufficient evidence to indicate that many such drugs are changed in their pharmacological properties by such light. Furthermore, it may be stated in this place, it was interesting to find that wherever such a change in the pharmacological properties was produced by polarized light, an inquiry into the chemical structure of the drugs studied, so far as is known, revealed that they were optically active. These results which are not final but require further study make it evident that optical activity of various chemical compounds is of even greater importance than has hitherto been supposed. We may conclude with a quotation from an article by the late Professor Arthur R. Cushny on the biological relations of optically isomeric substances, "This optical activity is, in fact, the most persistent evidence of life which we possess. An optically active alkaloid or acid may be kept for centuries after the plant which formed it and the chemist who isolated it are dead, but it will still possess its optical activity, testifying that it was formed by some living thing either directly or indirectly. When we find an optically active substance in the earth, we may know at once that it arose through the agency of life. The petroleum we burn, for example, must have arisen from living tissues, for it is optically active. Not only is it the most persistent sign of life, but it is the most definite physical characteristic of life. No other can be measured in actual numbers in the same way."

Summary

1. The effect of polarized light of different wave lengths was studied on a number of drugs and more particularly on tincture of digitalis solutions of cocaine and its salts, and quinine.

2. Polarized light was obtained by the two classical methods: (A) with a Nicol prism, (B) by means of piles of glass plates.

3. Controls were carefully performed with non-polarized light of exactly

the same physical properties and with the same physical constants except as to polarization.

4. It was found that polarized light beyond any doubt produced a very definite change in the pharmacological properties of the drugs studied as compared both with the normal specimens and with specimens irradiated by non-polarized light.

5. The practical significance of the above observations is discussed.

BALTIMORE, MARYLAND

[CONTRIBUTION FROM THE CHEMICAL LABORATORY OF THE U. G. I. CONTRACTING COMPANY]

THE SHIFT IN A NEAR INFRA-RED ABSORPTION BAND OF SOME BENZENE DERIVATIVES

BY JAMES BARNES AND W. H. FULWEILER

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In connection with a search for possible spectroscopic methods of identifying hydrocarbons some work was done on the absorption spectra of the aromatic series. Aside from the practical aspects of the results obtained it was thought that a preliminary report of some of the data might be of theoretical interest to those who are working in this field.

The extensive investigations of Coblentz[1] and Puccianti[2] on infra-red absorption spectra of organic liquids gave many important results. In recent years, certain special fields have been studied in a more detailed manner. We refer to the works of Márton,[3] Ellis[4] and others who showed that there appeared in the absorption spectra of the carbon-hydrogen compounds which they investigated a series of bands all at approximately the same wave length and that the values of these wave lengths almost formed an harmonic series, the fundamental being near 6.5μ.

Apparatus and Methods

All the above-mentioned observers used a prism spectroscope with a radiometer or a thermopile as the detecting instrument. In our work on the absorption spectra of some hydrocarbons and petroleum oils we have been using a grating spectroscope and photographing the bands on plates sensitized with neocyanine.

The source of radiation was a 65-watt lamp with a short, straight filament. It was surrounded by a hood with a hole in its side. The light passing through this hole fell on a lens which rendered it approximately parallel. It then passed through the absorption cell to another lens which

[1] Coblentz, *Carnegie Inst. Pub.*, **1905–8**.

[2] Puccianti, *Nuovo Cim.*, **11**, 241 (1900).

[3] Màrton, *Z. physik. Chem.*, **117**, 97 (1925).

[4] Ellis, *Phys. Rev.*, **27**, 298 (1926).