

A Further Note on Nile Blue

BY

A. J. CAIN

(From the Department of Zoology and Comparative Anatomy, Oxford)

IN a previous paper (Cain, 1947) it was shown that Nile blue can be used to distinguish between acidic and non-acidic lipoids, and that lecithin stained very deep blue with a 1 per cent. solution, while oleic acid was only faintly coloured. Since lecithin stains, it was suggested that other lipines might stain blue also and that the oleic acid used might be impure.

A sample of phosphorus-free oleic acid was dried over anhydrous sodium sulphate for three months. One portion was then shaken with 1 per cent. Nile blue aqueous solution, and solid Nile blue was added to another. The acid in contact with the solution coloured blue immediately. The other portion showed only traces of blue (appearing red by transmitted light because of fluorescence) after 2 hours. After 12 hours it had darkened considerably, but did not reach equality with the first portion for 24 hours. Since the dried acid takes up Nile blue so much more slowly than that in contact with water, it appears that the blue-staining of oleic acid is connected with its power of imbibing water, and as it is highly unlikely that dry oleic acid will be met with in animal tissues, no use can be made of blue-staining with 1 per cent. Nile blue to differentiate between lipines and fatty acids.

A sample of pure galactolipine (phrenosin and kersin) which gave a negative reaction with the acid haematein test was found to stain deep blue with 1 per cent. Nile blue. The substance was attached to a coverslip by gentle warming and then cooling. Kaufmann and Lehmann (1926) obtained a negative result with pure phrenosin and with pure kersin, probably because, as with lecithin, their method of impregnating pith was not suitable. Nile blue, therefore, cannot be used to distinguish between various members of the lipines.

REFERENCES

- CAIN, A. J., 1947. *Quart. J. micr. Sci.*, **88**, 383.
KAUFMANN, C., and LEHMANN, E., 1926. *Virchows Arch.*, **261**, 623.