

The Preparation and Properties of Chemiluminescent Compounds of the Lophine Type

EVANS W. COTTMAN, R. B. MOFFETT, and S. M. MOFFETT
The Cottman Laboratory of Photochemical Research, Madison

Introduction

Lophine was prepared by Radziszewski, using the two following methods:

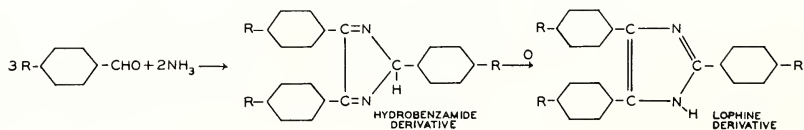
First Method. In 1877 he prepared lophine by treating benzaldehyde with an excess of ammonium hydroxide, and subsequently oxidizing the hydrobenzamide so formed, with atmospheric oxygen.¹

Second Method. In 1882 he prepared lophine by ammoniating equivalent amounts of benzil and benzaldehyde with dry ammonia gas in alcoholic solution at a temperature of 45°-50°.² He was the first to report on the chemiluminescent properties of lophine when oxidized in alcoholic potash solution.

Since little or nothing has been done along this line since 1882, the present writers have repeated Radziszewski's work, have attempted to obtain the optimum conditions for luminescence of lophine, attempted the preparation of a number of lophine derivatives, and studied the spectra of those lophine derivatives which show a marked degree of chemiluminescence. The writers are greatly indebted to the Eli Lilly Research Laboratories of Indianapolis, in which the analytical and spectrometric work was done.

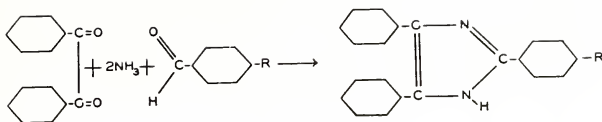
Discussion

Preparation of Lophine Derivatives.—Following the two methods of Radziszewski, outlined above, it was reasoned that if substituted benzaldehyde derivatives were used in place of benzaldehyde itself, the corresponding lophine derivatives would be prepared. For example, if the first method were used, a trisubstituted lophine derivative would result:



If Radziszewski's second method were used, it was reasoned that a monosubstituted lophine derivative should result from using a substituted benzaldehyde derivative, the substitution occurring in the 2-position of the imidazole ring:

¹ Radziszewski, 1877. Ber. 10:70-5.
² Radziszewski, 1882. Ber. 15:1493-5.



The following benzaldehyde derivatives were used in attempting to prepare the monosubstituted and trisubstituted lophine derivatives and the hydrobenzamide derivatives:

1. Piperonal (Eastman Kodak Co.).
2. *m*-Nitrobenzaldehyde (C. A. F. Kahlbaum Chemischefabrik, Adlershof bei Berlin).
3. *p*-Dimethylaminobenzaldehyde (Eastman Kodak Co.).
4. *p*-Hydroxybenzaldehyde.
5. Anisaldehyde (Eastman Kodak Co.).
6. *o*-Nitrobenzaldehyde (Dr. Theodor Schuchardt Görlitz).
7. Salicylaldehyde (Eastman Kodak Co.).
8. Resorcyaldehyde (Eastman Kodak Co.).
9. Veratraldehyde (Eastman Kodak Co.).
10. Vanillin (Eastman Kodak Co.).

The first method, described above, was used in preparing all hydrobenzamide derivatives and trisubstituted lophine derivatives. The second method was used in preparing all monosubstituted lophine derivatives. All the benzaldehyde derivatives were reactive toward aqueous ammonia except the *p*-dimethylaminobenzaldehyde. In general, the reactions paralleled those for the preparation of the normal lophine. A few of the compounds were gummy, and all attempts to purify them were unsuccessful. The nitrogen analyses of these compounds were valueless, but it was deemed worthwhile to include these compounds in the discussion because of their luminescent properties.

While definite proof is lacking, the evidence seems to point to the fact that a number of the desired lophine and hydrobenzamide derivatives were actually prepared, whereas some were doubtful. This evidence is as follows:

1. The method of preparation was similar to that of preparing lophine itself.
2. The physical appearances of the reactions were similar in many cases to those involved in normal lophine preparation.
3. Many of the nitrogen analyses checked fairly closely with the theoretical values.
4. Most of the compounds produced a chemiluminescence, similar to that of normal lophine, upon oxidation.

In view of the fact that complete proof of structure is lacking, the writers wish to be understood that the names used in this article for these compounds are employed for convenience and are provisional.

Optimum Conditions for the Luminescence of Lophine.—For luminescence of lophine to take place, the solution must be alkaline. Either sodium hydroxide or potassium hydroxide should be used. The solvent most generally used is ethyl alcohol, but methyl alcohol, acetone, dioxan, and several other solvents may be used successfully. To complete the

conditions, a proper oxidizing agent, together with a peroxide (usually hydrogen peroxide), must be used. The choice of oxidizing agent is an important point. Out of about fifty such reagents tried, the four following ones were found to be most successful: sodium hypochlorite, potassium ferricyanide, perchromic acid, haemoglobin. As the sodium hypochlorite was most extensively used in these researches, full directions for its preparation and use will be given in the Experimental Part.

Chemiluminescent Properties of the Derivatives.—Numbers I, II, V, VIII, IX and X, described in the Experimental Part, give no light and are assumed to be hydrobenzamide derivatives. Numbers IV, VII and XII are assumed to be monosubstituted lophine derivatives, and numbers III, VI and XI, trisubstituted lophine derivatives. The luminescent properties of the lophine derivatives will be described individually in the Experimental Part.

The spectra of lophine and eight of its most luminescent derivatives were studied with a Hilger spectroscope. All the luminescent derivatives give a pale yellowish light similar to that of normal lophine. The yellow color appears deeper in the more intense lights. Examined through the spectroscope, the spectra are found to be continuous, and lying approximately between 4800 and 6000 Angstrom units. The point of maximum intensity is about 5300 Angstrom units. The width of the visible band, of course, increases or decreases according to the intensity of the light. Since the intensity of the light is small, it was found necessary to strengthen it in order to take the spectroscopic readings. The device used for this purpose is described in the Experimental Part. An unsuccessful attempt was made to photograph the lophine spectrum, exposures up to three hours being made with a super-sensitive panchromatic film.

Experimental Part

A. Preparation of the Derivatives

I. *3, 4, 3', 4', 3'', 4''-tris (methylenedioxy) hydrobenzamide*: Pulverized piperonal is set aside in a stoppered flask with an excess of ammonium hydroxide. The derivative forms in about three days as a whitish crystalline cake. It is purified by rinsing several times on the filter with hot alcohol, in which it is slightly soluble. The yield is small.

II. *m, m', m''-tris (nitro) hydrobenzamide*: *M*-nitrobenzaldehyde is set aside in a stoppered flask with an excess of ammonium hydroxide for three days. The derivative forms as a very hard light brown cake. The brown color is dissolved out with hot alcohol, leaving the purified derivative as a flesh-colored residue.

III. *2, 4, 5-tris-m-nitrophenylimidazole*: No. II is fused in air and heated with constant stirring. It cools to a very dark brown brittle mass which is moderately soluble in hot alcohol. It is purified by recrystallization from hot alcohol. It gives rather poor luminescence of short duration.

IV. *2-m-nitrophenyl-4, 5-diphenylimidazole*: Equivalent amounts of benzil and *m*-nitrobenzaldehyde in alcoholic solution are saturated with

dry ammonia gas at 45°-50°. The derivative separates out as a bright yellow precipitate. It is purified by recrystallization from hot alcohol. It gives a fair degree of luminescence.

V. *p, p', p''-tris (methoxy) hydrobenzamide*: Anisaldehyde is set aside in a stoppered flask with an excess of ammonium hydroxide for three days. A brownish crystalline cake forms, and is steeped in hot alcohol several times to remove the impurities. The final residue is nearly white.

VI. *2, 4, 5-tris-p-methoxyphenylimidazole*: No. V is fused and heated in air with constant stirring. It fuses to an orange colored liquid which hardens, upon cooling, to a brittle vitreous mass. All attempts to get this into a crystalline form failed. However, a nitrogen analysis was made of the substance, with surprisingly close results. The chemiluminescent properties of this compound are exceptionally good. The light is of about the same intensity as that of lophine, but of longer duration. Of course, no melting point was obtainable.

VII. *2-p-methoxyphenyl-4, 5-diphenylimidazole*: Equivalent amounts of benzil and anisaldehyde in alcoholic solution are saturated with dry ammonia gas at 45°-50°. A white precipitate settles out and is filtered off. Upon standing, yellow crystals separate out from the filtrate. They are purified by washing with ether and recrystallizing from hot alcohol. This compound gives a yellow light noticeably brighter than that of lophine, but of shorter duration.

VIII. *o, o', o''-tris (hydroxy) hydrobenzamide*: An excess of ammonium hydroxide is added to salicylaldehyde and an immediate yellow precipitate forms. It darkens and hardens upon standing. It is dissolved in hot alcohol from which it separates very slowly into yellow crystals and an oil. The crystals are washed with ether to remove the oil.

IX. *2, 4, 2', 4', 2'', 4''-tris (dihydroxy) hydrobenzamide*: An excess of ammonium hydroxide, added to resorcydaldehyde, produces a very dark red solution. After standing several days, this is dried on a water bath, forming a red powder, coated with black. It is somewhat soluble both in alcohol and water. The red powder is purified by washing alternately with water and alcohol. The substance decomposes before reaching its melting point.

X. *3, 4, 3', 4', 3'', 4''-tris (dimethoxy) hydrobenzamide*: Veratraldehyde is set aside in a stoppered flask with an excess of ammonium hydroxide for three days. It swells up forming a flocculent white precipitate which fills the flask. This is purified by recrystallizing from hot acetone.

XI. *2, 4, 5-tris (3, 4-dimethoxyphenyl) imidazole*: No. X is fused and heated in air. It cools to form a very dark brittle substance, which is very soluble in alcohol. It is precipitated from alcoholic solution by water containing ethylene glycol. The final product is grayish brown. It gives a good luminescence.

XII. *2 (3, 4-dimethoxyphenyl)-4, 5-diphenylimidazole*: Equivalent amounts of benzil and veratraldehyde in alcoholic solution are saturated with dry ammonia gas at 45°-50°. The substance behaves similarly to the preparation of No. VII. First a white precipitate separates.

This is filtered off. Upon standing, yellow crystals separate from the filtrate. These are purified by washing with ether and recrystallizing from hot alcohol. This compound gives a luminescence slightly more intense than normal lophine.

TABLE I.—Numerical Data on the Derivatives

No. *	Probable empirical formula	Melting point	Nitrogen anal. calc.	Nitrogen analysis found	
I	C ₂₄ H ₁₈ N ₂ O ₆	94°	6.51%	6.21%	6.14%
II	C ₂₁ H ₁₅ N ₅ O ₆	140°	16.16%	15.88%	16.01%
III	C ₂₁ H ₁₃ N ₅ O ₆	126°	16.24%	16.05%	16.09%
IV	C ₂₁ H ₁₅ N ₃ O ₂	12.31%	12.21%	12.20%
V	C ₂₄ H ₂₄ N ₂ O ₃	127°	7.21%	7.14%	7.18%
VI	C ₂₄ H ₂₂ N ₂ O ₃	7.23%	7.16%	7.28%
VII	C ₂₂ H ₁₈ N ₂ O	202°	8.59%	8.38%	8.45%
VIII	C ₂₁ H ₁₈ N ₂ O ₃	136°	8.09%	8.03%	8.12%
IX	C ₂₁ H ₁₈ N ₂ O ₆	Decomposes	7.10%	6.82%	6.95%
X	C ₂₇ H ₃₀ N ₂ O ₆	132°	5.85%	5.77%	5.65%
XI	C ₂₇ H ₂₈ N ₂ O ₆	91°-95°	5.88%	5.78%	5.80%
XII	C ₂₃ H ₂₀ N ₂ O ₂	7.86%	8.00%	8.04%

* The names of these compounds are given in the text.

An attempt was made to prepare the following compounds by the methods outlined above:

- 2, 4, 5-tris (3, 4-methylenedioxyphenyl) imidazole from piperonal.
- 2 (3, 4-methylenedioxyphenyl) 4, 5-diphenylimidazole from piperonal.
- 2-*p*-dimethylaminophenyl-4, 5-diphenylimidazole from *p*-dimethylaminobenzaldehyde.
- 2-*o*-nitrophenyl-4, 5-diphenylimidazole from *o*-nitrobenzaldehyde.
- 2-*o*-hydroxyphenyl-4, 5-diphenylimidazole from salicylaldehyde.

These substances could not be purified sufficiently to give satisfactory analyses. However, they were all strongly chemiluminescent, which would seem to point to the fact that the lophine derivatives were probably prepared to some extent.

All attempts to ammonate *p*-hydroxybenzaldehyde and vanillin proved futile. Likewise, attempts to prepare the following compounds yielded no results, either with respect to analysis or to chemiluminescence:

- p*, *p'*, *p''*-tris (dimethylamino) hydrobenzamide by ammonation of *p*-dimethylaminobenzaldehyde.
- o*, *o'*, *o''*-tris (nitro) hydrobenzamide by ammonation of *o*-nitrobenzaldehyde.
- 2, 4, 5-tris-*o*-nitrophenylimidazole by atmospheric oxidation of the supposed hydrobenzamide derivative.
- 2, 4, 5-tris-*o*-hydroxyphenylimidazole by atmospheric oxidation of derivative No. VIII.
- 2, 4, 5-tris (2, 4-dihydroxyphenyl) imidazole by atmospheric oxidation of derivative No. IX.
- 2 (2, 4-dihydroxyphenyl)-4, 5-diphenylimidazole by ammonation of resorcyaldehyde and benzil.

B. Optimum Conditions for Luminescence of Lophine

Three solutions are prepared:

Solution 1 (Lophine solution): To a saturated alcoholic solution of lophine, an equal volume of alcohol is added.

Solution 2: 1 part 3% hydrogen peroxide is added to 7 parts water.

Solution 3: 5% sodium hypochlorite*.....25cc.
Sodium hydroxide10 gm.
Water175cc.

For each trial, 2cc. of Solution 2 is added to 4cc. of Solution 1. The room is then darkened and 2 cc. of Solution 3 is added to the mixture. An excellent luminescence results.

C. Device for Intensifying the Light

A glass tube, two feet long and one-half inch outside diameter, was sealed at both ends. Three small side entrance tubes were attached just back of the front end. An exit tube was attached at the rear end. The entire tube was silvered on the outside, except for a small window in the front end. This window was placed against the slit of the spectroscopy with the axis of the tube in direct line with that of the collimating tube of the spectroscopy. The three entrance tubes were connected to separate reservoirs containing the the various required solutions. Lophine solution was allowed to enter through one of the tubes, hydrogen peroxide solution through the second, and the mixture of potassium hydroxide and sodium hypochlorite through the third. The instructions for making up these solutions have already been given. As the solutions mix, luminescence occurs within the tube. This is intensified by the reflections and by the length of the tube.

Summary

1. A number of chemiluminescent substances were prepared through ammonation of benzaldehyde derivatives.
2. The methods used in preparing these substances followed those used in preparing hydrobenzamide and lophine.
3. Incomplete evidence indicates that a number of hydrobenzamide and lophine derivatives were prepared from the corresponding benzaldehyde derivatives.
4. The spectra of the brighter lights were studied.
5. Optimum conditions for the luminescence of lophine were determined.

* The commercial cleaning agent, "Clorox," may be used successfully for this purpose