

**Studies on Derivatives of Malachite Green.
Protolytic Equilibria and Reaction-Rate Constants of
m-Hydroxy and *m*-Methoxy Malachite Green**

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Dyestuffs of the Malachite Green type give in water solutions five species, two coloured (B and G) and three colourless (C, S', and S'') between which the following reversible reactions take place:

$B + H^+ \rightleftharpoons G$; $C + 2H^+ \rightleftharpoons S' + H^+ \rightleftharpoons S''$; $G + H_2O \rightleftharpoons S''$;
 $B + H_2O \rightleftharpoons S'$; $B + OH^- \rightleftharpoons C$. The equilibria between B and G and between C, S', and S'' are reached instantaneously whereas the hydration equilibria are reached only after a certain time.

Through spectrophotometric measurements dissociation, hydration and reaction-rate constants for the above indicated reactions have been determined for *m*-hydroxy and *m*-methoxy Malachite Green in water solutions having a constant ionic strength 0.5 M at 20°C. The method was the same as that used by the author in previous investigations of a similar nature.

A comparison between the constant values obtained shows that there is a marked difference between the *m*-hydroxy and *m*-methoxy derivatives only in strongly alkaline solutions.

A comparison between Malachite Green and the dyes now investigated shows that the influence of a *m*-hydroxy or a *m*-methoxy substituent is relatively small in acid and weakly alkaline solutions.

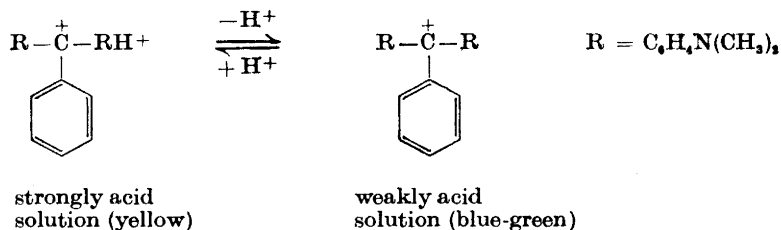
Previously, Malachite Green (MG) and a number of its *ortho* and *para* derivatives, with the substituent in the amino free phenyl group, were investigated¹. In the present paper the corresponding investigations on the *m*-hydroxy MG and *m*-methoxy MG are reported. Further, the measurements have now been extended to also include the reactions in strongly alkaline solutions.

The reactions of the dyestuffs now investigated can be expressed with the same equilibrium scheme that was previously used by the author² in studies

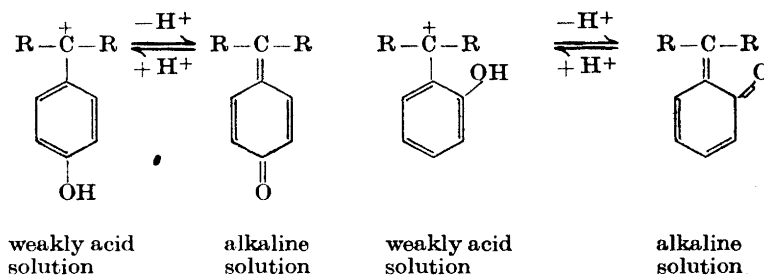
on the influence of temperature on protolytic equilibria and reaction rates of Methyl Green.

m-Hydroxy and *m*-methoxy MG show very great similarities in regard to absorption curves, protolytic constants, and reaction rates. Only in strongly alkaline solutions is there found a marked difference between the two compounds. Previously investigated *p*-hydroxy and *p*-methoxy MG (Ref.¹) show a striking mutual similarity when $\text{pH} < 6$. At a higher pH the *p*-hydroxy derivative, on account of its acidic $-\text{OH}$ group, can give rise to two additional forms, one coloured and one colourless, which cannot be formed by the corresponding methoxy compound. *o*-Hydroxy MG and *o*-methoxy MG have also been investigated and the results will soon be published in this journal. Like the corresponding *para*-derivatives the *ortho*-derivatives show great similarities in acid solutions and striking dissimilarities in alkaline solutions where the hydroxy compound can form two additional species, one coloured and one colourless.

Malachite Green can give two coloured ionic species,

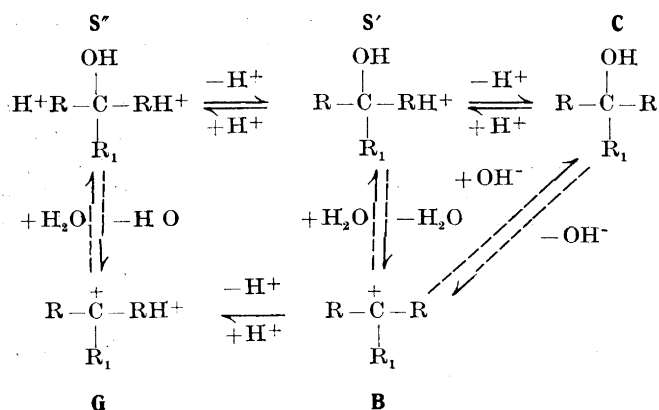


p-Hydroxy and *o*-hydroxy MG can give another coloured form through splitting off a proton from the OH-group. The following quinoid structures then become possible:



On the other hand, *m*-hydroxy MG can not give rise to a certain coloured form through splitting off a proton from the OH-group, since this does not give any possibility of an additional quinoid structure. Therefore we can formally use the same equilibrium scheme as for the *m*-methoxy derivative. In the Discussion part some differences in strongly alkaline solutions will be treated.

Neutral water solutions of the dyestuffs now investigated are beautifully blue-green. In strongly acid solutions they at first turn yellow and then are strongly discoloured. In alkaline solutions they are colourless. The reactions may be written schematically in the following way, where $R = C_6H_4N(CH_3)_2$ and $R_1 = C_6H_4OH$ or $C_6H_4OCH_3$



Definitions of constants and symbols

$$K_1 = \frac{[G]}{[B][H^+]} ; K_2 = \frac{[S']_\infty}{[G]_\infty} ; K_3 = \frac{[S'][H^+]}{[S'']} ; K_4 = \frac{[C][H^+]}{[S']} ; \\
 K_6 = \frac{[B]_\infty}{[S']_\infty}$$

$$h = [H^+]; oh = [OH^-]; K_w = [H^+][OH^-]$$

$$k_1 = \text{rate constant for the reaction } G + H_2O \rightarrow S''$$

$$k_2 = \text{» » » » » } S'' \rightarrow G + H_2O$$

$$k_3 = \text{» » » » » } B + H_2O \rightarrow S'$$

$$k_4 = \text{» » » » » } S' \rightarrow B + H_2O$$

$$k_5 = \text{» » » » » } B + OH^- \rightarrow C$$

$$k_6 = \text{» » » » » } C \rightarrow B + OH^-$$

$$k = \text{rate constant for the total reaction}$$

$$\epsilon_B, \epsilon_G = \text{the molar extinction coefficient of B and G}$$

$$e_0, e_t, e_\infty = \text{the absorbance per cm of a solution at the times 0 and } t \text{ and at equilibrium (thickness of absorbing layer } d = 1 \text{ cm)}$$

$$E_0, E_t, E_\infty = \text{the absorbance when } d \neq 1 \text{ cm}$$

$$e_B, e_G = \text{the absorbance per cm of a solution containing only B or G}$$

$$C_M = \text{the total concentration of dyestuff}$$

The constants in the equilibrium scheme can be calculated if the following data are determined for a large number of solutions with varying pH: (1) The absorbance, e_0 , at the moment of mixing a dye stock solution with appropriate

buffers; (2) The absorbance, e_∞ , when equilibrium is reached; (3) The pH of the solutions; (4) The total rate constant, k , for the reactions that change the intensity of the colour of the solutions either through hydration of the coloured forms (B and G) to the colourless (C, S', and S'') or vice versa.

The following equation for e_0 can be derived

$$e_0 = \frac{e_B + hK_1 e_G}{1 + hK_1} \quad (1)$$

Eqn. (1) requires that the dyestuffs are present only as the coloured form B (or G) when it is mixed with the buffer solution. This can not be realized for *m*-hydroxy MG or *m*-methoxy MG in water solutions. In acetate buffers, pH \approx 5, these dyestuffs have a maximum colour intensity when about 97 % of the individual dye is present as the coloured ion B while the remaining 3 % is distributed among the colourless forms S'', S', and C. Such dye-solutions, very weakly buffered, were used as stock solutions.

We can transform eqn. (1) and at the same time replace e_B and e_G with e'_B and e'_G , where

$$\frac{e'_B}{e_B} = \frac{e'_G}{e_G} = \frac{[B]_0 + [G]_0}{[B]_0 + [G]_0 + [S'']_0 + [S']_0 + [C]_0}$$

and we obtain

$$\frac{h}{e'_B - e_0} = \frac{h}{e'_B - e'_G} + \frac{1}{K_1(e'_B - e'_G)} \quad (2)$$

The terms K_1 and e'_G can be obtained from eqn. (2) by a graphical method when h , e_0 , and e'_B have been determined.

For the equilibrium absorbance, e_∞ , the following equation can be derived

$$e_\infty = \frac{e_B + hK_1 e_G}{1 + 1/K_6 + K_4/hK_6 + hK_1(1 + K_2)} \quad (3)$$

In acid solutions, pH $<$ 2, we can approximate eqn. (3) and write

$$K_2 = \frac{e_G}{e_\infty} + \frac{e_B - e_\infty(1 + hK_1 + 1/K_6)}{hK_1 e_\infty} \quad (4)$$

From eqn. (4) K_2 can be determined when e_B and e_G are known.

In weakly alkaline solutions eqn. (3) is approximated and transformed to

$$\frac{h}{e_\infty} = h \frac{1 + K_6}{K_6 e_B} + \frac{K_4}{K_6 e_B} \quad (5)$$

Eqn. (5) gives a straight line from which $(1 + K_6)/K_4$ is obtained.

KINETIC MEASUREMENTS

The equilibrium between the two coloured forms B and G is reached instantaneously. The same is true for the equilibrium between the colourless forms C, S', and S". The hydration equilibrium between a coloured and a corresponding colourless form and the equilibrium $B + OH^- \rightleftharpoons C$, on the other hand, are reached only after a measureable time. The following rate expression can be derived (cf. Refs.²⁻³).

$$k = \frac{k_2 h^2 + k_4 h K_3 + k_6 K_3 K_4}{h^2 + h K_3 + K_3 K_4} + \frac{k_1 h K_1 + k_3 + k_5 oh}{h K_1 + 1} = \frac{1}{t} \ln \frac{e_0 - e_\infty}{e_t - e_\infty} \quad (6)$$

In strongly acid solutions eqn. (6) is approximated to

$$k = k_2 + \frac{k_1 h K_1 + k_3}{h K_1 + 1} \quad (7)$$

and in strongly alkaline solutions to

$$k = k_3 + k_6 + k_5 oh \quad (8)$$

Between these two extreme approximations we can make a smaller approximation of eqn. (6) in the pH-range 4-8.

$$k = \frac{k_4 K_3 h}{h^2 + h K_3 + K_3 K_4} + k_3 \quad (9)$$

Eqn. (9) can be simplified further to

$$k = k_4 h / K_4 + k_3 \quad (10)$$

when pH = 7-8.

Approximate values of k_1 , k_2 , and k_4 can be determined from eqns. (7) and (9) while eqns. (8) and (10) give exact values of k_3 and k_5 but not of k_6 since $k_6 \ll k_3$. Then non-approximated values of k_1 , k_3 , and k_4 can be calculated from eqn. (6). The rate constant k_6 is obtained from the expression $k_6 = k_5 K_w K_6 / K_4$. Finally K_3 , K_4 , and K_6 are obtained from eqns. (5) and (6) by combining equilibrium measurements with kinetic measurements.

EXPERIMENTAL

Chemicals and solutions. *m-Hydroxy Malachite Green.* The leuco compound was prepared in a nitrogen atmosphere, through condensation of *m*-hydroxybenzaldehyde with *p*-dimethylaniline in conc. hydrochloric acid and urea. The leuco compound was recrystallized from benzene-ethanol. Light green needles, m.p. 134°C, were obtained. The dyestuff was prepared by oxidizing, with lead dioxide, and the leuco compound dissolved in a calculated amount of 5 M HCl and diluted with 10 % acetic acid. From the oxidized product a perchlorate was prepared that was obtained as small green crystals after repeated recrystallizations from 2 % acetic acid. (Found: C 62.6; H 5.68; O 17.6; N 6.22; Cl 7.72. Calc. for $C_{23}H_{15}O_5N_2Cl$: C 62.1; H 5.66; O 17.98; N 6.30; Cl 7.97).

m-Methoxy Malachite Green was prepared in the same manner as the *m*-hydroxy derivative. The leuco compound was obtained as white needles, m.p. 126°C, and the perchlorate of the dyestuff as small dark green crystals. (Analysis of the perchlorate. Found: C 63.1; H 5.87; O 17.60; N 6.14; C 7.73. Calc. for $C_{24}H_{27}O_5N_2Cl$: C 62.8; H 5.93; O 17.43; N 6.11; Cl 7.72).

Stock solutions of the dyes were prepared by weighing and dissolving the perchlorates in acetone that then was diluted with a potassium chloride solution giving a final solution with ionic strength 0.5 M and containing 4 % of acetone. The stock solutions, with $C_M = 2 \times 10^{-5}$ M, were buffered very weakly with an acetate buffer, $\text{pH} \approx 5$, and were diluted, when necessary, with 0.5 M potassium chloride containing 4 % of acetone. The perchlorates are so slightly soluble in water that it is almost impossible to prepare reproducible stock solutions with a fixed C_M by dissolving the perchlorates directly in 0.5 M KCl.

Hydrochloric acid, sodium acetate, disodium hydrogen phosphate, potassium dihydrogen phosphate, and sodium hydroxide, all of analytical grade, were used in the buffer solutions. *Potassium chloride* of analytical grade was added to keep the ionic strength constant (0.5 M).

Apparatus. The determinations of $[\text{H}^+]$ were carried out titrimetrically ($\text{pH} \leq 2$ and $\text{pH} \geq 12$) and potentiometrically ($2 < \text{pH} < 12$). For the potentiometric measurements a reference solution ($\text{pH} = 2$) having the composition 0.01 M HCl + 0.49 M KCl was used. In the present paper pH always means $-\log[\text{H}^+]$ and not $-\log a_{\text{H}^+}$. For the electrodes quinhydrone and platinumplate were used for $\text{pH} \leq 8$ and platinum-black and hydrogen gas for $\text{pH} > 8$. The temperature was kept at 20.0°C .

For the spectrophotometric measurements a modified Beckman DU quartz spectrophotometer with photomultiplier was used. The absorption cell house was kept at 20.0°C by pumping water from a thermostat through its walls and the whole apparatus was placed in a room kept constant at 20°C . Further details of the experimental technique may be found in Refs.^{2,3}

MEASUREMENTS AND RESULTS

The absorption curves of the two dyestuffs now investigated show such a similarity that only curves for one of the dyestuffs have been drawn in Fig. 1. In Fig. 1 the absorption curves are seen for the blue-green ion B in acetate buffer, $\text{pH} \approx 5$, and the yellow ion G in dilute hydrochloric acid. The molecular extinction coefficients were calculated from experimentally determined absorption curves and eqn. (3). ($e_B = C_M \epsilon_B$ and $e_G = C_M \epsilon_G$). The following values were obtained:

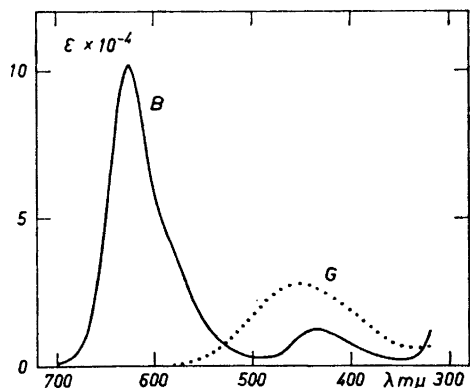


Fig. 1. *m*-Hydroxy Malachite Green. Absorption curves for the blue-green ion, B, in acetate buffer and the yellow ion, G, in dilute hydrochloric acid.

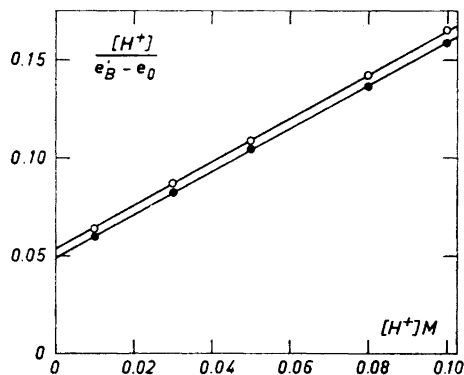


Fig. 2. $[\text{H}^+]/(e'_B - e_0)$ as a function of $[\text{H}^+]$ for the determination of K_1 for *m*-hydroxy MG (●) and *m*-methoxy MG (○).

m-hydroxy MG

$$\begin{aligned}\lambda_{\text{Bmax}} &= 621 \text{ m}\mu \text{ resp. } 430 \text{ m}\mu \\ \varepsilon_{\text{Bmax}} &= 9.4 \times 10^4 \text{ resp. } 1.8 \times 10^4 \\ \lambda_{\text{Gmax}} &= 454 \text{ m}\mu \\ \varepsilon_{\text{Gmax}} &= 2.9 \times 10^4\end{aligned}$$

m-methoxy MG

$$\begin{aligned}\lambda_{\text{Bmax}} &= 622 \text{ m}\mu \text{ resp. } 430 \text{ m}\mu \\ \varepsilon_{\text{Bmax}} &= 9.5 \times 10^4 \text{ resp. } 1.8 \times 10^4 \\ \lambda_{\text{Gmax}} &= 455 \text{ m}\mu \\ \varepsilon_{\text{Gmax}} &= 2.9 \times 10^4\end{aligned}$$

Determination of K_1 . The dye stock solutions were mixed with an equivalent volume of suitable HCl + KCl-buffers and e_0 was determined by extrapolation of the absorbancies to $t = 0$. The measurements were carried out at the wavelengths $\lambda = 621$ and $622 \text{ m}\mu$ where *m*-hydroxy MG and *m*-methoxy MG, respectively, have an absorption maximum. In Fig. 2 $[\text{H}^+]/(e'_\text{B} - e_0)$ is plotted against $[\text{H}^+]$. We get linear relations, and the slopes, l , and the intercepts, y , on the y -axis give according to eqn. (2) for *m*-hydroxy MG ($e'_\text{B} = 0.918$): $y = 0.0492 \text{ M}$, $l = 1.096$, $e_\text{G} = 0.006$, $K_1 = 22.3 \pm 0.3 \text{ M}^{-1}$ and for *m*-methoxy MG ($e'_\text{B} = 0.912$): $y = 0.0540 \text{ M}$, $l = 1.104$, $e_\text{G} = 0.006$, $K_1 = 20.4 \pm 0.3 \text{ M}^{-1}$.

Determination of $K_4/(K_6 + 1)$. The dye stock solutions were mixed with appropriate phosphate buffers and the equilibrium absorbancies, E_∞ , were read off after 24 h. Table 1. $C_\text{M} = 5 \times 10^{-6} \text{ M}$, $d = 2 \text{ cm}$. In Fig. 3 $[\text{H}^+]/E_\infty$ is plotted as a function of $[\text{H}^+]$. According to eqn. (5) linear relations are obtained which give for *m*-hydroxy MG: $y = 1.20 \times 10^{-7} \text{ M}$, $l = 1.088$, $K_4/(K_6 + 1) = 1.103 \times 10^{-7} \text{ M}$ and for *m*-methoxy MG: $y = 1.31 \times 10^{-7} \text{ M}$, $l = 1.070$, $K_4/(K_6 + 1) = 1.224 \times 10^{-7} \text{ M}$.

Table 1. *m*-Methoxy and *m*-hydroxy Malachite Green. Determination of $K_4/(K_6 + 1)$. $C_\text{M} = 5 \times 10^{-6} \text{ M}$, $d = 2 \text{ cm}$, $\lambda = 622 \text{ m}\mu$, $T = 20.0^\circ\text{C}$.

	<i>m</i> -methoxy MG	<i>m</i> -hydroxy MG
$[\text{H}^+] \times 10^7 \text{ M}$	$E_\infty = 2 e_\infty$	$E_\infty = 2 e_\infty$
14.48	0.858	0.857
8.32	0.812	0.812
5.30	0.758	0.761
3.45	0.689	0.696
2.26	0.605	0.617
1.400	0.500	0.513
0.824	0.376	0.390
0.372	0.215	0.229
0.1844	0.122	

Determination of K_2 . The dye stock solutions were mixed with suitable HCl + KCl-buffers. The solutions were thermostatted and the equilibrium absorbancies, E_∞ , were measured in 5 cm absorption cells. The constant K_2 was calculated from eqn. (4) when e_B , e_G , and K_6 were determined. The results

Table 2. *m*-Hydroxy Malachite Green. e_{∞} -, e_0 -, and k -values for determination of K_2 , K_1 , and k_1 . $\lambda = 621 \mu\text{m}$, $C_M = 1 \times 10^{-5} \text{ M}$, $d = 1, 2, \text{ and } 5 \text{ cm}$, $e_B = 0.946$, $e'_B = 0.918$.

[H ⁺] M	e_{∞}	K_2	e_0	$k \text{ min}^{-1}$	$k_1 \text{ min}^{-1}$
0.010	0.1190	30.2	0.750	0.0403	0.152
0.020	0.0630	30.5			
0.030	0.0440	29.8	0.553	0.0728	0.152
0.040	0.0330	30.2			
0.050	0.0270	29.8	0.439	0.0917	0.152
0.060	0.0226	29.8			
0.080	0.0174	29.3	0.330	0.1091	0.152
0.100	0.0140	29.3	0.287	0.1163	0.151
Mean		29.9			0.152

are recorded in Tables 2 and 3. From these tables the following mean values are obtained: *m*-hydroxy MG, $K_2 = 29.9 \pm 0.6$; *m*-methoxy MG, $K_2 = 33.7 \pm 0.7$.

Kinetic measurements. If the dye stock solutions are mixed with NaOH + KCl-buffers, the solutions are decolourized with a rate that is directly proportional to [OH⁻]. In Fig. 4 the rate constants of the decolourization, k in eqn. (8), are plotted against [OH⁻]. Straight lines are obtained which pass almost through the origin. This means that in this pH-range $k_3 + k_6 \ll k_5 \text{oh}$. Therefore, only k_5 can be determined from Fig. 4. For *m*-hydroxy MG, $k_5 = 11.7 \pm 0.2 \text{ min}^{-1} \text{ M}^{-1}$ and for *m*-methoxy MG, $k_5 = 32.0 \pm 0.5 \text{ min}^{-1} \text{ M}^{-1}$.

The rate constant k_3 was determined from kinetic measurements in phosphate buffers, pH = 7–8. The approximate eqn. (10) permits a good determination of k_3 but not of k_4/K_4 . In Fig. 5 k is plotted as a function of [H⁺]. The straight lines give the following k_3 values: *m*-hydroxy MG, $k_3 = 0.00654 \pm 0.00010 \text{ min}^{-1}$; *m*-methoxy MG, $k_3 = 0.00622 \pm 0.00010 \text{ min}^{-1}$.

Table 3. *m*-Methoxy Malachite Green. e_{∞} -, e_0 -, and k -values for determination of K_2 , K_1 , and k_1 . $\lambda = 622 \mu\text{m}$, $C_M = 1 \times 10^{-5} \text{ M}$, $d = 1, 2, \text{ and } 5 \text{ cm}$, $e_B = 0.948$, $e'_B = 0.912$.

[H ⁺] M	e_{∞}	K_2	e_0	$k \text{ min}^{-1}$	$k_1 \text{ min}^{-1}$
0.010	0.1170	33.9	0.755	0.0373	0.151
0.020	0.0624	33.9			
0.030	0.0428	33.7	0.568	0.0689	0.152
0.040	0.0324	33.8			
0.050	0.0264	33.5	0.451	0.0894	0.155
0.060	0.0220	33.6			
0.080	0.0166	33.7	0.349	0.1050	0.152
0.100	0.0134	33.6	0.307	0.1131	0.152
Mean		33.7			0.152

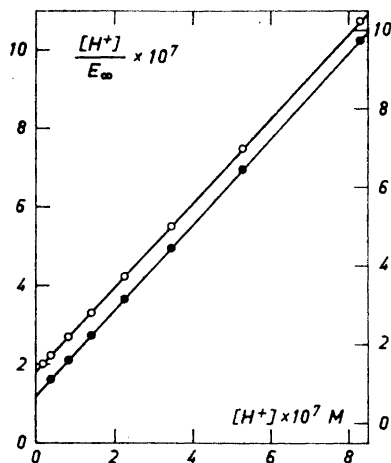


Fig. 3. $[H^+]/E_{\infty}$ as a function of $[H^+]$ for the determination of $K_4/(K_6 + 1)$ for *m*-hydroxy MG (●), ordinate left, and *m*-methoxy MG (○), ordinate right.

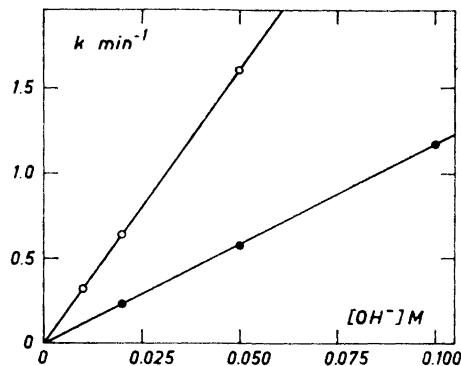


Fig. 4. k plotted against $[OH^-]$ for the determination of k_5 . *m*-Hydroxy MG (●), *m*-methoxy MG (○).

Determination of k_1 and k_2 . These rate constants were determined by kinetic measurements in HCl + KCl-buffers. The fading rate was determined and approximative values of k_1 and k_2 were initially calculated from eqn. (7). Tables 2 and 3 contain the k -values and the correct values of k_1 and k_2 calculated later from eqn. (6). The following values were obtained: *m*-hydroxy MG, $k_1 = 0.152 \pm 0.002 \text{ min}^{-1}$ and $k_2 = 0.00508 \pm 0.00015 \text{ min}^{-1}$; *m*-methoxy MG, $k_1 = 0.152 \pm 0.002 \text{ min}^{-1}$ and $k_2 = 0.00451 \pm 0.00015 \text{ min}^{-1}$.

Determination of k_4 , K_3 , K_4 , and K_6 . If k is differentiated with respect to $[H^+]$ in eqn. (9) k is found to have a maximum when $[H^+] = \sqrt{K_3 K_4}$. The dye stock solutions were partially decolourized by making them 0.02 M with respect to hydrochloric acid. The following reactions then occur $B + H^+ + H_2O \rightarrow G + H_2O \rightarrow S'$ and $B + H^+ + H_2O \rightarrow S' + H^+ \rightarrow S''$. These partially decoloured dye solutions were then quickly mixed with acetate buffers, and the colour was restored owing to the reactions $S'' \rightarrow S' + H^+ \rightarrow B + H^+ + H_2O$ and $S'' \rightarrow G + H_2O \rightarrow B + H^+ + H_2O$. The rate constants for the restoring of the colour were determined.

In Fig. 6 k is plotted as a function of pH. The following values are obtained: for *m*-hydroxy MG, $k_{\text{max}} = 0.167 \text{ min}^{-1}$ when $\text{pH} = 4.88$ which gives $K_3 K_4 = 1.74 \times 10^{-10} \text{ M}^2$ and for *m*-methoxy MG, $k_{\text{max}} = 0.160 \text{ min}^{-1}$ when $\text{pH} = 4.87$ which gives $K_3 K_4 = 1.82 \times 10^{-10} \text{ M}^2$.

If K_6 is fixed, K_4 can be determined from the known value of $K_4/(K_6 + 1)$, and K_3 from the known value of $K_3 K_4$. Further, k_4 can be obtained from $k_4 = k_3 K_6$ and k_6 from $k_6 = k_5 K_w K_6 / K_4$. Different values of K_6 were tried using eqn. (6). Within the pH-range 4–6 the terms $k_4 h K_3$ and k_3 are dominating in eqn. (6). Therefore the approximate values of k_1 and k_2 , obtained from

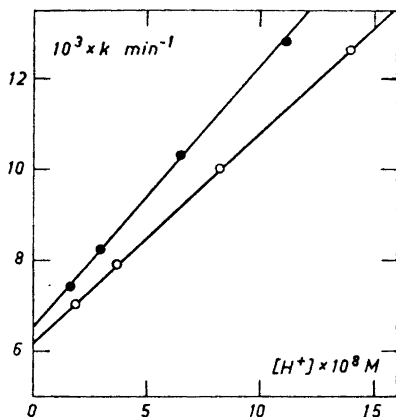


Fig. 5. k plotted against $[H^+]$ for the determination of k_5 . m -Hydroxy MG (●), m -methoxy MG (○).

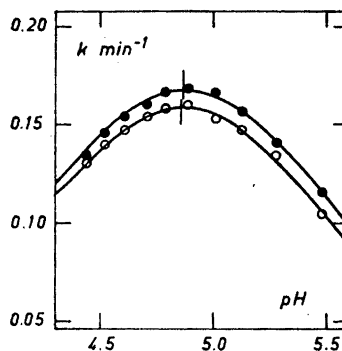


Fig. 6. k as a function of pH. The solid curves have been calculated from eqn. (6). The symbols have been determined by kinetic measurements in acetate buffers. m -Hydroxy MG (●), m -methoxy MG (○).

eqn. (7) by earlier measurements, can be used. The values of K_6 were chosen so that the curve calculated from eqn. (6) coincides best with the experimentally found k -values.

The best coincidence was found for m -hydroxy MG when $K_6 = 42.0 \pm 0.5$ which gives $K_3 = (3.68 \pm 0.05) \times 10^{-5}$ M, $K_4 = (4.73 \pm 0.06) \times 10^{-6}$ M, $k_4 = 0.275 \pm 0.008$ min⁻¹ and for m -methoxy MG when $K_6 = 44.0 \pm 0.5$ which gives $K_3 = (3.32 \pm 0.05) \times 10^{-5}$ M, $K_4 = (5.49 \pm 0.07) \times 10^{-6}$ M, $k_4 = 0.274 \pm 0.008$ min⁻¹. From a previous investigation we have $K_w = 1.24 \times 10^{-14}$ M² and k_6 can now be calculated from the relation $k_6 = k_5 K_w K_6 / K_4$. Thus for m -methoxy MG, $k_6 = (1.29 \pm 0.06) \times 10^{-6}$ min⁻¹ and for m -hydroxy MG, $k_6 = (3.18 \pm 0.15) \times 10^{-6}$ min⁻¹.

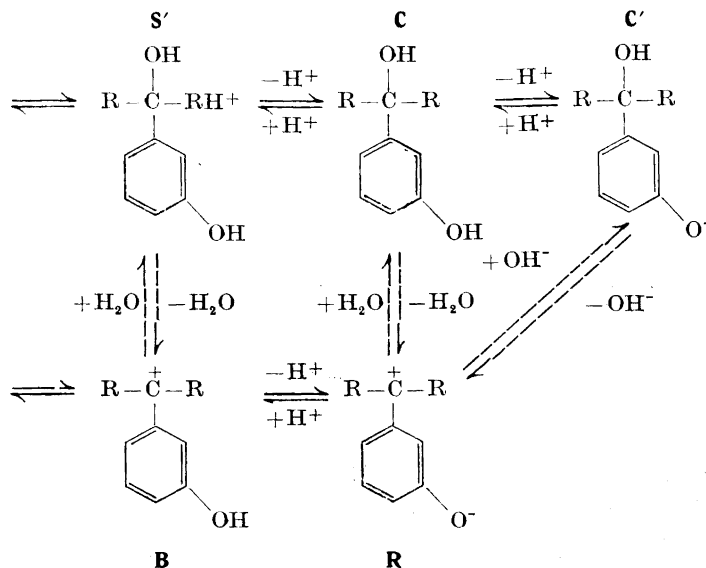
In Fig. 6 it can be seen that the curves calculated from the non-approximated eqn. (6) agree with the experimentally obtained k -values.

By definition, $K_1 K_2 K_3 K_4 = 1$. The values of this product that were determined are as follows: m -hydroxy MG, $K_1 K_2 K_3 K_4 = 1.03$ and m -methoxy MG, $K_1 K_2 K_3 K_4 = 1.00$.

DISCUSSION

A comparison between the values of the constants of m -hydroxy MG and m -methoxy MG shows a very great similarity when $pH < 8$. Corresponding values differ only by 0–16%. On the other hand, in strongly alkaline solutions, there is a marked difference between the rate constants. The rate constants k_5 and k_6 for m -methoxy MG are, respectively, 2.7 and 2.5 times as great as those for m -hydroxy MG.

The explanation of the difference in strongly alkaline solutions is probably that the equilibrium scheme on page 1894 does not cover all of the possible reactions of *m*-hydroxy MG. The hydroxy-group gives off a proton in alkaline solutions, and the following reactions are obtained some of which can not be established by measurements.



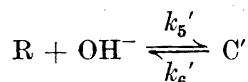
The following constants are defined as follows:

$$K_7 = \frac{[\text{R}][\text{H}^+]}{[\text{B}]} ; K_8 = \frac{[\text{C}'][\text{H}^+]}{[\text{C}]} ; K_9 = \frac{[\text{C}]_\infty}{[\text{R}]_\infty}$$

If B and R have a different colour, K_7 can be determined in the same way as K_1 . However, B and R do not show any noticeable difference in colour and K_7 can not be determined. An idea of K_7 might be obtained if the equilibrium measurement were influenced by K_7 in the pH-range 7–8 where $K_4/(K_6 + 1)$ was determined. This is not the case and therefore $\text{p}K_7$ is certainly >9 . Nor are the kinetic measurements in strongly alkaline solutions ($\text{pH} \gtrsim 12$) appreciably influenced by K_7 which means that $\text{p}K_7$ is certainly <11 .

When $\text{pH} > 9$ *m*-hydroxy MG, at reequilibrium, is present only in the form of the colourless species S', C, and C' and this is the reason why K_8 and K_9 can not be determined. For the *para*- and *ortho*-derivatives the following values of the constants were obtained: *p*-hydroxy MG, $K_7 = 1.1 \times 10^{-7}$ M, $K_8 = 1.20 \times 10^{-10}$ M, and $K_9 = 8.4$ (Ref.¹); *o*-hydroxy MG, $K_7 = 2.7 \times 10^{-9}$ M, $K_8 = 6.1 \times 10^{-10}$ M, and $K_9 = 0.15$ (to be published). For *m*-hydroxy MG both $\text{p}K_7$ and $\text{p}K_8$ probably lie in the range 9–10 which gives a value of 100–1 000 for K_9 .

The reason why k_5 and k_6 are considerably greater for *m*-methoxy MG than for *m*-hydroxy MG, therefore, probably lies in the fact that the kinetic measurements at $\text{pH} \gtrsim 12$ for the latter dyestuff give the total rate-constant for the reaction



From the kinetic measurements k_5' is obtained which is numerically identical with k_5 on page 1899. Of the two structures, corresponding to B and R, R has the greater electron density on the central methane carbon atom. Therefore it seems reasonable that the reaction $\text{R} + \text{OH}^- \rightarrow \text{C}'$ has a smaller rate constant than the reaction $\text{B} + \text{OH}^- \rightarrow \text{C}$. The latter reaction may be assumed to proceed with approximately the same speed for *m*-hydroxy MG as for *m*-methoxy MG. However, the rate constants of the reactions $\text{B} + \text{OH}^- \rightleftharpoons \text{C}$ or $\text{R} + \text{H}_2\text{O} \rightleftharpoons \text{C}$ can not be determined, for *m*-hydroxy MG as K_7 , K_8 or K_9 are not known and the reaction $\text{R} + \text{OH}^- \rightarrow \text{C}'$ dominates the fading reactions at high pH.

Table 4. Comparison between corresponding constants of Malachite Green and its *m*-hydroxy and *m*-methoxy derivatives.

	K_1	K_2	$K_3 \times 10^5$	$K_4 \times 10^5$	K_6	$k_1 \text{ min}^{-1}$	$k_2 \text{ min}^{-1}$	$k_3 \text{ min}^{-1}$	$k_4 \text{ min}^{-1}$
Malachite Green	28	22	2.7	5.8	68	0.168	0.00783	0.00647	0.439
<i>m</i> -Hydroxy »	22	30	3.7	4.7	42	0.152	0.00507	0.00654	0.275
<i>m</i> -Methoxy »	20	34	3.3	5.5	44	0.152	0.00451	0.00622	0.274

If the values of the constants of Malachite Green, *m*-hydroxy MG, and *m*-methoxy MG are compared, it is found from Table 4 that a *m*-hydroxy or a *m*-methoxy group in the amino free phenyl ring has a comparatively small influence on the equilibrium and reaction-rate constants. The meta-substituents do not give any marked resonance or steric effects. The inductive influence of the negative groups $-\text{OH}$ and $-\text{OCH}_3$ may be seen in an about 30 % increase in the acid constants $1/K_1$ and K_3 and a somewhat greater increase in the hydration constants K_2 and $1/K_6$.

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