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Trump B F, Smuckler E A & Benditt E P. A method for staining epoxy sections for light microscopy. *J. Ultrastruct. Res.* **5**:343-8, 1961.
[Department of Pathology. University of Washington, Seattle, WA]

A technique for staining sections of osmium-fixed epoxy-embedded tissues without prior removal of embedment for light microscopy using aqueous toluidine blue at alkaline pH was presented. When sections are stained in this manner, their images under the light microscope are striking due to their great definition and resemblance to electron micrographs They are useful, therefore, not only for the identification and mapping of areas seen in electron microscopy, but also because they permit better utilization of the full resolving power of the light microscope [The *SCI*® indicates that this paper has been cited in over 820 publications since 1961]

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"The stage was set in Kansas City, Missouri, in 1944, when my father and I were home together with chicken pox for two weeks. I had had microscopes since about the fourth grade; my main purpose was to study cells and tissues or, indeed, anything that I could see. It didn't matter that I hadn't acquired a commercial microtome because my father was my personal purchasing agent, mechanic, technician, and general supervisor who constructed the microtome and helped me shoot the rabbits and squirrels and catch the fish (the favorite experimental animals in those days) and then admired the results that I obtained.

"I think I was born destined to stain things to look at in a microscope but, in any case, I have had a lifelong love affair with microscopes that is not only amusing and soul-fulfilling but also useful in supporting my family and my research group.

"I readily accepted the advice of Robert E. Stowell in 1957 when he said that the foreseeable future in histopathology required expertise in electron microscopy (EM). So I set out for the University of Washington in August of 1959 to the laboratories of H.S. Bennett. Those times were exciting for EM since J. Luft was developing new epoxy resin embedding techniques.[1-2] This resulted in many solutions but also caused problems as epoxy sections were difficult to stain for light microscopy (LM). The only available technique for LM visualization was phase microscopy. This problem remained unsolved, and the need to stain epoxy sections for LM became acute, especially in pathology.

"We had the usual labs available, among them a histology-histochemistry lab which was always bustling with activity. One night, I was determined to solve this problem of staining epoxy sections before going to sleep. I cut a large number of semi-thin sections and proceeded to stain such with every stain-filled Coplin jar in the lab. By 7 a.m., the results were clear, amazing, and gratifying. I observed the same contrast in organelles as one could see in the electron microscope. This immediately extended the range of magnifications possible and gave images in photomicrographs which correlated well with those in low power electron microscopes. I could scarcely wait until Bennett and Benditt arrived. However, in analyzing my notes, I realized that the one Coplin jar which had worked was unlabeled and, thus, I had absolutely no idea what the responsible dye might be. Much to my delight, Smuckler identified the Coplin jar as one containing an alkaline solution of toluidine blue. I demonstrated the results to Bennett and Benditt and proceeded to write the paper, which was my first, including, of course, Smuckler and Benditt as coauthors. The paper was accepted immediately and has since been cited often because the technique was so useful.

"Since that time, of course, many variances of this technique and many additional techniques have been developed."[3]

1. **Luft J H. Improvements in epoxy resin embedding methods.** ***J.*** *Biophys. Biochem. Cytol.* **9:**409-14, 1961.
2. ..... Citation Classic. Commentary on *J. Biophys. Biochem. Cytol.* **9**:409-14. 1961. *Current Contents* (20):8. 16 May 1977.
3. **Burns W A.** Thick sections: techniques and applications. (Trump B F & Jones R T, eds.) *Diagnostic electron microscopy.* New York: Wiley, 1978. Vol. 1. p. 141-66.