

## METHOD FOR CUTTING LAMELLAR GRAFTS IN FROZEN CORNEA. NEW ORIENTATION FOR REFRACTIVE SURGERY

(Preliminary Report)

BY

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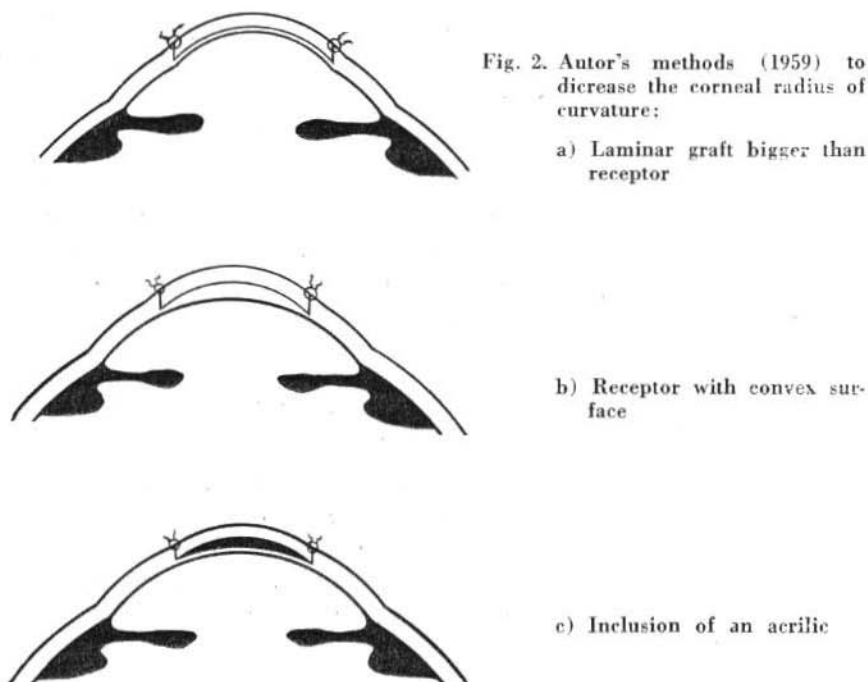
Although attempts to use lamellar transplants were made in the early period of development of keratoplasty (Walther-1840)<sup>1</sup> they were very soon given up in favor of penetrating keratoplasty which, by that time, was producing better results and which continued to do so until 1948, when Paufique and Sourdille, with the advent of the antibiotics, proved the benignity of the lamellar method.<sup>2</sup> This characteristic has become more pronounced since the entrance of the corticosteroids into therapeutic use.

However, the visual results of anterior lamellar keratoplasty are frequently less satisfactory than those obtained in favorable cases by penetrating keratoplasty, a fact which is due to one or more of the following causes:

- 1) Residual opacities in the deep plane.
- 2) Opacity in the plane of junction.
- 3) Defects of refraction.

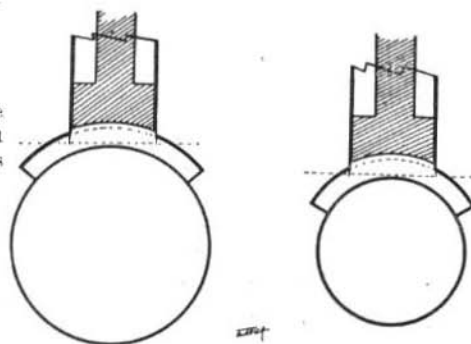
Defects of refraction, which are those dealt with in this article, are caused by irregularities:

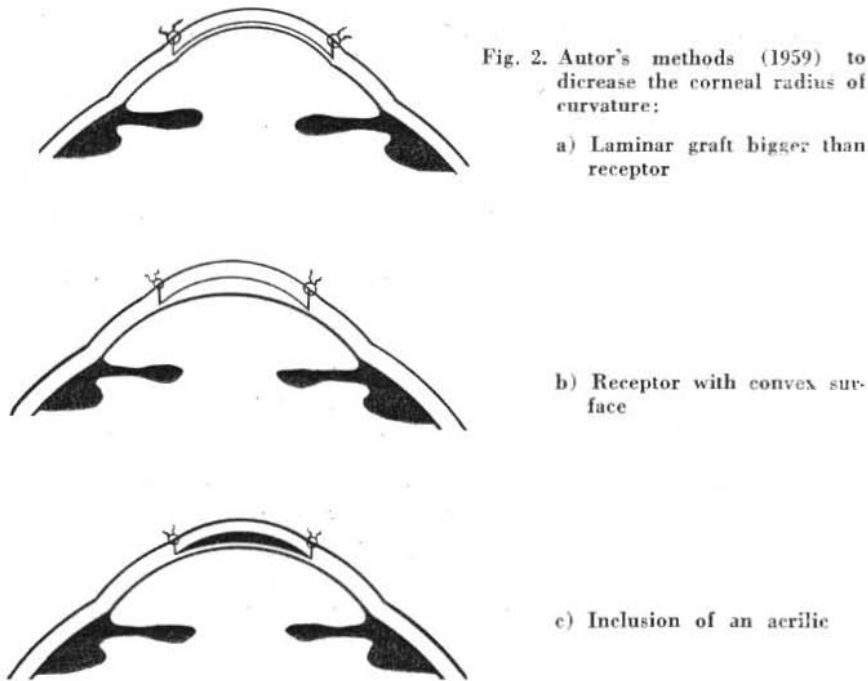
- a) In the cutting of the recipient bed.
- b) In the cutting of the graft.



The lack of a technical procedure for cutting the recipient bed in the desired form led us to consider the possibility of modifying the form of the graft. By so doing we succeeded in minimizing pre-existing hypermetropic or myopic defects of refraction in eyes with superficial leukoma, using transplants cut with a knife from a donor cornea, the form of which had previously been modified by fitting it over a metallic sphere of adequate size (Fig. 3). Although, because of imperfections connected with manual cutting, the grafts did not possess sufficient exactitude, the results obtained encouraged us to continue to make experiments along the same road.

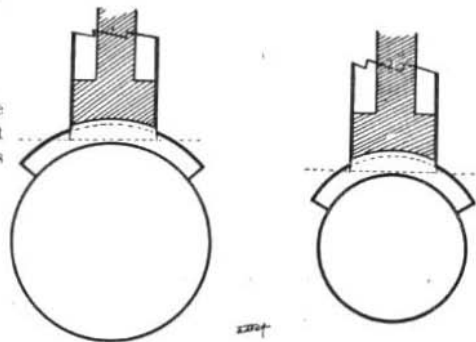
Fig. 3. The cornea placed on the surface of a sphere, is cut horizontally once the graft has been limited.





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Fig. 3. The cornea placed on the surface of a sphere, is cut horizontally once the graft has been limited.



Our experiments were then directed toward the cutting of either the graft or the recipient bed with mechanical aids, similar to those used in the cutting of optic lenses. We tried in vain a large number of methods and of instruments (curved knives, and burrs of metal, emery, carborundum, diamond, etc.).

The discoveries of Eascott<sup>8</sup> in regard to the viability of frozen grafts opened to us a new horizon for our idea of cutting perfect corneal grafts and eventually grafts with a predetermined refractive power, because the hardness of frozen cornea makes it possible to handle it just as any other material in common use, let us say, for instance, plastic material, can be handled.

The method as it has been developed consists in cutting the frozen graft by turning the parenchymatous face of the cornea, in special lathe which enables one to give the graft the proper curvature, thickness, and size with the greatest precision.

With this technique we have been able to obtain grafts that are absolutely uniform and that have a predetermined dioptric value. With these grafts it has not only been possible to minimize or suppress postoperative cylindrical and spherical defects of refraction, or those present before lamellar keratoplasty, but also to carry out lamellar autokeratoplasty, in experimental animals for the purpose of producing, at will, ametropias which can compensate for a pre-existing ametropia.

The method can even be adapted to another use, which at present is in the course of experimentation, namely, the cutting of lenses of corneal tissue of precise form and size which, when included in the thickness of the cornea, will produce modification of its refractive power. (Fig. 4).



Fig. 4. Interlamellar inclusion of lens of "corneal parenquima", to increase or decrease the corneal radius curvature.



This article, being a preliminary report, does not allow us to give a minute description of technical details; in fact, such a description would be premature at the present time. We will describe only the essential steps of the technic as it was

followed in our experiments, and we will give examples of the results obtained, which have prompted us to publish this report.

The complexity of the method does not constitute an obstacle to its use. Everything is complicated in the beginning. In cases of homokeratoplasty, and this is the only procedure in which, up to the present time, we have used this technic on human beings, all the technical work can be done in the eye bank by a specialist who will be able, by using this technic, to deliver to the eye surgeon a graft perfectly adapted to the specifications requested by him.

With corneas cut according to this technic, two of the aforementioned causes of tectonic irregularity in lamellar keratoplasty can be diminished, because:

1) It makes it possible to obtain grafts of perfect uniformity in thickness and in radius of curvature, which radius can be calculated so as to compensate for a pre-existing ametropia.

2) It makes it possible to compensate for deformations of the recipient cornea by means of astigmatic grafts.

3) Irregularities due to defects of coaptation are minimized in lamellar keratoplasty either by the use of edge-to-edge sutures or by the use of a contact lens.

4) The method is not capable of modifying irregularities due to a defective cutting of the recipient bed, irregularities that can be diminished, at present, only by scrupulously careful dissection under the control of the surgical microscope with the slit lamp.<sup>11</sup>

## EXPERIMENTAL WORK

In evolving this method we studied successively the following possibilities:

a) The possibility of freezing the recipient cornea for the purpose of cutting it in the desired form.

b) The viability of lamellar grafts frozen after cutting.

c) The viability of lamellar grafts cut during freezing.

We did not investigate the viability of grafts cut from globes previously preserved at 79° Centigrade below zero and defrosted before the cutting of the graft, because that point has already been demonstrated by the results of the experiments carried on by Eascott, Cross, Leigh, North, Rycroft and others.

### a) *Freezing of the recipient.*

Experiments carried out in rabbits showed us that corneal lesions caused by freezing of the cornea produced by means of the direct application of a little rod of carbon dioxide snow upon the cornea, or by insertion through the concave

surface of a little tube of metal, 7 millimeters in diameter, filled with carbon dioxide snow are reversible (Fig. 5).

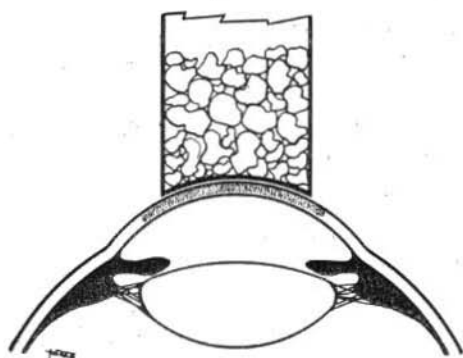


Fig. 5. Freezing of cornea, applying a tube full of carbonic snow.

Freezing for more than ten seconds caused, in all cases, irreversible lesions of the iris, (atrophy and paralytic mydriasis) and produced in most cases, opacity in the anterior capsule of the crystalline lens.

This time (ten seconds) was not sufficient to enable us to cut the recipient bed in the predetermined form, as was our plan.

In the hope of prolonging the available useful time of freezing of the cornea in situ, without producing irreversible lesions in the iris and in the crystalline lens, we attempted to isolate these structures by first filling the anterior chamber with air. By so doing we succeeded in prolonging the useful time of freezing for another three seconds. For the purpose of increasing it even more, we tried circulating air, at a temperature of zero degrees Centigrade, in the anterior chamber (Fig. 6) and with this we succeeded in prolonging the time to 15 seconds. Freezing for more than that length of time produced in all cases irreversible lesions of the iris and of the crystalline lens. Consequently, the idea of cutting the frozen cornea in situ was dismissed and we started new studies on the possibility of cutting it in the isolated graft.

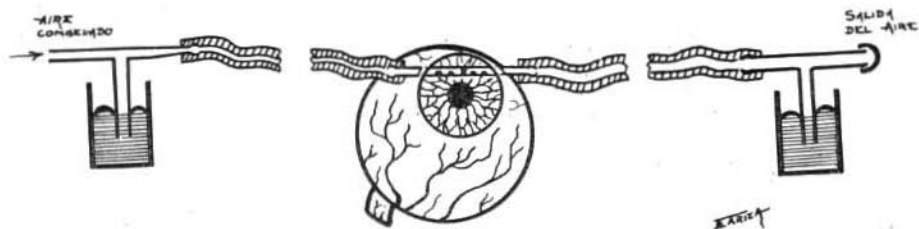


Fig. 6. Diagram showing a desing made to keep the circulation of cold air in the anterior chamber, at a uniform pressure. The containers have mercury.

b) *Investigation of the viability of lamellar grafts frozen after cutting.*

Being aware of the results of experiments carried on by Eascott, Cross, Leigh and North on the role that glycerol plays in protecting the viability of grafts frozen at 79° Centigrade below zero, we started to test their results in isolated cornea, carrying out autoplasties and homoplasties in dogs, pigs, rabbits and guinea pigs.

For the freezing of the graft we used, at first, the platen of the freezing microtome and autoplasic and homoplastic corneas which were obtained from animals killed immediately before the experiments.

One half of these grafts were frozen without any previous preparation, while the rest were submerged in an isotonic saline solution with 15% of glycerol for a period of time which varied between 15 minutes and one hour, before the freezing was started. Freezing was maintained for 5 minutes, a span of time which we considered sufficient for the maneuvers to which we had decided to subject the frozen tissue. Defrosting was accomplished slowly in the ambient temperature. With this technic we did not obtain any case of transparency.

Accordingly, we changed our procedure for freezing by submerging the cornea into an ambient temperature of 70° Centigrade below zero, which was obtained in a small chamber covered with a paste of carbon dioxide snow with alcohol or glycerol (Fig. 7). By this means we obtained 60% of transparent grafts, both in the group of grafts which had been treated with glycerol and also in those which had not previously been treated.

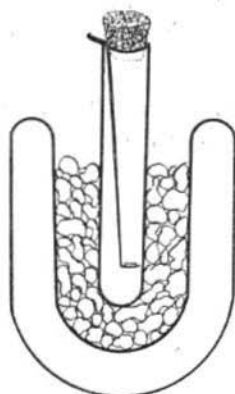


Fig. 7. Freezing chamber for grafts.

Because of the fact that a prolonged stay of the grafts in the glycerol and saline solution caused them to become edematous and of a mucous consistency which

destroyed their usefulness for our purpose optic cutting, we then decided to use exclusively grafts without any protection and we restricted our experiments to the use of eyes of rabbits because of the greater facility with which they can be manipulated. Thus we were able to observe that opacities are due, in the greatest number of cases, to defects in the coaptation of the graft, because sutures are torn prematurely and as a result the graft becomes detached in a small zone through which the tears infiltrate.

By resecting the third eye-lid at the end of the operation and by practicing a tarsorrhaphia, these accidents were diminished. Later on, they were practically eliminated when the experimental animals were treated with antibiotics and with ACTH during the postoperative course.

In a control lot we obtained one hundred per cent of transparencies by using homoplastic corneas which were frozen without any protection, and circular lamellar keratoplasty 6-mm. in diameter and 0.3 mm. in thickness, the grafts being fixed with a continuous edge-to-edge suture.

*c) Investigation of the viability of lamellar grafts cut during freezing.*

Once the viability of homogenous and autogenous lamellar grafts cut according to our customary technique (piriform spatula) and frozen for a period of 5 minutes at 79° Centigrade below zero was confirmed, we proceeded to determine whether the cutting of the graft frozen in a lathe would modify its viability or affect the postoperative course.

The results obtained were identical with those already described in paragraph b), that is, manipulation of the graft in a frozen state does not modify its subsequent behaviour.

The grafts thus obtained (Fig. 8) exhibited a perfectly uniform section and it was impossible to detect, even under the microscope, the slightest irregularity or any trace left by the cutter.



Fig. 8. Grafts without optical power; spherical power positive, and negative.

#### OPTIC CUTTING OF FROZEN GRAFTS

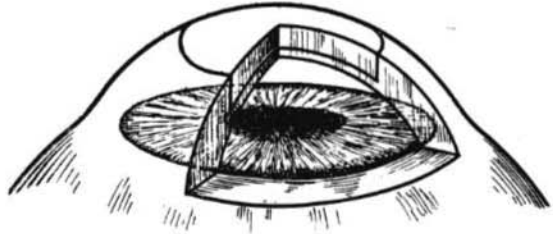
The question was how could be turning the frozen corneas in a lathe, for the purpose of giving them the desired form and thickness. We must briefly con-



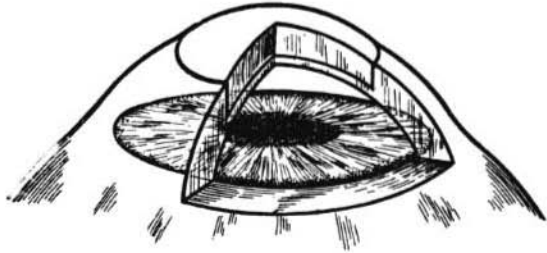
sider at first the characteristics of the lathe used and then the devices employed to fix the cornea so that it can be turning on in the same frozen state for as long as may be necessary.

*Lathe:* In our early experiments we employed a small lathe of the kind used by watch-makers, but provided with a special carriage and a device for reproducing curves (Fig. 10). This carriage and device were constructed for us by the Rendix firm.

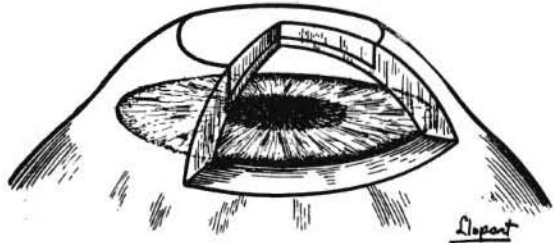
Fig. 9. Anterior Lamellar Keratoplasty with graft:  
a) Without optical power



b) Of positive dioptric value



c) Of negative dioptric value



The radii of curvature were obtained by using spheres of adequate dimensions of the kind commonly used in ball bearings. At the present time we use an industrial lathe with a device for cutting curves which permits us to obtain a precision of 0.01 mm.

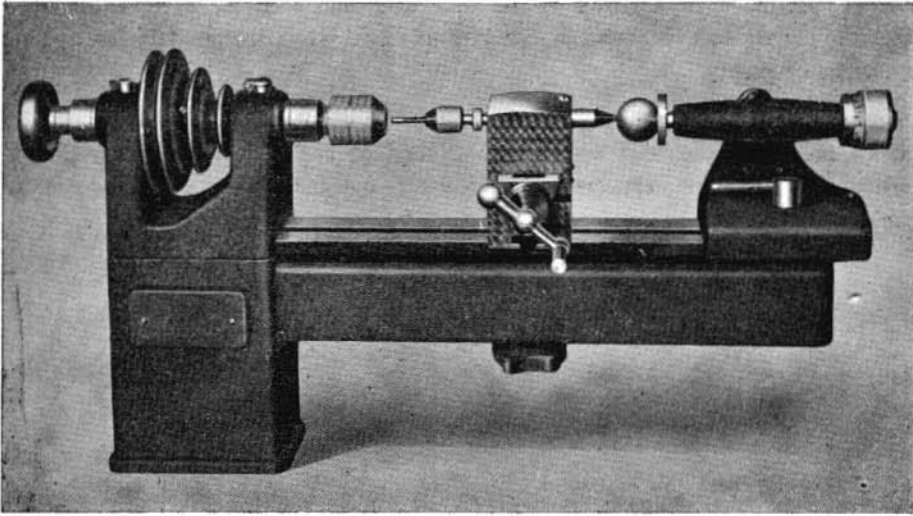


Fig. 10. Jewellers lathe machine, with an special dispositive to reproduce curvs.

*Fixation of the graft.* Fixation of the frozen graft and preservation of the graft in the frozen state presented to us at first many difficulties, which were finally overcome by the construction of a lathe platen with a chamber for carbon dioxide snow (Fig. 11) and a plastic cornea-holder which is fixed to the lathe platen by means of a bayonet device (Fig. 12).

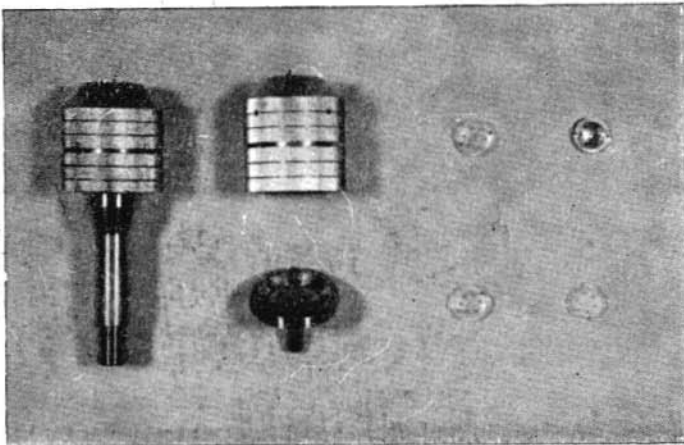


Fig. 11. Two models of plates, with chamber for carbonic snow, and cornea holder.

The modus operandi is as follows:

The plate, without carbon dioxide snow but with the plastic cornea-holder already attached to it, is placed in the spindle of the head where a concavity of an adequate radius of curvature and of the exact diameter of the cornea which is to be worked upon, is cut in the cornea-holder. Next, a ring of perforations is made at about 1 mm. from the periphery of the said concavity. The perforations are about 0.2 mm. each, and they serve to increase the solidity of the fixation (Fig. 13).

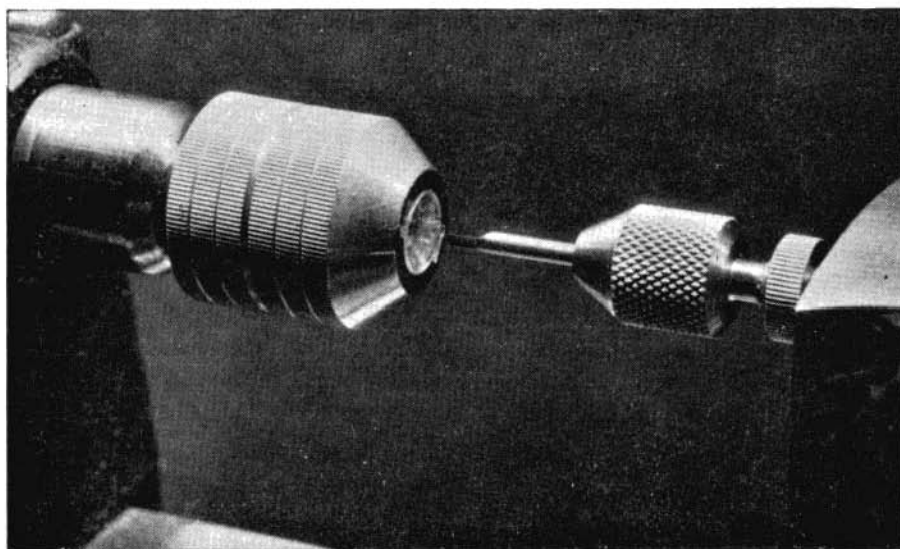


Fig. 12. Plate with corneal-holder placed in lathe.

The cornea-holder is then removed from the plate and sterilized. Then the cornea previously obtained with a trephine of adequate dimensions, is placed with its epithelial surface contacting the sterilized cornea-holder, in such a manner that it remains perfectly centered in the concavity of the plastic. It is then placed in the freezing chamber, where it should remain for 3 or 4 minutes.

In this lapse of time the chamber of the platen is filled with carbon dioxide snow and the lathe is conveniently regulated. The plastic cornea-holder, with the frozen cornea adhering to it is then fixed in the platen and the whole system is placed in the Lathe's spindle head. The graft is then cut from the parenchymatous surface of the cornea, following the curve which has previously been selected. It is advisable to carry out this maneuver in an atmosphere with a low hydrometric degree, in order to prevent the excessive formation of ice on the frozen parts. In some cases

we have been forced to work in a nitrogen atmosphere because we did not have within our reach adequate means for making conveniently dry the ambient atmosphere of our laboratory.

The cutting of the graft in itself does not present any difficulties from a mechanical point of view. The only provisions worth mentioning are the convenience of using a burin with a curved cutting surface, the radius of curvature of which should be approximately 1 mm. less than that of the concavity which is to be cut and the maintaining of the burin at a low temperature as long as the operations lasts. This is easily accomplished by placing a small depository filled with carbon dioxide snow near its point.

The value of the radii of curvature constituted a reason for experimentation to determine which of the two procedures we followed is the most adequate. The second of these two procedures, which we are going to describe, deserves our preference on account of its simplicity.

The first procedure consists of cutting the bed in the cornea-holder with a curve exactly similar to that of the donor cornea and then calculating mathematically what the curve of the parenchymatous surface of the cornea must be for it to adapt itself to the raw surface of the recipient cornea; this gives us at the epithelial level of the graft, the radius of curvature that we wish to obtain.

The second procedure consists in cutting the concavity in the cornea-holder with the radius which we wish to obtain definitively and to which the donor cornea will be adapted because of the elasticity of the cornea and the relatively small difference between the two radii of curvature. The posterior or parenchymatous surface of the cornea should be cut with exactly the same radius of curvature as that possessed by the recipient cornea at the level of the bed. This radius is 0.3 or 0.4 mm. less than the radius of curvature of the recipient cornea at the level of the epithelium, depending on whether the graft has a thickness 0.3 or 0.4 mm. This point should be given great attention because of the fact that 0.3 or 0.4 mm. in the value of the radius is the equivalent of about 2 diopters.

This method has the advantage of not requiring calculations, but it still calls for a large series of experiments to determine whether the adaptation of the graft to the new concave bed permits sufficient exactness.

When it is a question of obtaining optically neutral grafts, —the kind we have used up to the present time in human beings— we cut the recipient bed in the cornea-holder with a radius exactly equal to that of the donor cornea, cutting the parenchymatous surface with the same radius reduced by exactly the thickness of the graft, usually 0.3 mm. so that both surfaces are parallel to one another.

In these cases the corneas were obtained from human globes which had been preserved in liquid paraffin up to the very moment in which the graft was taken from to be frozen and optically cut.

The astigmatic grafts can be obtained by a procedure which differs slightly from the others previously mentioned, and although they may theoretically be secured by either of the same techniques herein described for grafts either optically neutral or with spherical refractive value, we have been able, on account of difficulties of a mechanical character, to try out only the one that is equivalent to the second technique herein described, with some variations.

This technique is as follows: The cornea-holder is cut in a toric form with the degree of astigmatism we wish to obtain. When the cornea is applied to the toric surface, it suffers a deformation by which it becomes elliptical. For this reason it is necessary to utilize a larger graft, in order to cut its parenchymatous surface and then immediately either to change the instrument in the lathe or, still better, to transfer the platen to an auxiliary machine in which the graft is given the form of a cylinder and its diameter is reduced to the proper size.

When it is a question of homoplasties, we have, in all cases cut, with the trephine, a graft of the whole thickness of the cornea, of the desired dimensions, reducing it from its endothelial surface, to the necessary thickness.

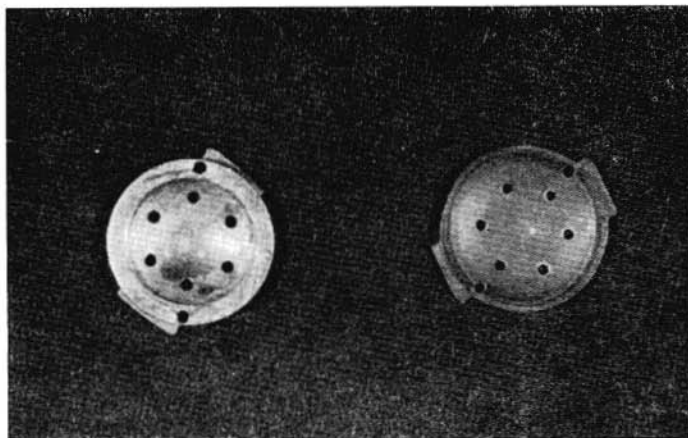


Fig. 13. Corneal-holder:  
a) Metal for 6 mm. grafts  
b) Plastic for 7 mm. grafts.

In cases of autoplasties (experimental up to this date) we have obtained the lamellar graft by our usual technique (piriform spatula) reducing it only as much as necessary in order to obtain the modification needed in the radius of curvature.

Once the graft has been cut we have used one of the three following procedures. Up to the present time we have not been able to decide which of the three is the best. They are as follows:

- a) The graft is defrosted and used immediately.
- b) The graft is left to thaw-out by itself and it is preserved in liquid paraffin at a temperature which may vary between  $2^{\circ}$  and  $4^{\circ}$  C, up to the moment when it is used.
- c) The graft attached to the cornea-holder is kept frozen at  $79^{\circ}$  C. below zero up to the very moment of use.

## RESULTS

As an example we report the case of a rabbit which had a cornea with a radius of curvature of 47 diopters and into which a graft of 37 diopters was inserted. A final result of 38 diopters with perfect transparency of the graft and visibility of the ocular fundus was obtained.

Figure 14 illustrates the case of a rabbit eye, the cornea of which had a value of 43,25 diopters and into which a graft of 47,25 diopters was inserted. A cornea of 48 diopters with perfect transparency and normal visibility of the ocular fundus was obtained.



Fig. 14. Myopic keratoplasty in a rabbit

Fig. 15 shows a 6mm. lamellar keratoplasty in a human eye, six months after the operation, which was performed with an optically neutral graft. The visual acuity in this eye at the present time is 0.62 with a correction of - 1.25 cil x 120.

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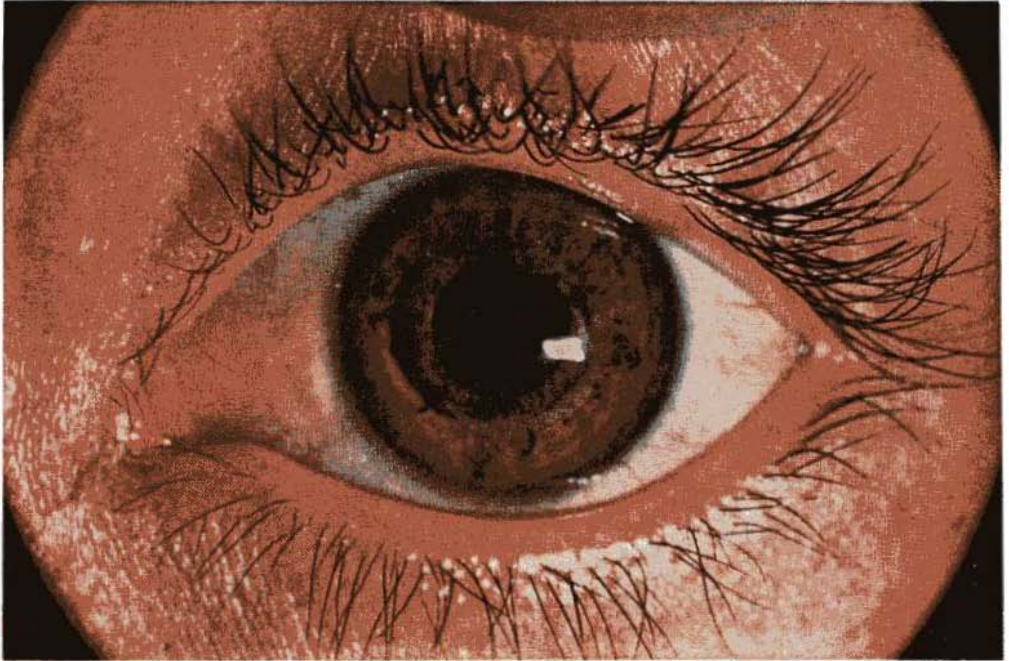


PLATE I

Keratoplasty in human eye, one year after the operation. Graft obtained by means of the procedure described.

V = 0.62 with - 1.25 cil  $\times$  120.

#### REFRACTIVE GRAFTS

These cases show the viability of the cornea when it is cut in lathe in a state of deep freezing. This is the only fact that we wish to establish in this article, since the determination of the precision which can be obtained in refractive correction should be the subject of wide experimentation.

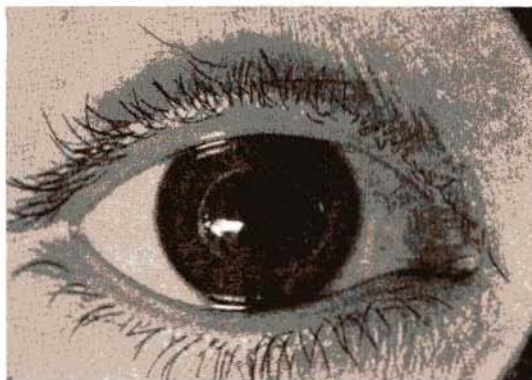


Fig. 15. Keratoplasty in human eye, 6 months after surgery. Graft turned under freezing state.

#### CONCLUSION

From the experiences of the author, as briefly reported in this article, it is obvious that it is possible to cut the corneal tissue in a state of deep freezing by mechanical means without causing it to lose its qualities, in order to make lamellar grafts for optical purpose. (Plate I).

#### SUMMARY

- a) The author describes a method for the uniform cutting of lamellar grafts from frozen cornea.
- b) He believes that the viability and the preservation of transparency of these grafts should be regarded as a demonstrated fact, from the results obtained and reported in this article.
- c) He describes the fundamentals of a method which opens a new horizon to the surgical treatment of refractive defects.

The procedure consists in turning the frozen cornea at  $79^{\circ}$  C. below zero in order to give it the desired curvature, size and thickness. By this procedure the Eye Banks would be enabled to deliver lamellar grafts perfectly cut and of the dimensions and refraction specified by the eye-surgeon.



By this method the author has been able to modify experimentally the spherical value of the cornea, in both autoplasties and homoplasties.

This article likewise suggests the possibility of correcting defects of astigmatism and also that of cutting lenses of corneal tissue for interlamellar implantation.

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#### BIBLIOGRAPHY

- 1) RYCROFT B. W. (1955) *Corneal Grafts*, Butterworths, London p. 5.
- 2) PAUFIQUE L., SOURDILLE G. P., OFFRET G. (1948) *Les Greffes de la cornée*. Masson. Paris.
- 3) BARRAQUER M. J. I. (1951) *La Histioterapia en Oftalmología*. Arch. Soc. Hisp. Amer., 11, 458.
- 4) BOCK R. H. (1949) *New Instrument for obtaining lamellar grafts*. Arch. Ophthal., 9, 458.
- 5) FRITZ A. (1950) *Sutured contact lenses for contention of the graft in keratoplasty*. Amer. Jour. Ophthal., 33, 1236.
- 6) WALSER E. (1949) *New Fixation procedure in keratoplasty*. Ber. Dtsch. Ophthal. Ges., 55, 382.
- 7) BARRAQUER M. J. I. (1949) *Queratoplastia refractiva. Estudios e informaciones oftalmológicas*, 2, Nº 10.
- 8) EASCOTT H. H., CROSS A. G., LEIGH A. G. y NORTH D. P. (1954) *Preservation of grafts by freezing*. Lancet, 1, 237.
- 9) CASTROVIEJO R. (1958) *Comunicación al Congreso Internacional de Bruselas*.
- 10) ARIZA H. E. (1958) *Lentes y Prótesis acrílica intracorneales*. Arch. Soc. Amer. Oftal. Optom., 1, 192.
- 11) BARRAQUER M. J. I. (1956) *The Microscope in ocular Surgery*. Amer. Jour. Ophthal., 42, 916.