change in the resting potentials in the unstained fibres on being exposed to light for several minutes. On being exposed to light the stained fibres showed varying degrees of depolarization depending on the duration of exposure and the depth of the situation of the fibres. The superficial fibres were depolarized more easily. In 50% of the exposures (65 observations) there was repetitive discharge during the exposure time (1 min) and the fibres irreversibly depolarized from the resting membrane values of 85.5 ± 1.8 mV to 54.5 ± 1.5 mV. In 20% of the fibres the repetitive discharge ensued immediately after switching off the light and the fibres were irreversibly depolarized. Repolarization was observed in those fibres that did not attain threshold firing level. After a number of exposures the muscle went into a state of contracture and the fibres were difficult to impale. The phenomenon was reproducible in curarized preparations ($1 \cdot 10^{-4}$ g/ml of *d*-tubocurarine). In choline Ringer solution and under lack of oxygen the photodynamic effect was not demonstrable.

Zusammenfassung. Kurarisierte und nichtkurarisierte Frosch-Sartorii zeigten einen veränderlichen Grad der Depolarisation, wenn sie mit Bengalrot (1:25000) gefärbt und durch scharf zentriertes Licht einer 300-Watt-Tungsten-Lampe beleuchtet wurden. Die Depolarisation wurde niemals im äusseren Natrium und ohne Sauerstoff beobachtet.

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The Study of the Cysteine Content of the Chicken Erythrocyte Histone by Polarography

The cysteine content of the histones is not well known; only some contradictory information is available[1-7]. Since the polarographic method is a sensitive test for the –SH groups[8], we studied the polarographic activity of the chicken erythrocyte histones.

Experimental. The chicken erythrocyte nuclei were isolated with a citrate method[9] and were homogenized in saline-EDTA solution[10]. The whole histone (WH) from the washed homogenate was extracted with 0.25 *N* HCl and precipitated with 6 vol acetone. The precipitate was washed and dried *in vacuo*. The WH was fractionated by using a carboxymethyl-cellulose column with the method of Johns et al.[11]. The protein content of fractions was followed turbidimetrically[5]. The polarographic assay was carried out according to our previous report[12].

Results. As previously indicated[13] the WH was polarographically inactive at a concentration of 10 µg/ml. However, a mild catalytic wave appeared when the WH concentration was increased in the polarographic test-solution. The height of the wave reached its maximum at a concentration of 100 µg/ml (Figure 2, curves a–d). The height of the protein waves decreased with the increase of histone concentrations, until estimation of the polarographic curve became impossible (Figure 2, curves e–g).

The WH chromatographed on a carboxymethyl-cellulose column gave four fractions, similar to thymus histone[11] and to a tumour histone[6]. From these four fractions only one histone (F3) proved to be active polarographically (Figures 1 and 2, curves i–k).

Discussion. The polarographic analysis of the chicken erythrocyte histones showed in all cases that one of them contains –SH groups. This consideration was substantiated by the following facts: that the observed catalytic waves of the WH and F3 fraction are characteristic protein waves, and that the oxidized histones gave no cata-

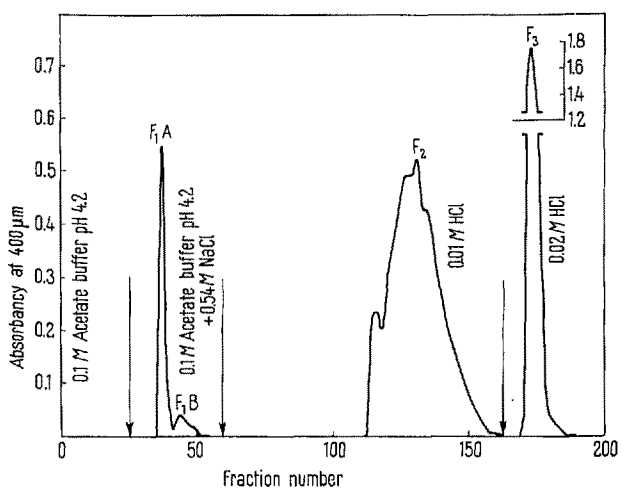

Fig. 1. Elution pattern of chicken erythrocyte histones (117 mg) on a carboxymethyl-cellulose column (14 × 2.3 cm).

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lytic waves (Figure 2, curve h). From the height of the F3 fraction wave it may be deduced that it contains a slight amount of cysteine.

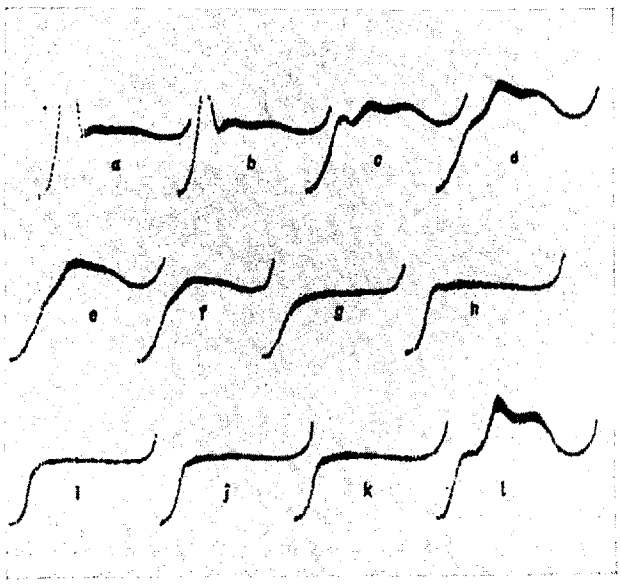

Fig. 2. Polarographic curves of the chicken erythrocyte histones. Curves a–g: WH (20, 30, 50, 100, 150, 200 and 2500 μg/ml); curve h: WH oxidized with performic acid (100 μg/ml); curve i: F_1A histone (60 μg/ml); curve j: F_1B histone (50 μg/ml); curve k: F_2 histone (58 μg/ml); curve l: F_3 histone (45 μg/ml).

The decrease, deformation or disappearance of the catalytic waves of histones at increased protein concentration (Figure 2, curves e–h) may be explained by the aggregation of the histone molecules at basic pH[14]. Therefore, formation of Co^{3+}-protein complexes causing the polarographic activity is met with spatial difficulties. This phenomenon may explain the fact that polarographically HAMER[15] did not find any cysteine in calf thymus histone.

Zusammenfassung. Das Erythrocytenhiston des Hühnchens zeigt eine polarographische Aktivität, die für das Vorhandensein von SH-Gruppen spricht. Extrahiertes Gesamthiston wurde an einer Carboxymethylcellulosesäule chromatographiert. Von den erhaltenen 4 Fraktionen enthielt nur eine (F3) polarographisch nachweisbares Cystein.

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Formate Oxidation in *Saccharomyces cerevisiae*

Oxidation of formate to carbon dioxide is a significant pathway in a variety of animal and plant tissues. Evidence has been presented by CHANCE[1] and others[2,3] that in animal tissues the peroxide-catalase complex is responsible for this oxidation. In the case of microorganisms, diverse mechanisms, such as a true oxidase in *Aspergillus niger*[4] and cytochrome-specific dehydrogenase in *Escherichia coli*[5] and *Nitrobacter agilis*[6], have been shown to be operative.

A *de novo* synthesis of catalase on aeration of anaerobically grown cells of *Saccharomyces cerevisiae* has been reported earlier[7,8]. Formate oxidation in such a system has now been studied to examine the possibility of a catalase-dependent reaction in this organism.

A locally isolated strain of *Saccharomyces cerevisiae* was grown on a medium consisting of glucose 2%, Bactopeptone 1%, yeast extract 0.2% for 20 h at 30°C. Anaerobic condition was ensured by using 500 ml conical flasks filled to the neck with the medium. The cells were then harvested by centrifugation and were suspended in a fresh non-growth medium of composition glucose 1%, potassium dihydrogen phosphate 1%, magnesium sulphate 0.01% and calcium chloride 0.01%. This suspension was divided into 10 ml portions and was aerated in 50 ml conical flasks by shaking in a reciprocating shaker for different periods, after which the cells were separated, washed with distilled water and used for the assay of catalase activity and formate oxidation. Catalase activity was estimated by a modified titanium colour reaction method[9] and is expressed as *Katf*, where

$$Katf = (\ln X_0/X_t)/et \cdot 10^2$$

in which X_0 and X_t are the residual hydrogen peroxide at '0' and 't' minutes and 'e' the concentration of the enzyme source. The extent of formate oxidation was followed by incubating the cells in standard Warburg respirometer vessels at 37°C for 15 min with Krebs-Ringer phosphate buffer pH 7.4, 0.1 M and formate-C^{14} ($1.4 \cdot 10^4$ cpm, Radiochemical Centre, Amersham, England) to a total volume of 3 ml. The respired carbon

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