

The vessel segments are then taken to a clean but not sterile dissecting table and with sharp dissection all extraneous tissue is removed. The vessel is next washed with tap water to be sure that no postmortem clots cling to the intima. The vessel is examined for evidence of damage or atherosclerosis and if none is found it is ready for sterilization.

Sterilization

For the past two years tissue sterilization has been carried out with specially purified Beta-Propiolactone,* a method described by Szylagi and associates.¹ This has been a very satisfactory method and cultures following sterilization have been uniformly sterile.

The chemical solutions necessary to carry out this phase of the program are:

1. Beta-Propiolactone.* This is an extremely unstable chemical and must be handled with care.

2. Buffered saline solution. This is made unsterilely as follows:

16.8 gm. sodium bicarbonate
8.5 gm. sodium chloride
5.0 ml. 1% phenol red

This solution is made up to 1 liter, is unsterile and is stable for several months.

3. Molar phosphate buffer which is made as follows:

0.2 M anhydrous dibasic sodium phosphate (28.396 g/liter)
0.2 M anhydrous monobasic potassium phosphate (27.218 g/liter)
808 ml. of sodium phosphate is added to 192 ml. of the potassium phosphate

pH is taken on a Beckman meter and adjusted to 7.4.

Place 700 ml. in a vacuum seal pyrex bottle and autoclave for 45 min. at 15-18 lb. pressure at 120° C.

Steps in Sterilization

1. Because the BPL is so unstable it is necessary to dilute the concentrated solution in ice cold water. With a pipette 2.2 cc. BPL is added to 25 cc. of ice cold distilled water in a small flask.

2. Place 225 cc. of buffered saline solution into an autoclaved wide-mouth jar such as the ethicon suture jar. This solution has a phenol red indicator and should be a reddish pink.

3. Pour the diluted BPL into the buffered saline stirring gently. The solution should not change color at this time.

4. Place the grafts into the solution and loosely apply the cap. One graft to each solution is recommended; however, this depends on tissue mass. If the grafts are small, we have processed two in one such solution.

*Furnished by B. F. Goodrich Chemical Co., Rose Bldg., 2060 E. Ninth Street, Cleveland 15, O.

5. The jar is next placed in a water bath for two hours at 37° C. During the two hours, the solution is gently agitated and care must be taken to see that the grafts are completely covered by solution. At the end of this two-hour period the solution will have assumed a salmon pink color.

Buffer Washing and Packaging

This step must be done under the strictest aseptic conditions. A table is prepared with a sterile pack containing a small saline basin, sterile scissors and long forceps. Two or three large pyrex test tubes (a good size is a 1½ inch diameter and 8 inch length) with rubber stoppers to match, plus two or three sterile rubber condoms, are added to this equipment. The operator next puts on sterile rubber gloves and with the long forceps places the grafts in the saline basin. The graft is then washed two or three times with the 0.2 molar phosphate buffer solution. A small piece of the vessel is then snipped away and placed in a thioglycolate culture tube for culture. The vessel is now placed with the long forceps into the large pyrex test tube and the rubber stopper applied. Starting at the capped end the rubber condom is rolled on the tube and fixed in position with two or three encircling rubber bands. All subsequent steps may now be done without aseptic precautions.

Quick-Freezing

Three or four pounds of dry ice are pulverized and placed in a flat pan. The pan we have used is 8 x 12 x 2 inches. It is advantageous but not essential to have this insulated. Seventy per cent ethyl alcohol is gradually added to the dry ice dust. This mixture is constantly stirred until it forms a viscous liquid, with a temperature of minus 70 to minus 74° C.

The prepared stoppered tubes with enclosed grafts are placed in the alcohol-dry ice mixture. The pyrex tubes are rotated gently until well embedded in the solution and the flat pan is covered with towels for insulation. This quick-freezing process continues for 20 minutes.

The tubes are then removed, labeled and placed in a standard deep freeze refrigerator which has a temperature of minus 30° C. The tag on the graft tube contains all the essential information about the configuration, size, source, date of procurement and condition of the graft. This information and records concerning the operative use of the grafts are kept in a vessel registry for purposes of long range follow-up. The vessel registry form is shown in Figure 1.

DE
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BLOOD VESSEL GRAFT
DONOR: Name

Age

Cause of Death

Date of Death

No. of hours

No. of hours

No. of hours

Name of doctor

Name of doctor

Culture report

GRAFT: Diagram

Measurement

RECIPIENT: Name

Age

Date of operation

Place of operation

Diagnosis

Surgeon's name

FOLLOW UP: (As registered in
Laboratory)

1. Discharge from hospital
2. Three months postoperative
3. One year postoperative

Fig

DEPARTMENT OF SURGERY
UNIVERSITY OF MIAMI SCHOOL OF MEDICINE

BLOOD VESSEL GRAFT No.

DONOR: Name Record No.....

Age Sex..... Color.....

Cause of Death

Date of Death.....

No. of hours between death and autopsy.....

No. of hours between autopsy and procurement.....

No. of hours between procurement and processing

Name of doctor procuring graft.....

Name of doctor processing graft.....

Culture report (to be attached by doctor who processed graft)

GRAFT: Diagram

Measurements: lengths

internal diameters (both ends)

branches and major intercostals

RECIPIENT: Name Record No.....

Age Sex..... Color.....

Date of use of graft.....

Place of surgery.....

Diagnosis (reason for graft)

Surgeon's report: Copy of operative record

Diagram, length, branches, etc.

Culture report (to be attached by
surgeon)

FOLLOW UP: (As regard success of graft. Presence of pulsations?
Laboratory studies? Arteriograms?)

1. Discharge from hospital.
2. Three months postop.
3. One year postop.

Figure 1. The vessel registry form.

The Use of the Grafts at Surgery

After an adequate exploration has been accomplished at the operating table, a suitable graft is selected and removed from the freezing compartment. In a few minutes it will have thawed enough to allow it to be rolled around in the pyrex tube. Under aseptic conditions, the rubber covering and rubber stopper are removed, and the graft is placed in a sterile saline basin on a sterile side table. The basin contains 150 cc. of normal saline, 1 gram streptomycin and 1,000,000 units of aqueous penicillin. After a few minutes the graft assumes the supple characteristics of a normal artery.

All branches of the graft are ligated with 000 silk or, if they have been cut flush with the vessel, they are sutured with 5.0 arterial silk. The vessel is then occluded at one end and the saline solution is injected under mild pressure from the other end to test the graft for leakage. The graft is now ready for use. Since all grafts are of necessity tailored to fit a particular situation, the portion of the blood vessel not used is sent to the bacteriology laboratory for culture.

Comments

The grafts preserved in this manner have been easy to work with and in our experience have been entirely satisfactory.² Grafts processed in this manner we believe to be satisfactory at least one year and probably two years after the initial freezing.

If equipment and personnel common to most hospitals are used, the cost will be negligible. Refrigerators, water baths, and the like are already in use and can be utilized without inconvenience or extra expense. Chemical solutions, bacteriologic studies and sterile supplies can be obtained at little expense from the appropriate hospital department.

References

1. SZYLAGI, D. E., OVERHULSE, P. R., SHONNARD, C. P., and LOGRIPPO, G. H. "Sterilization of Human Arterial Homografts with Beta-Propiolactone," *Surg. Forum* 4: 244, 1954.
2. COOKE, FRANCIS N. "Blood Vessel Replacement Therapy," *Journal of the Florida Medical Association*. To be published.

Sumario en Español

Los recientes avances en la necesidad de establecer en todos los hospitales en aquellos donde el costo y caro no es satisfactorio de un personal habitual e insignificante. Refri- den utilizar sin ma- micas, estudios bac- poco costo de los di- Metodos de obten- detalle.

Sumario en Español

Los recientes avances en cirugía cardiovascular hacen enfática la necesidad de establecer y mantener un banco de vasos sanguíneos en todos los hospitales donde se resuelven problemas cardiovasculares o en aquellos donde se contempla hacerlo en el futuro. Equipo complicado y caro no es un requisito necesario para el funcionamiento satisfactorio de un banco de vasos sanguíneos. Si se usa equipo y personal habitual a la gran mayoría de los hospitales el costo será insignificante. Refrigeradores, equipo de laboratorio ya en uso se pueden utilizar sin mayor inconveniente o gastos extra. Soluciones químicas, estudios bacteriológicos y equipos estériles pueden obtenerse a poco costo de los distintos departamentos hospitalarios apropiados.

Metodos de obtención, esterilización y preservación se explican en detalle.