A Semi-automatic Needle for Implanting Tissues into Experimental Animals

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The method employed by most laboratories for the implantation of tissue fragments into experimental animals requires the loading of a cannula (usually a spinal needle) prior to each injection. Moistness of the tissue, gauge of the needle, creation of sufficient vacuum in the lumen upon retraction of the stylet, size and shape of the fragment to be loaded, and manipulation of the fragment at the tip of the needle are factors that contribute to the time-consuming task of loading a conventional cannula. When large numbers of animals are to be injected rapidly, it is helpful to have an assistant for the loading of the cannulas. With each loading there is increased danger of contamination.

We have designed a modified spinal needle to facilitate successive implants of solid tissue fragments under aseptic conditions and without the need of an assistant.

The device consists essentially of three parts (Fig. 1): (A) a spinal needle, (B) spring-controlled stylet, and (C) a tissue-holding chamber with a micrometer-like ram. All materials selected for construction of the apparatus are noncorrosive and nontoxic to tissue.

Two modifications are made of an 18-gauge (other sizes may be used) spinal needle. The tip of the cannula is cut off to give an over-all length of 40 mm.; the tip is filed smooth and round to minimize trauma. At a point 3 mm. from the cannula-hub junction, a hole 0.5 mm. in diameter is bored through the wall of the cannula.

The spring-controlled stylet is housed in a 2-ml. metal tip Luer hypodermic syringe. The handle of the stylet is ground off to compensate for the presence of the stylet head and the spring in the barrel (over-all length of the plunger, 58 mm.). The diameter of the plunger is reduced to eliminate friction between plunger and barrel. To prevent the plunger from being forced out of the barrel by the compressed spring, a stainless steel chain is used to connect a wire around the neck of the plunger with the spring holder around the barrel.

The tissue-holding chamber with micrometer-like ram consists of three parts (Fig. 1): (C) a tissue-holding chamber, (D) a sleeve, and (E) a ram. A hole 3.5 mm. in diameter is bored through one end of a solid brass rod (28 mm. long and 5 mm. in diameter) for a distance of 26 mm. The exterior of this end of the rod is threaded for a distance of 20 mm. (28 threads/2.5 cm.). Two millimeters from the opposite end of the rod a hole 1 mm. in diameter is bored through the diameter of the rod.

The sleeve, constructed of stainless steel tubing (22 mm. long, 7 mm. outside diameter, and 5 mm. inside diameter of the chamber) is threaded internally, one end threading over the external threads of the tissue-holding chamber and the opposite end receiving the threads of the ram. The external surface of the end of the sleeve threading over the tissue-holding chamber is scored at four equidistant points of its circumference. The scores act as markers for determining the amount of rotation of the sleeve required to force tissue into the lumen of the needle.

The diameter of the head of the stainless steel ram is sufficiently tight-fitting to the wall of the tissue-holding chamber to prevent fragments of tissue from becoming lodged between them (diameter of the head of the ram, 5.4 mm., over-all length 33 mm., with 3 mm. as the handle and 3 mm. for threading into the sleeve). The ram is detachable from the sleeve to permit cleaning.

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Fig. 1.—(A) Modified spinal needle; (B) spring-controlled stylet housed in a hypodermic syringe; (C) tissue-holding chamber; (D) sleeve; and (E) ram.
The tip of the modified spinal needle is inserted through the hole drilled through the diameter of the blind end of the tissue-holding chamber. The hole in the cannula is aligned with the center of the tissue-holding chamber, and the two pieces are joined from the outside with silver solder. The ram is threaded into one end of the sleeve, and the opposite end of the sleeve is threaded over the tissue-holding chamber.

The spring is inserted into the syringe barrel; the tip of the stylet is inserted through the long axis of the spring and through the tip end of the barrel. The plunger is inserted into the barrel, and the two parts are connected by means of a chain. The assembled tissue-holding chamber and the spring-controlled stylet are wrapped separately and sterilized in an autoclave.

The tissue to be implanted is minced with sterile scissors and packed into the mouth of the tissue-holding chamber. (A vibrating pipette shaker was found to produce the vibration necessary to pack the tissue fragments into the chamber and to prevent air bubbles from remaining between the fragments.) The head of the rod is inserted into the mouth of the tissue-holding chamber, and the sleeve is threaded over the chamber to force tissue into the lumen of the needle. The desired size of the tissue fragment to be implanted is determined by the amount of clockwise turning of the sleeve over the tissue-holding chamber. Upon compression of the plunger of the spring-controlled stylet, the tip of the stylet forces tissue into the lumen to the tip of the needle.

For efficient use of the instrument the operator must: (a) release the plunger to permit the tip of the stylet to return to rest behind the tissue-holding chamber-cannula junction and (b) load the instrument by forcing tissue into the lumen of the needle before attempting to implant into an animal.

Since the tissue to be implanted is enclosed in a sterile container there is no danger of contamination of the tissue source during implantation into a large number of animals.

Fifty to sixty animals can be implanted in an hour without the necessity of reloading the instrument. Tissue fragments implanted into the anterior chambers of eyes can be observed to leave the instrument. For subcutaneous implants the operator may reassure himself of the presence of the tissue in the lumen by first forcing the fragment to the tip of the needle. Release of the plunger will result in return of the fragment into the lumen.

In this laboratory the instrument has been used to implant mouse Sarcoma 37 and mouse tumor E 0771 into the anterior chambers of the eyes of 256 mice and into the subcutaneous tissues of the flanks of 480 additional animals. ABC and C57BL strains were used; 98 per cent of the implants were successful.

**SUMMARY**

A device for the rapid implantation of solid tissue fragments under aseptic conditions is described. One individual can operate the instrument and do 60 implants an hour. The percentage of successful implants is equivalent to that obtained by loading a separate cannula for each injection.

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