254 CD
DOI: 10.1017/S1431927606061460
Microsc Microanal 12(Supp 2), 2006

A Comparative study of two procedures of tissue processing for TEM: with and without the use of propylene oxide

Sultan Al-Siyabi*, Issa S. Al-Amri** and Taher Ba-Omar***

*Department of Physiology and ** Department of Pathology, EM Unit, College of Medicine, Sultan Qaboos University, P.O.Box 35 Muscat 123, Sultanate of Oman.
***Department of Biology, College of Science, Sultan Qaboos University, P.O.Box 36 Muscat 123, Sultanate of Oman.

It is a common practice among electron microscopists to pass tissue fragments through a transitional solvent and pure propylene oxide (PO) for a few minutes immediately prior to infiltration[1]. PO is well known for its hazardous effects reported in technicians and researchers working with electron microscope.

The present study was carried out to explore the possibility of excluding the use of PO as a transitional solvent in processing of both hard and soft tissues for transmission electron microscopy (TEM).

Two different experimental techniques were used for preservation of kidney and skeletal muscle tissues with minor changes from the living state. Freshly dissected kidney and skeletal muscle tissues of a White albino (WKY) rat were fixed in Karnovsky's for 2.5h at 4^{o}C, postfixed in 1% osmium tetroxide for an hour and dehydrated in graded acetone. Tissues were prepared following two different protocols. In the first, tissues fragments were passed through a transitional solvent, propylene oxide (PO) and were infiltrated with various proportion of propylene oxide and epoxy resin. In the second, the use of transitional solvent was eliminated and tissue samples were infiltrated with various proportions of acetone and epoxy resin. The specimens were embedded in epoxy resin and polymerised over night at 60^{o}C. Ultrathin sections were cut and stained with uranyl acetate and lead citrate. Sections were examined under JEOL electron microscope JEM-1230.

Electron micrographs of the kidney tissue and the skeletal muscle revealed very well preserved irrespective of the use of PO. The glomerular basement membrane, the capillary endothelial cells, mesangial cells and podocytes were shown to be well preserved(**Fig. 1.a-d**). Similarly, electron micrographs of the skeletal muscle showed very good preservation of the striations, myofilaments and the organelles including mitochondria and sarcoplasmic reticulum (**Fig.2.a,b**).

Some previous studies have suggested the elimination of the use of PO as a transitional step especially for soft biological tissues [2]. In light of our results, we suggest that the use of PO may also not be essential for processing of hard biological tissues for transmission electron microscopy.

References

[1] Hayat M A.(2000), Principle and Techniques of Electron Microscopy Biological Application, 4th Ed. Cambridge university Press.
[2]. Mascorro J A. (2004). Propylene oxide: to use or not to use in biological tissue processing. Microscopy Today 12: 45.

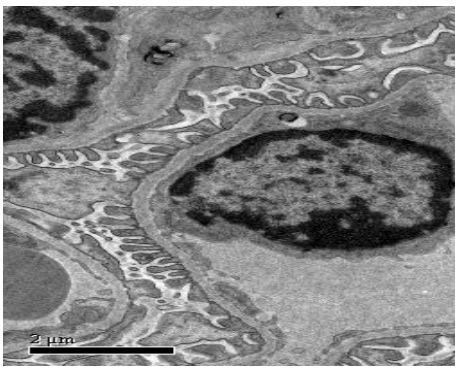

Fig 1.a

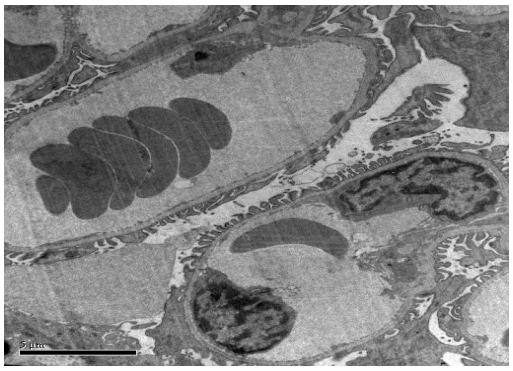

Fig 1.b

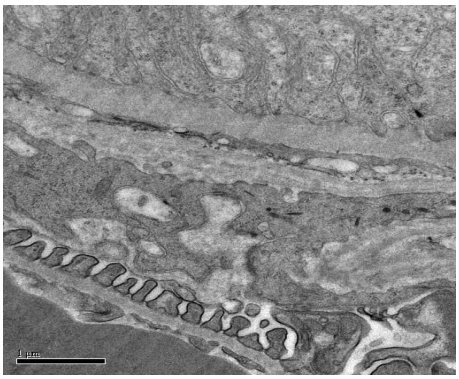

Fig. 1.c

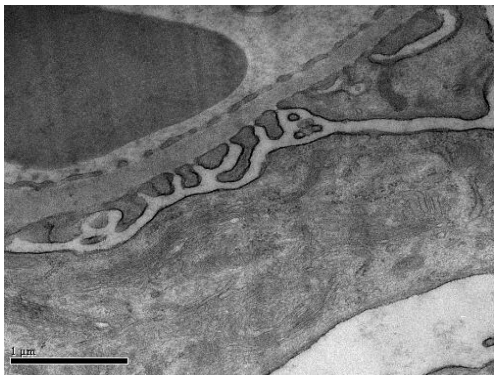

Fig. 1.d

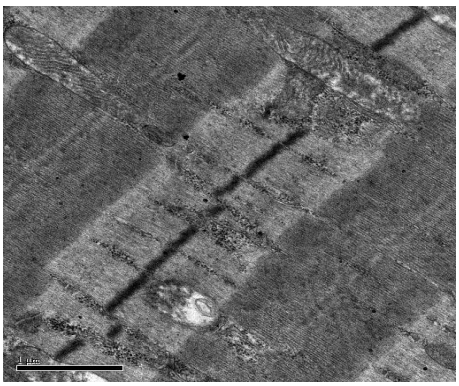

Fig. 2.a

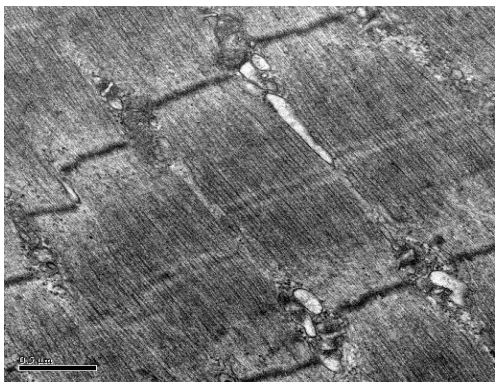

Fig. 2.b

Fig.1.a - d. Electron micrograph of the kidney tissue (glomerulus) processed without PO as in (a & c) and with PO as in (b & d). Scale bar =2µm (a) and 5 µm for (b) and 1 µm for both(c) and (d).

Fig. 2.a,b.Electron micrograph of the skeletal muscle processed without PO as in (a) and with PO as in (b). Scale bar=1 µm for (a) and 0.5 µm for (b).

https://doi.org/10.1017/S1431927606061460 Published online by Cambridge University Press