

the tetraploid level. The haploids would seem to provide an excellent opportunity for effecting gene transfer from the numerous cultivated and wild diploid ($2n=24$) *Solanum* species. They also represent promising material for studies on the nature of ploidy in *S. tuberosum* and chromosome differentiation within the tuber-bearing solanums.

S. J. PELOQUIN

R. W. HOUGAS

Crops Research Division, U.S.
Agricultural Research Service, and
University of Wisconsin, Madison

References and Notes

1. R. W. Hougas and S. J. Peloquin, *Nature* 180, 1209 (1957); R. W. Hougas, S. J. Peloquin, R. W. Ross, *J. Heredity* 49, 103 (1958).
2. This work was done in cooperation with the Inter-Regional Potato Introduction (IR-1) Project.
3. H. C. Choudhuri, *Trans. Roy. Soc. Edinburgh* 61, 199 (1944); M. S. Swaminathan and H. W. Howard, *Bibliographia Genet.* 16, 1 (1953).
4. J. G. Hawkes, *Scot. Plant Breeding Sta. Ann. Rept.* (1956), pp. 37-109.
5. E. V. Ivanovskaja, *Doklady Akad. Nauk, S.S.S.R.* 24, 517 (1939).

17 June 1958

Phosphorescence Spectra and Analyses of Some Indole Derivatives

Abstract. Phosphorescence spectra of solutions frozen and at low temperatures (77°K) possess more characteristic structure and detail than fluorescence spectra. They contain no background due to scattering of the exciting radiation. Thus greater analytical specificity and sensitivity are obtained. Some indole derivatives indistinguishable by spectrofluorimetry are easily differentiated by spectrophosphorimetry.

The substitution (1) of two monochromators for the customary light filters in fluorimetry, one for selecting the wavelengths of light for excitation, the other for the analysis of the resulting fluorescence, has made fluorimetry a rather general method of analysis of high sensitivity. The modification in technique was appealed to in the beginning especially for analyses of indole derivatives, which are of importance in the study of the central nervous system. In this report (2) we discuss characteristics of the closely allied spectra of phosphorescence (3) and their applicability for analyses, again with reference to indole derivatives.

The apparatus employed for fluorescence may serve for phosphorescence also, but the sample instead of being mobile as in a fluid solution must be rigid as in a glass or a crystal. Both fluorescence and phosphorescence spectra are emitted simultaneously, partially superimposed on each other. The time-intervals in which the intensities of the two

types of radiation diminish to half their values differ by orders of magnitudes, a characteristic which may be utilized to separate them. Indeed, a rotating phosphoroscope (3) between the two monochromators disposed at right angles as in fluorimetry was adequate for isolating the phosphorescence with half-life of the order of 1 second.

The phosphoroscope consisted of a rotating vertical cylindrical drum having two vertical slits 180 deg apart. On the axis was the solution in a fused silica tube within a fused silica Dewar flask. The retardation was the time required for a slit facing the first monochromator to make one quarter turn and face the second. At a rotation of 3000 rev/min the phosphorescence registered arose 5 msec after its excitation. A consequence was the elimination of the principal back-ground present in fluorescence which arises from the radiation of the light source scattered by the first monochromator.

The intensity of fluorescence increased with reduction in temperature and after solidification of the solution phosphorescence appeared at the expense of fluorescence. When some indoles had been dissolved only in water and frozen, the phosphorescence at 77°K was extremely feeble, but the addition of almost any solute such as an electrolyte increased the intensity (4). It appears that imperfection in the ice lattice favors trapping of excited states or electrons, forming what may be regarded as momentary color centers, in the same way as, under x-irradiation, color centers in crystals are favored by lattice imperfections introduced by impurities or mechanical strain (5). Variation of luminescence with solvent was also investigated. Some nonaqueous solutions yielded higher intensities than did aqueous solutions. Solvents were chosen that formed transparent glasses at the low temperature in order to reduce scattering and thereby achieve more intense luminescence.

Figure 1 shows recordings (6) of the phosphorescence spectra of the following substances (7) in transparent glasses of methanol and ethanol in ratio of 9:1. Indole (curve I) and tryptophan (curve II) exhibit different patterns. However, the fluorescence spectra of these substances at room temperature consist of virtually the same bell-shaped form with the same single maximum (1) at 360 mμ. 5-Hydroxytryptamine (serotonin) (curve III) shows still a different pattern. At room temperature only one maximum appeared at 330 mμ. At the low temperatures, indications of structure are evident in fluorescence but they are not so distinct as they are in phosphorescence.

Reserpine (not shown here) had an entirely different shape for its phospho-

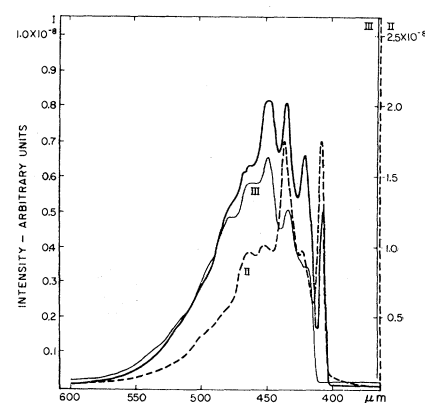


Fig. 1. Phosphorescence spectra of indole derivatives in alcohols at 77°K. I, Indole (2.5 mg/ml); II, tryptophan (0.5 mg/ml); III, 5-hydroxytryptamine (serotonin) creatinine sulfate (0.1 mg/ml) in glasses of methanol and ethanol, 9:1 by volume. Ordinates are in amperes.

rescent spectrum, a tall mound with two minor peaks at 450 and 470 mμ. Indoleacetic acid and tryptamine, also indistinguishable from indole and tryptophan at room temperature, could be differentiated from them and from each other by means of their phosphorescence spectra, which resembled the spectrum of tryptophan rather closely. Greater differences in relative maxima were shown by indoleacetic acid than by tryptamine.

In activation spectra different wavelengths produced phosphorescence spectra of different shapes as well as intensities. Such behavior furnishes highly specific criteria for identification.

We defer discussion of sensitivity to some later date. With our present equipment the sensitivity by phosphorescence was about 10 times as great as by fluorescence in the submicrogram region. The limitation appears as background due to the phosphorescence of solvents and of fused silica of containers. However, the phosphorescence contained no background due to scattered exciting radiation, which is chiefly limiting in the sensitivity of measurements by fluorescence.

SIMON FREED

WILLIAM SALMRE

Chemistry Department, Brookhaven
National Laboratory, Upton, New York

References and Notes

1. R. L. Bowman, P. A. Caulfield, S. Udenfriend, *Science* 122, 32 (1955).
2. This research was performed under the auspices of the U.S. Atomic Energy Commission.
3. P. Pringsheim, *Fluorescence and Phosphorescence* (Interscience, New York, 1949).
4. R. H. Steele and A. Szent-Györgyi [Proc. Natl. Acad. Sci. U.S.A. 43, 477 (1957)] found that the addition of glucose to aqueous solutions increased the intensity of phosphorescence.
5. N. F. Mott and R. W. Gurney, *Electronic Processes in Ionic Crystals* (Clarendon, Oxford, 1948). Impurities in fused silica also increase the number of color centers produced by radiation from uranium reactors.

6. The apparatus consisted of mercury arc (BH6, General Electric Co.), monochromators (Bausch and Lomb Optical Co.), photomultiplier tube (6903, Radio Corporation of America), and recorder (Brown Instrument Co.). The activation of the phosphorescence spectra consisted of a region from 280 to 310 mμ.
7. Sources of our materials: Serotonin was used as serotonin creatinine sulfate kindly furnished us by Dr. K. E. Hamlin, Jr., Abbott Laboratories, North Chicago, Ill., and by Sandoz Pharmaceuticals, Hanover, N.J. Both sources gave identical spectra. Reserpine was furnished by Mr. Louis Dorfman, Ciba Pharmaceutical Products, Summit, N.J. Indole was purified by crystallization and by several successive sublimations. Tryptamine hydrochloride, L(-) tryptophan, and indoleacetic acid, Eastman Organic Chemicals.

8 July 1958

Effect of Post Partum Separation of Mother and Kid on Maternal Care in the Domestic Goat

Abstract. Twenty-four goat mothers were separated from their newborn kids for 1 hour immediately following birth. Two months later these mothers were observed to nurse their own kids less and alien kids more than nonseparated mothers. Separation of mother and young in half the flock also resulted in abnormal "rejecting" behavior in some nonseparated mothers.

Recent studies (1) have demonstrated that the appearance of some types of normal, species-typical maternal behavior, often classed as "instinctive," is dependent upon the occurrence of specific experiences during critical periods in the life of the individual animal. Experimental or accidental changes in the "natural" environment at these times often result in the development of decidedly abnormal, species-atypical behavior.

Domestic sheep and goats normally rear their young within an individual-specific family structure. The suckling relationship between mother and young is typically limited to a particular parent and her offspring, and any attempt by a lamb or kid to nurse, or sometimes even

to approach, a mother not its own results in that mother's withdrawal from and often violent repulsion of the alien offspring.

Descriptions of sheep and goat parturition indicate that the experience of the mother immediately following birth is critical to the development of this individual-specific infant-rearing pattern. Separation of mother and newborn for a short time at birth results in at least the temporary rejection of the young by the mother when mother and offspring are reunited (2).

Some observations of the behavior of separated mothers, however, suggest that maternal behavior often is unstable. Separated mothers who at first reject their own or foster young may occasionally later accept them, and mothers who at first accept young sometimes later reject them (2, 3). The study described in this report was undertaken to investigate the long-term, general effects of early mother-young separation on the individual mother and on the population as a whole.

Twenty-four domestic goat mothers were separated from their newborn kids for periods ranging from ½ hour to 1 hour, 5 to 10 minutes immediately following birth. The kids were permitted, or helped, to nurse their own mothers when mother and kid were reunited. A control group of 21 mothers, equated for age and parity, were allowed to follow the normal newborn care-taking pattern. The usual life of the flock was not specifically interfered with further until approximately 2 months (Table 1, observation 1) and, again, 3 months (Table 1, observation 2) after birth, when mother-kid interaction was studied in the following manner.

All the kids in the flock were housed in a room apart from all the adult goats for from 6 to 10 hours. The kids were deprived of both food and water during this time; the mothers received food and water as usual. Three of the kids, of ap-

proximately the same age, were then brought into the experimental room. One minute later the mother of one of the three kids joined them, and all four were observed for 15 minutes through a one-way-vision glass. An observer recorded the time in seconds of the mother's nursing and butting behaviors by activating separate electric clocks for the duration of each type of behavior as it occurred. This procedure was repeated until each mother had been observed with her kid and two others.

In each instance, the three kids appeared highly excited and fearful when they were first brought into the unfamiliar room. With the appearance of the mother, all three kids rushed toward her and attempted to nurse. The immediate reaction of all the mothers was to back away from this onslaught for the first 30 seconds. After this initial period, most nonseparated (control) mothers began the process of establishing a private territory for herself and her own offspring, by butting away the two other kids each time they approached her. After several minutes of being butted the other kids kept their distances, while the mother nursed her own kid in the usual manner.

In contrast, the separated (experimental) mothers behaved in a distinctly abnormal manner during both observation periods (Table 1), nursing their own kids less than the nonseparated mothers ($p=0.01$) and nursing other kids more ($p=0.01$). Separated mothers nursed other kids as long as they nursed their own, whereas nonseparated mothers nursed other kids relatively little, as compared with the time spent nursing their own ($p=0.01$) (mean differences were tested for significance by t tests, which were computed separately for each observation period).

All butting behavior for both groups was normal; none of the mothers butted their own kids, although they butted other kids frequently.

An unexpected result of the study was the appearance of "rejecting" behavior (nursing neither their own nor other kids) among the mothers of the nonseparated group. Similar experimental analysis of the *post partum* behavior of another herd in which none of the kids had been separated at birth revealed no instances of "rejecting" behavior or of "indiscriminate" behavior (nursing other kids as long as, or longer than, their own), suggesting that the act of separating half the kids in the experimental herd had probably been the principal factor affecting maternal-young relationships of the nonseparated "rejecting" mothers. Although the specific cause of this effect on the nonseparated mothers is unknown, the "rejecting" behavior of the nonseparated mothers may have developed because their own kids wandered off shortly after birth and were

Table 1. Mean duration, in seconds, of maternal activities occurring during the 15-minute observation periods.

Condition	Separated mothers (N = 24)		Nonseparated mothers (N = 21)	
	Observation 1	Observation 2	Observation 1	Observation 2
Nursing own kids				
Mean	36.2	23.2	50.4	61.8
Standard deviation	11.4	9.7	16.5	22.3
Nursing other kids				
Mean	37.2	27.6	7.1	10.5
Standard deviation	9.3	10.1	6.8	8.8
Butting own kids				
Mean	0.0	0.0	0.0	0.0
Standard deviation	0.0	0.0	0.0	0.0
Butting other kids				
Mean	16.1	14.8	19.3	13.9
Standard deviation	15.3	12.6	15.7	10.0