In-Vitro Fusion of the Human Secondary Palate

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Introduction

In a recent communication (4), reference was made to unpublished earlier work in the North East of England (5). In order to establish the graciously acceded priority of our investigation, a brief account is now presented.

'In-vitro' organ culture of palatal shelves has been used to study the mechanism of palatal fusion in rats (8, 10), rabbits (2), and mice (13), and it has been shown, both morphologically and histologically, that 'in-vitro' fusion is comparable to the process 'in vivo' (7, 12). More recent studies have used this method to evaluate the effects of known teratogenic agents on mouse and rat embryonic palatal tissue (9, 6, 1).

In this laboratory, the technique has been successfully extended to human embryonic oral tissue.

Procedure

Human fetuses at about 6–7 weeks fertilization age as determined from external features (5) were obtained sterile at hysterotomy for sterilization and at hysterectomy. At this stage, the secondary palate is not yet fused. Six palates from embryos of 43–47 days fertilization were studied.

The head was detached from the body, and the tongue and mandible were removed. An incision was then made parallel to the roof of the mouth just below the level of the orbits. The nasal septum and any excess tissue posterior to the palatal shelves was trimmed away. The dissection was carried out in human amniotic fluid. The shelves were then washed in the culture medium (BGJ supplemented with 15 per cent fetal calf serum and 150 mg/L ascorbic acid) and placed, oral surface uppermost, on a millipore filter. The preparation was supported on an expanded metal grid in a 3 cm. borosilicate glass culture dish containing 1.5 ml. of culture medium and enclosed in a 9 cm. glass petri dish

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The work was supported by the Medical Research Council.
This paper was presented at the Meeting of the Research Group of the British Association of Plastic Surgeons at Newcastle upon Tyne in October 1974.
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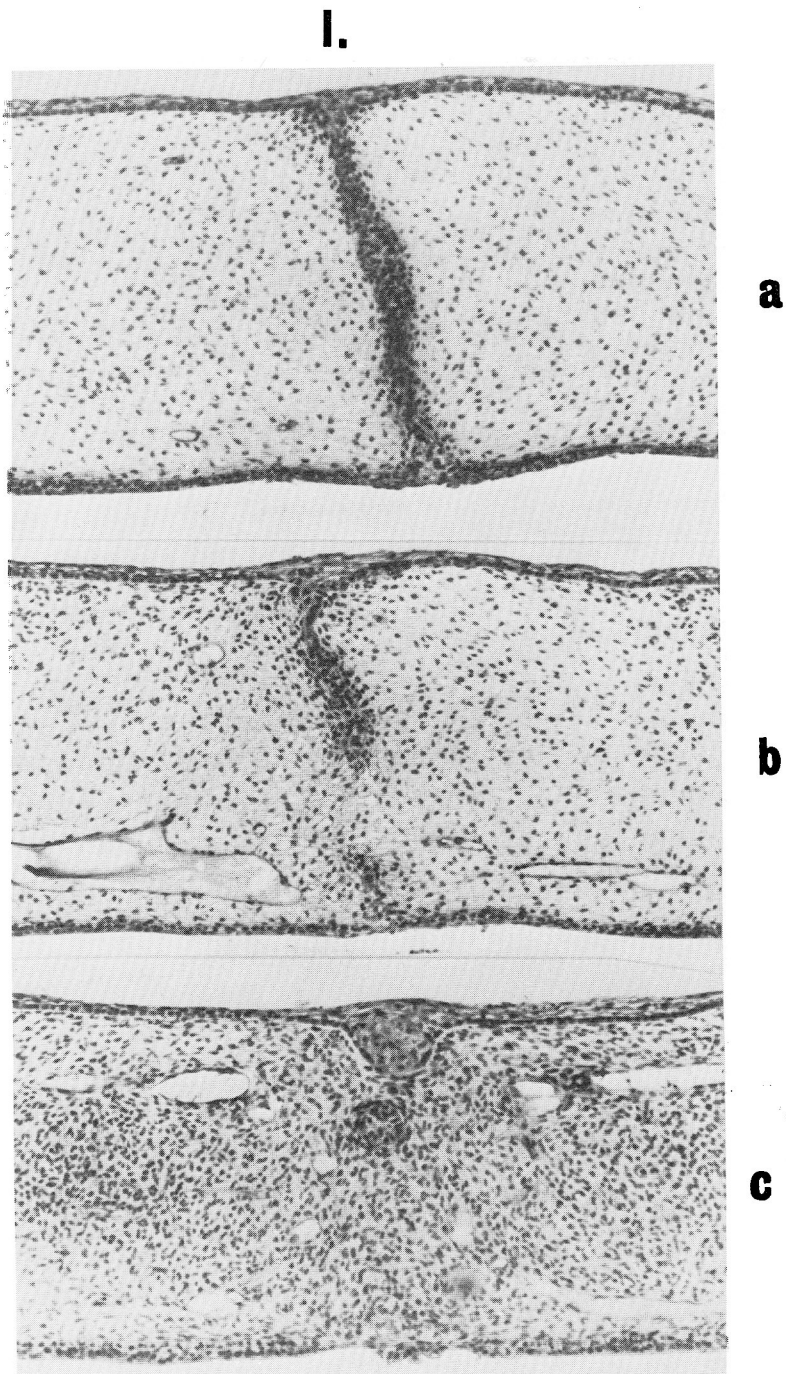

FIGURE 1. A, B, C, Successive stages in the fusion of the secondary palate of a 47-day fertilization age human embryo after 6 days in culture. Sections stained with Delafield's haematoxylin and chromotrope 2R.

containing filter paper saturated with 0.9 per cent sodium chloride. (For further details see references 3 and 11). The cultures were maintained at 37.5°C. in an atmosphere of 5 per cent Co_2 in air for 6 days. The culture medium was renewed every two days.

Results

The six palates that were cultured all showed both epithelial fusion (Fig. 1a.) and mesenchymal penetration (Figs. 1b. and 1c). In five of these cases, the palatal shelves were placed with their medial edges in contact at the commencement of culture; in one case contact was delayed for 24 hours.

This work was undertaken in the latter half of 1972 and early 1973. At that time a new trend in abortion practice was becoming obvious. With increasing experience, alternative forms of therapy were almost universally used so that sterile fetuses, never common, ceased to be available.

Acknowledgements: Our thanks are expressed to Dr. K. T. Rajan for his advice and to Dr. M. Thompson for providing laboratory facilities. This work was given financial support by the Medical Research Council.

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