Conservation techniques for specimens preserved in fluid.

(Notes to accompany Fluid Preservation Course)

Simon Moore, August 2025.

Preparing glass for mounting specimens

Glass requires cutting to a correct size to fit either as a flat lid for a battery jar, a circular lid for a flanged and cylindrical jar or as a backing plate upon which specimens may be mounted. For older battery jars the manufacturing process results in the width at the bottom of the jar being slightly narrower than at the top. These jars also have a slightly thinner and evenly-flat side for displaying the specimen whereas the back surface of the jar is normally as it came out of the mould and will have a slightly ridged surface which can distort viewing a specimen if the specimen is placed back to front.

Tools and materials

For the successful mounting of biological specimens in glass display (battery) jars the following items will be required.

Glass preparation tools

Glass-cutting protective gloves. Always check a sheet of 3 to 4mm glass for sharp edges and run a fine grit paper over any suspected sharp edges prior to handling.

Eye protection, laboratory protective spectacles.

Oil-filled glass cutters (Toyo is a good make), bicycle oil is good to fill them (oil acts as a flux and prevents glass micro-splinters from flying off the glass during scoring).

Cutter for making glass circles – essential if using cylindrical flanged jars that require a flat and circular lid.

Plastic glass-breaking / running pliers, don't get the metal-jawed versions with plastic covers as the covers rupture after a short time.

For grinding – 10mm thick glass lapping plate, 120 carborundum grit, glycerine water and pipette.

Glass drill bits - diamond dust burrs are ideal: Silverline 722878 Rotary Tool Diamond Burr Set 30pce 3.17 mm Mandrel. https://amzn.eu/d/9dDjYfB

Drilling flux - 20g of camphor dissolved in 100 ml of turpentine but 50% aqueous glycerine also works well.

Specimen adhesives

• Collodion / pyroxylin and gelatine sealant:

Collodion: 8% nitrocellulose (pyroxylin) in ether-alcohol other names are pyroxylin, diluted to 2-4% with the solvent below. This should be stored in a refrigerator but monitor it to ensure that the solution stays clear or it may need further dilution with the solvent.

Collodion solvent mixture comprises di-ethyl ether [tlv 1200] and absolute ethanol or methanol (50:50/wv). This mixture is highly flammable and should be stored in a refrigerator.

Gelatine sealant: leaf gelatine (pale or blonde), glycerine, glacial acetic acid (ref. pp 4-5).

Specimen mounting tools

Curved sewing/ embroidery needles – small and medium sizes.

Scissors.

Preparation and mounting of specimens

For large or firm specimens

Arrange the specimen on a backing plate of 3 to 4 mm thick soda glass. The glass will require marking with attachment points (using a diamond scriber/pen) just behind the specimen, to denote the position of the holes to be drilled.

Check distribution of the specimen's weight to avoid placing too much strain on one particular area - heavier areas will require greater support (use circles or cut shapes of clear plastic / acetate sheet) or the filament may scar or even tear through the specimen.

Dry the backing plate and then drill the attachment holes using the scribed marks to locate the exact position for the drill bit. If the backing plate is thicker (< 5mm), a hole may have to be drilled from each side. Whilst cutting, the drill handle should be gently rotated to allow the point enough room for penetration. Always keep the drill bit moist with flux, which will also prevent a powdered glass hazard, and use very slight pressure only. When the hole is complete, a slight click will be heard or felt. Drilling should be briefly continued with the drill vertical to remove sharp inner edges from either side of the hole.

Using a curved and long needle, pass some nylon monofilament through the areas of the specimen to be attached. For more fragile tissues, a long hollow needle (hypodermic) can be used to pierce the hole first and then the monofilament passed through the tissue. If the needle has a clearing wire through it, pierce the tissue with this inside the needle to prevent it from clogging or becoming blocked.

Turn the backing plate over onto the specimen passing the monofilament through the drilled holes and tie double knots that are firm but not so tight as to cut into the tissue. Then trim off surplus filament ends to about 2mm.

Use the collodion technique for alcohol-preserved or melted gelatine for formalinpreserved specimens to stick soft areas of tissue to the backing plate.

Supporting the specimen with a gloved hand, lower it into the jar and leave overnight to rid itself of any trapped air pockets formed during ligaturing. Check for any faults or problems, and then seal the jar.

Hollow specimens

For specimens such as a fish swim-bladder or alimentary tract, fill out the corners by injecting with 2% collodion or dilute (50%) gelatine for formalin-preserved specimens, using glass or metal syringes (no rubber or plastic fittings). Inject the hollow interior with alcohol or 5% formalin and leave to harden. Note that the injection needle must be pre-warmed to prevent gelatine from gelling whilst inside the needle. A needle with a teat bulb attached is better and will avoid a syringe becoming clogged with gelatine!

Small or fragile specimens

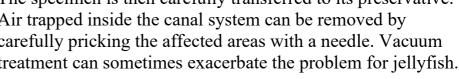
Use the collodion technique. This has been carried out on salmon eggs that have remained attached to glass for over fifteen years. The successful adhesion of a perfect sphere to a flat surface shows the efficacy of this technique. Internal labels can also be attached to specimens using the collodion technique and it can be most useful to repair damage to a fragile specimen or to reinforce it. Molten gelatine (also used as a jar sealant) is used for mounting small specimens instead of collodion and is gelled by

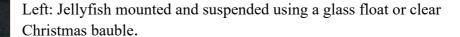
contact with formalin.



These can be supported by a small, centrally pierced circle of clear acetate sheet placed centrally under the bell and inside the stomach. A piece of monofilament knotted at the lower end is then passed upwards through the medusa's stomach and umbrella (for example) using a needle so that the acetate disc sits centrally under the umbrella or the specimen may tilt slightly in the preservative.

The specimen is then carefully transferred to its preservative. Air trapped inside the canal system can be removed by carefully pricking the affected areas with a needle. Vacuum treatment can sometimes exacerbate the problem for jellyfish.





Adhesives for mounting specimens in fluid preservatives.

Collodion technique

The following method is recommended for attaching alcohol-preserved specimens to glass:

- 1. Arrange specimen on a glass plate and drain off excess alcohol.
- 2. Moisten attachment points (or edges of specimen) with ether-alcohol mixture.
- 3. Pipette small amounts of 2-4% Collodion onto attachment points and leave to gel. Do not allow to dry out moisten with ether-alcohol if necessary; avoid breathing on Collodion as condensation will react with it forming an opaque film.
- 4. When Collodion surface has gelled (after about 5-10 minutes it takes on a crinkled appearance), slowly immerse the plate and specimen into alcohol. Leave for c. 10 minutes for the Collodion to harden. The Collodion should be completely transparent; if not, re-dissolve with solvent and allow to re-gel.
- 5. Attach any parts of the specimen trailing in fluid.
- 6. Leave the specimen in alcohol overnight (at least) to check the bond is firm before sealing the jar.

Gelatine technique

Follow the same technique as above bearing in mind, that this is used for aqueous preservatives such as 5% formalin or glycol mixtures, and using molten gelatine sealant that is sufficiently mobile for easy application and so that it can adhere to the specimen's tissues.

This has also been successful when mounting alizarin transparency specimens onto glass plates.

Sealing glass display/battery jars

The following processes should lead to the successful sealing of glass lids to display jars (battery jars): the gelatine method has been tried and tested and a well-sealed jar can remain sealed for over twenty years. Other sealants such as bitumen and Stockholm tar with lead sesquioxide are both hazardous and messy; paraffin wax is ideal for short-term sealing and the lids are easier to remove than those sealed with gelatine. Alternatively, Dow Corning silicones (available from Merck) provide a good seal for jars although these products have only been tested for up to fifteen years and not over the longer term.

Lid grinding is essential, otherwise the sealant has no key for attachment. Pour about 5 ml of water and glycerine on to a sheet of scrap plate-glass and sprinkle on some medium grit (20 grade) carborundum powder. Grind away any sharp edges to the lid and then press it on to the grit and glycerine mixture and move the lid around on this mixture in a circular motion until the glass surface has been sufficiently ground. Wash and dry the lid.

Jar grinding

For a cylindrical, flanged or a battery jar with a flat lid, the sealing surface can become damaged with small chips or spalls (tiny pits in the ground glass surface). These can be ground out by the following technique which will also ensure that the surface remains level.

- 1. Using the lapping plate add some 30% aqueous glycerine and 120 carborundum grit (the grade should have sufficient sharpness for the job and neither be too coarse (it will be nearly impossible to use) or too fine (such as a polishing grade).
- 2. Invert the jar and gently rotate it, using both hands, on the grinding mix. It will be quite a noisy process.
- 3. After about 50 turns, remove the adherent grinding mixture from the jar surface and replace it onto the lapping plate with a spatula, then wipe away any further adherent grit before rinsing it under the tap. The grit is heavy and will block any plumbing system quite quickly.
- 4. Check process the spalls, chip marks become smaller and will eventually disappear leaving a clean ground glass surface, ideal for sealing.

Gelatine sealing technique

This is a tried and tested method of unknown origin but the recipe for the sealant was discovered in an index museum day-book dating from the mid-nineteenth century at The Natural History Museum, London. The technique for its use has since been improved by Moore (1980).

Preparation of sealant

The sealant is made up from gelatine *coignets* or leaves - wafer-thin sheets that are used in preparing food. Commercial powdered and fibrous gelatine, although cheaper, are unsuitable as they do not melt below 100°C following hydration.

Weigh out 22 g of gelatine sheet and leave to hydrate in tap water. The leaves should be adequately hydrated in three minutes; thicker sheet gelatine will take longer (maybe overnight). The gelatine will feel loose and floppy when hydrated.

Melt the hydrated gelatine in a beaker in a water-bath or *bain-marie* (gelatine will char if brought into direct contact with heat). If the molten gelatine is as mobile as water, it will have absorbed too much water during hydration - leave it for a further twenty to forty minutes (preferably using a magnetic stirrer) to evaporate the surplus water.

Using fume removal facilities take the gelatine from the heat and stir in 6 ml of glycerol and 3 ml of glacial acetic acid. Pour the mixture into a metal tray and leave to set for 30 to 40 minutes.

Cut it into squares and store in an air-tight Le Parfait or other bail jar with a crystal of menthol as a mould inhibitor.

Sealing a jar with gelatine

- 1. With the specimen in the jar, fill the jar only three-quarters full with IMS (alcohol) preservative.
- 2. Ensure that the lid has a filler hole drilled in one corner not too near the edge, and that areas in contact with sealant have been ground to give some key the top edge of the jar and lower edge of the lid. These should be clean, grease-free and dry. Old and dried gelatine can be scraped off with a scalpel or rehydrated for easier removal.
- 3. Melt some gelatine sealant in a hot water-jacketed beaker and heat the lid in a tray of hot water to about 50°C. Ensure the lid is kept wet.
- 4. Brush sealant fairly generously on to the outer edges of the jar. Press/grind down the preheated lid (drained only) on to the applied gelatine seal with the filler hole at the front.
- 5. Heat and moisture from the lid should re-melt the gelatine if it has cooled and gelled; brush in further gelatine if any gaps appear.
- 6. Apply weights to the lid overnight.
- 7. Inspect for white streamers of excess gelatine which may have run down into the alcohol. If present, the jar will have to be cleaned and then resealed using less sealant.
- 8. To remove lid, invert the jar into a shallow, hot water-bath and leave until the sealant has hydrated and swelled. Take care that the lid does not fall off! If appearance is satisfactory, leave for a further 24 hours until sealant takes on the frosted appearance of the underlying ground glass. If the sealant is opaque white, then some alcohol has affected it (the jar may be too full of alcohol) and the jar should be resealed.
- 9. If the jar seal looks good, top up the level of alcohol in the jar to c. 2 mm below the lid, using a syringe, through the filling hole. If the jar is filled to the brim, the seal will be put under pressure in warmer weather.
- 10. Larger jars can be filled using a separating funnel clamped on to a stand, and with a piece of rubber tubing connected to a needle wired on to the end of the tubing. This apparatus can be left unattended while the jar is filled although the jar should be placed in an alcohol-proof dish in case of accidental overflow.
- 11. If the jar becomes overfilled, remove surplus alcohol using a syringe.
- 12. Insert a piece of polypropylene rod into the filler hole and trim it flush with a sharp blade.
- To prepare polypropylene rod bungs, this must be done away from a smoke detector as it may trigger fire alarms! Rotate some 5mm rod just beyond a gas flame until it becomes clear (slightly melted). It can then be slowly stretched whilst still rotating it over the heat. Once the rod turns opaque as it cools, it cannot be stretched further unless re-heated. The rod itself will burn gently if held too close to the source flame. Blow it out if it catches fire and disperse resultant smoke.
- 10. Seal the hole with a blob of molten gelatine and add a 5mm circular microslide coverslip pressed down onto the gelatine.

11. If many jars are to be plugged and time is short, a circle of the flowable-fluid silicone around the filler hole and a 5mm microslide coverslip will work just as well but will be more difficult to remove.

Other sealants

Silicone sealing

Flowable-fluid silicone is suitable for glass jars containing formalin, alcohol, phenoxetol-based preservatives and glycerol-acetate mixtures, although Horie (pers. comm.) states that formalin will eventually break down a silicone seal but so far, this sealant has lasted for over 30 years. Permatex flowable silicone (car windscreen) glass sealant is available from Amazon in 42g tubes or Dow Corning 3140 RTV.

- 1. Apply to rim of jar (not too generously or it will flow down inside the jar) using a small-bladed spatula.
- 2. Smear around the seal to ensure an even application and then apply the lid. Press down and the sealant will join up all the way round.
- 3. Check that the sealant has not started to flow down the jar sides, or has any gaps in the seal. If so, remove the lid quickly, wipe away in excess with strong paper towelling or add some more to fill any gaps and re-apply the lid pressing down and twisting slightly.
- 4. The seal will take about 6 hours to set but overnight is better.
- 5. This provides a really effective seal but is difficult to reverse, even with silicone release agents but is slowly reversible in hazardous organo-chlorine solvents such as tri-chloromethane or dimethyl chloride. Be sure to do this work on a reverse flow bench that draws the fumes backwards and/or wear breathing apparatus as these solvents are powerfully anaesthetic and are toxic.
- 6. Alternatively, carefully push a thin-bladed (47) Tiranti spatula against the seal until a weaker spot is found, continue to push the head of the spatula into the seal and do not prise it up or you may crack the glass lid and break or bend the spatula blade. Once inserted, the spatula may be moved carefully from side to side, cutting into the seal until the lid detaches. A spatula is better than a scalpel as the latter tends to break easily.
- 7. Despite this slight problem this silicone has been found to be a very effective sealant. Other more viscous silicones have been tried but none has the ease of use and permanence of the flowable silicone.

Older sealants

Gutta percha

This technique was first devised by Schorr in 1907 and has been improved over the years. Although it is considered to be old-fashioned, some curators still use it by tradition in preference to the even older gelatine technique.

- 1. Apply cement evenly to the edge of the jar using a spatula.
- 2. Warm the lid in hot water, dry it and apply it to the cemented edge of the jar.
- 3. Apply weights and leave for three days to a week for cement to harden.
- 4. Trim off excess cement using a pre-warmed spatula.

Stockholm tar and red lead

This technique can be used in conjunction with a pig's bladder. The sealant has a life of five to ten years, becoming increasingly brittle.

- 1. Pour Stockholm tar into a shallow dish and mix in lead sesquioxide (this is a toxic health hazard and usage should be checked under local health and safety regulations) until the mixture forms a sticky paste, firm enough not to flow.
- 2. Apply cement evenly to the edge of the jar using a spatula.
- 3. Warm the lid in hot water, dry it and apply it to the cemented edge of the jar.
- 4. Apply weights and leave for three days to a week for cement to harden.
- 5. Trim off excess cement using a pre-warmed or hot spatula.

The conservation unit at the Royal College of Surgeons of England have recently completed a short training video entitled 'Core principles of fluid preservation: Routine maintenance of the specimen', which can be viewed at: http://youtu.be/Oe48q7B6IoY.

This project has been generously funded by the John Ellerman Foundation and the Hunterian Museum Trustees.

Pig's bladder technique

The pig's bladder technique considerably extends the life of a jar seal and gives the jar a better and antique-look finish. It is possibly the oldest and most traditional way of sealing a museum jar but usually requires some experience before a successful result is achieved. Pigs' bladders can be obtained from slaughterhouses.

- 1. Cut the pig's bladder open and pin it out in a dissecting tray; scrape away as much fatty tissue as possible then immerse it in a formalin-based fixative. Cover the container and place it into a fume cupboard.
- 2. After fixation, unpin and remove the bladder and wash it in running water until formalin is undetectable.
- 3. Stretch the bladder over the top of the jar and attach the corners of the bladder to the side of the jar using adhesive tape. Experienced curators fold the corners in just below the jar rim to give a thicker bed for ligaturing.
- 4. Bind the bladder to the jar just below the rim using several turns of fine twine and tie it off.
- 5. Apply some varnish or button polish to the twine to prevent it from working loose. Allow the bladder to dry.
- 5a. If the resulting seal fails then remove the bladder and rehydrate it using Decon-90 or 2% sodium orthophosphate; go back to stage 1.
- 6. When completely dried, remove the tape and trim corners off the bladder just below the twine.
- 7. The bladder can be painted black paint or traditional, using bitumen dissolved (20%) in toluene (carried out in a fume cupboard), if desired. If it shows signs of becoming brittle some lanolin cream, gently applied, can slightly extend the life of the bladder and improve its appearance.

References

Moore, S.J. (1980) Problems with glass museum jars solved. *Biology Curators' Group Newsletter*, **2**, 384-389.

Moore, S. J. 1999: Fluid Preservation (pp. 92-132). In: Care and conservation of Natural History Collections. Eds Carter, D. J. & Walker, A. Butterworth Heinemann, Oxford.