SUMMARY

1. Wool, silk and nylon, but not cotton, sorb hydrogen peroxide from aqueous solution and the concentration range 0–45 % has been studied. The sorption has not reached its limit at 45 %, but stronger solutions could not be studied.

2. With wool the sorption is independent of the pH of the peroxide solution in the range

pH 2.5–9, but decreases at both lower and higher pH values.

3. It is concluded that the hydrogen peroxide is held by both the amino and carbimino groups and that the sorption process resembles the formation of complexes with simple organic nitrogen-containing compounds such as urea.

The authors wish to thank Mr R. H. Reed for experimental assistance and the directors of Messrs Wolsey Ltd. for permission to publish this paper.

REFERENCES

- Alexander, P. & Earland, C. (1950). *Textile Res. J.* **20**, 296.
 Alexander, P. & Whewell, C. S. (1946). *B.I.O.S. Report* 1472, p. 91. London: H.M. Stationery Office.
 Astbury, W. T. (1942). *J. chem. Soc.* p. 337.
 Elöd, E. & Zahn, H. (1940). *Kolloidzshr.* **93**, 50.
 Elöd, E. & Zahn, H. (1942). *Melliand Textilber.* **23**, 313.
 Hailwood, A. J. & Horrobin, S. (1946). *Trans. Faraday Soc.* **42B**, 84.
 Harris, M. & Smith, A. L. (1936*a*). *Bur. Stand. J. Res., Wash.*, **16**, 301.
 Harris, M. & Smith, A. L. (1936*b*). *Bur. Stand. J. Res., Wash.*, **17**, 557.
 Harris, M. & Smith, A. L. (1937*a*). *Bur. Stand. J. Res., Wash.*, **18**, 623.
 Harris, M. & Smith, A. L. (1937*b*). *Bur. Stand. J. Res., Wash.*, **19**, 81.
 Krepelka, J. H. & Buksa, R. (1937). *Chem. Listy*, **31**, 447.
 Lu, C.-S., Hughes, E. W. & Giguère, P. A. (1941). *J. Amer. chem. Soc.* **63**, 1507.
 Maass, O. & Matheson, G. L. (1929). *J. Amer. chem. Soc.* **51**, 674.
 Machu, W. (1937). *Wasserstoffperoxyd und die Perverbindungen*. Vienna: Springer.
 Rutherford, H. A. & Harris, M. (1938). *Bur. Stand. J. Res., Wash.*, **20**, 559.
 Tanatar, S. (1901). *Z. anorg. Chem.* **28**, 225.
 Theis, E. R. (1946). *Trans. Faraday Soc.* **42B**, 244.

Acid-soluble Pigments of Molluscan Shells

5. IDENTITY OF SOME SUBSIDIARY FRACTIONS DERIVED FROM *PINCTADA VULGARIS*

By A. COMFORT

Department of Physiology, London Hospital Medical College, London, E. 1

(Received 28 March 1950)

Reference was made in a previous paper (Comfort, 1949) to the violet and blue subsidiary fractions which accompany uroporphyrin in the shell of the pearl oyster *Pinctada vulgaris*. A fuller examination of these substances, isolated by the methods already described, shows that they exhibit reactions characteristic of biladienes.

EXPERIMENTAL

The pigments were obtained as by-products of a large-scale extraction of uroporphyrin I from 5 kg. of broken shell, which yielded about 150 mg. of uroporphyrin, purified as its methyl ester. The blue and violet fractions (3 and 3*a*) from talc chromatograms were pooled, dried, rechromatographed on talc from N-hydrochloric acid, and the pigments eluted with acetone.

The main subsidiary pigment (3) is violet in acid solution, and is separated with difficulty from a sky-

blue fraction (3*a*). Both these fractions are green in alkaline solution, with a strong red fluorescence in ultraviolet light. The spectrophotometric curve of pigment 3 is given in Fig. 1. The alkaline spectrum is greatly intensified by the addition of methanolic zinc acetate, but without displacement of the bands. Pigment 3 is converted to a red material ($\lambda_{\max.} = 494 \text{ m}\mu$.) by concentrated nitric acid. Reduction by zinc dust in N-hydrochloric acid gives a yellow solution with strong blue fluorescence.

Pigment 3 cannot be extracted from aqueous solution at any pH by chloroform, and is very slightly extracted by ether at pH 7.5. Pigment 3*a* appears to be slightly more soluble in ether. Both pigments are soluble in acetone and in methanol.

A portion of pigment 3 was dried in a desiccator and esterified with a saturated solution of hydrogen chloride in methanol for 2 hr. at room temperature. Chloroform (1 ml.) was added, followed by water to

separate the phases. The ester was extracted by the chloroform. Its greenish neutral solution showed a red fluorescence, similar to that of the alkaline, free pigment and reversibly quenched by acidification.

Fig. 1. Spectral absorption of violet pigment of *Pinctada vulgaris* (Beckman spectrophotometer). ●—● *Pinctada* '3' fraction (N-HCl); ×—× *Pinctada* '3' fraction methyl ester (neutral CHCl_3).

The spectral curve is given in Fig. 1. Both 3 and 3a pass slowly into amyl alcohol from solution in N-hydrochloric acid.

DISCUSSION

The behaviour and properties of the violet fraction conform closely to those of a biladiene. The red, porphyrin-like fluorescence is exhibited by the alkaline, free pigment, by its ester in neutral and alkaline solution, and by the zinc complex. The acid absorption maximum of the free and esterified pigment at 560 mμ. corresponds with that of meso-biliviolin methyl ester shaken with 2N-hydrochloric acid in chloroform solution ($\lambda_{\text{max}} = 560 \text{ m}\mu.$; Siedel, 1935), although the other bands described by Siedel

(diffuse, between 631 and 621–594, max. 607 mμ.) are not present. Both the zinc complex and the ester hydrochloride of mesobiliviolin exhibit red fluorescence (Siedel, 1935; Watson, 1933), the zinc complex showing bands at 625 and 575 mμ. in methanol (Lemberg & Legge, 1950) compared with 628, 595 mμ. for our pigment 3. The behaviour of pigment 3a closely resembles that of 3, except that it is unstable in N-hydrochloric acid solution at room temperature. The amount of this pigment, even from 5 kg. of shell, was too small for further study.

The red fluorescence of the combined violet and blue fraction on talc columns is variable, and fails to appear in the presence of strong acids, but can be strong enough near neutrality to be mistaken for an additional porphyrin band (Comfort, 1949). A small peak at 410 mμ. in all spectrophotometric tracings derived from these fractions may indicate an admixture of porphyrin, but this could not be separated on chromatographing again, and the fluorescence of the total pigment in a sample of material not exceeding 5 mg. total weight has been shown to equal that of several mg. of free porphyrin. The red fluorescence is therefore undoubtedly proper to the non-porphyrin pigments themselves, and resembles that of aplysiopurpurin, another molluscan pigment which has been shown to contain a mixture of biladienes (Lederer & Hutterer, 1939).

SUMMARY

1. The violet pigment fractions extracted from 5 kg. of *Pinctada vulgaris* shell consist of at least two components.
2. The spectral and chemical properties of these fractions conform to those of the known biladienes of the biliviolin series.

Acknowledgement is due to Prof. C. Rimington, of University College Hospital Medical School, and to Dr E. R. Holiday, of the Medical Research Council Spectrographic Unit, London Hospital, for their kind co-operation in this study.

REFERENCES

- Comfort, A. (1949). *Biochem. J.* **45**, 199.
 Lederer, E. (1940). *Biol. Rev.* **15**, 273.
 Lederer, E. & Hutterer, C. (1939). Unpublished. Cited by Lederer, E. (1940).
 Lemberg, R. & Legge, J. W. (1950). *Hematin Compounds and Bile Pigments*. New York: Interscience.
 Siedel, W. (1935). *Hoppe-Seyl. Z.* **237**, 9.
 Watson, C. J. (1933). *Hoppe-Seyl. Z.* **221**, 145.