The main purpose of fixation is:
1) to cross-link cellular structures into a matrix so as to preserve the structure of the cells with the minimum of alteration from the living state (i.e. with no changes in morphology, volume, or spatial relationships, and with minimum loss of cellular constituents) and,
2) to protect and stabilize cellular structures from changes during subsequent treatments and from irradiation by the electron beam.

Fixation is the first and most important step in any EM study, since mistakes made at this point render the whole project useless. Below are a number of fixation protocols that should provide good starting points for most EM studies.

SAFTEY NOTE: Most of the chemicals used for processing specimens for electron microscopy are extremely hazardous

Glutaraldehyde, formaldehyde and osmium tetroxide are volatile and can fix any cells/tissues they contact (they will fix your respiratory epithelium, corneas, and epithelial cells on your hand, etc): it is therefore essential to use them in a fume hood. Although dangerous these fixatives cannot penetrate more than 1-2 mm into tissues, so exposure to them rarely causes any permanent damage.

The epoxy resins used to embed the specimens are potentially more dangerous than the fixatives. Many of the resin components are known to cause cancer in rats or mice. During the embedding process the resins are dissolved in solvent(s) that can carry the resin into your skin EVEN through plastic gloves. In contrast to fixatives, whose actions are immediate and apparent, the consequences of exposure to the resins are not apparent for years. Therefore be careful with all resins prior to polymerization into hard blocks.

A) Preparation schedule for tissues cut into 1-2 mm thick sheets or <1 mm^3 blocks

Primary fixation
2.5% glutaraldehyde in 100 mM phosphate buffer at pH 7.0 2-24 hours (2-3 preferred)
Fixation is usually started at room (or body) temperature and after 15-30 min then continued at 4°C. Fixation at 4°C slows down autolytic processes and reduces tissue shrinkage. Nevertheless, with some specimens, fixing at 4°C can cause problems. For example, some organelles such as microtubules can be cold labile and may be lost at low temperatures.
(if you cut up the specimen in fixative, place samples in to fresh fixative)

Wash in 200 mM phosphate buffer that has been adjusted to the osmolarity of the sample to prevent tissue damage

Postfixation
1% osmium tetroxide in 100 mM phosphate buffer 1-2 hours at 4°C.

Wash at least 5 times in distilled water (need to remove all excess phosphate ions to prevent uranyl acetate (UA) from being precipitated – same applies for cacodylate or any other buffer)

En bloc stain with 2% aqueous uranyl acetate for ~2 h at 4°C IN DARK (must be carried out in the dark as UA is photo reductive and will precipitate – this you don't want to happen as UA crystals make sectioning difficult and don't look nice in the specimen!).

Dehydrate through a series of ethanols or acetones and propylene oxide.
Embed in epoxy resin.

For reference here is a typical embedding schedule:

Dehydration

30% acetone	15 minutes
50% acetone	15 minutes
70% acetone	15 minutes
90% acetone	15 minutes
100% acetone	3 changes, each of 30 minutes

Resin embedding (Epon mix)

Propylene oxide	2 changes, each of 15 minutes	
(N.B. since propylene oxide is much more volatile than ethanol or acetone be careful not to allow the sample to be exposed to the air as damage will occur due to the rapid evaporation of the solvent)		
Resin infiltration	2:1 mix of propylene oxide:resin	1 h
	1:1 mix of propylene oxide:resin	1 h
	1:2 mix of propylene oxide:resin	1 h
	100% resin	overnight
	Change in to fresh resin	1 h
Embedding	Fresh resin in embedding moulds – remember to label the samples.	
Polymerise	12-24 hours at 60-70°C	

The above protocol uses phosphate buffer (not PBS) as the buffering vehicle for the glutaraldehyde and osmium fixatives. As alternatives cacodylate, PIPES or HEPES buffer can be used instead.
The type of buffer in which the fixatives are made up can affect the appearance of the specimen. However, one advantage of cacodylate and PIPES or HEPES is that $CaCl_2$ and/or $MgCl_2$ can be added to the primary fixative. Calcium (and Mg) ions reduce the extraction of cellular components and also enhance the retention of phospholipids.

B) Fixation protocol for cells in suspension

There are a number of ways to fix cells grown in suspension.
i) gently pellet the cells (800 g for 5-10 min), remove most of the culture medium and then fix the cells by adding an excess volume (5-10 times the cell volume) of fixative and re-suspend the cells in the fixative. After –10 min the cells should be pelleted in an eppendorf tube (3-5 min at max rpm), the fixative removed and replaced by fresh fixative.
ii) cells can be fixed by adding 1 volume of 25% glutaraldehyde to 9 volumes of cell suspension in the original culture medium. After 5-10 min the cells can be pelleted and fixation continued in the original fixative. Alternatively, after pelleting, the original fixative can be replaced by a 'standard' glutaraldehyde fixative and the fixation process continued for the required time.

After the initial fixation step the sample(s) can be further processed as described above. The cells do not need to be re-suspended during the subsequent steps and if treated carefully the pellet can be freed from the tube and 'flat embedded' allowing sections to be cut through which ever part of the pellet is required.

C) Fixation protocol for cells grown in culture dishes

Primary fixation
After removal of most of the culture medium, the cells should be fixed with an aldehyde-based fixative as described in protocol A.

After the initial fixation the cells can be further processed by a number of different ways depending on the aim(s) of the experiment.
i) the cells can be released from the plastic by gentle scraping and then pelleted. The samples can then be post-fixed and processed as normal.

ii) the samples can be post-fixed with osmium tetroxide (and uranyl acetate if required) and then dehydrate through a series of ethanols and embedded in situ.
N.B. In this case, since most plastics dissolve in acetone and propylene oxide the samples must be dehydrate using ethanol and a series of resin-ethanol mixes used during the infiltration process (instead of the more usual resin-propylene oxide mixes).

iii) the samples can be further processed and dehydrated using ethanol as described above. However, after fully dehydrating the samples, the cells can be released from the plastic using propylene oxide. By gentle pipetting propylene oxide over the cells they will detach from the plastic either as individual cells or in sheets. The detached cells should be pelleted, washed several times with propylene oxide to remove and solubilized plastic, and then embedded as described above.

D) Fixation protocol for pathology samples

Fix tissues in a mixture of 2.5% glutaraldehyde, 2% (para)formaldehyde in 100 mM cacodylate buffer (pH 7.0) with 2 mM $CaCl_2$.
After an initial ~30 min fixation, cut the specimens into small (~1 mm^3) pieces and continue fixation in fresh fixative for 16-24 h at 4°C.
Wash briefly with 200 mM cacodylate buffer (pH 7.0).
Post-fix with 1% osmium tetroxide in 100 mM cacodylate buffer (pH 7.0); 2 h at 4°C.
Wash with excess distilled water [to remove any free cacodylate and/or phosphate ions].
En bloc stain with 2.0% aqueous uranyl acetate, ~2 h at 4°C (in dark).
Dehydrate with acetone (or ethanol), propylene oxide and embed in resin as outlined above.

General references for TEM

Glauert, A.M., Lewis, P.R. (1998). Biological Specimen Preparation for Transmission Electron Microscopy. Practical Methods in Electron Microscopy: vol. 17. This tells you all about the fixatives, buffers etc. and also covers dehydration and embedding.

Reid, N., Beesley, J.E. (1991). Sectioning and Cryosectioning for Electron Microscopy. Practical Methods in Electron Microscopy: vol. 13.
This book covers section cutting for both conventional resin sections as well as cryosections, although the Griffiths book (see below) is the best for cryosectioning and cryo-immunocytochemistry.

Lewis, P.R., Knight, D.P. (1977). Staining Methods for Sectioned Material. Practical Methods in Electron Microscopy: vol. 5.
This is a useful reference for section staining etc. You can get the actual references for the lead citrate and uranyl acetate staining protocols from here – if you want to quote the original papers.

Griffiths, G. (1993). Fine Structure Immunocytochemistry. Springer-Verlag.
Although this book is mainly about immunocytochemistry (and especially the techniques of cryosectioning) there are useful chapters on the chemistry of aldehyde fixatives.

All EM supplies are from Agar Scientific or TAAB Laboratories – these are 2 of the main suppliers of EM materials in the UK.