

INTRODUCTION

Prolonged transport time and processing delays of tissue specimens are known to affect cell vitality. The aim of this study was to test the impact of storing tissue samples under vacuum condition prior to use for mouse implantation.

MATERIALS AND METHODS

Samples were obtained from patients who underwent surgical resection for pancreas ductal adenocarcinoma (PDAC). Samples were used from a total of 10 cases of PDAC. 80 SWISS-nu/nu mice were used for tumor implantation.

5 cases of fresh pancreas tumor tissue were cut in 3 samples: one was processed immediately (T0) and the other two were placed in a vacuum packed using Tissue Vacuum (Kaltek)[®] (Fig. 1) and stored refrigerated at 4°C for 24 hours (T24) and 48 hours (T48).



Fig.1: Tissue Vacuum (Kaltek)[®]



Fig.2: Athymic mice Swiss-nu/nu with tumor in the nape and right flank

Each sample was then fragmented into four pieces which were implanted in two immunodeficient SWISS-nu/nu mice, one fragment in each of the nape and right flank of each mouse (Fig. 2).

Based on results of 48 hours we then successfully tested other 5 cases up to 96 hours using the same methods. Cases with larger tumor size were selected to permit 5 samples from each case to be used for the study for implanting immediately (T0), at 24 hours (T24), at 48 hours (T48), at 72 hours (T72) and at 96 hours (T96).

RESULTS

Tumor fragments implanted in the right flank of each mouse grew within 17 days of implantation (Fig 3) showing the viability of tumor tissue stored vacuum refrigerated for up to 48 hours for 3 of the first 5 cases (Table 1).



Fig.3: tumorgraft

Table 1: Growth of tumor implant in mice up to 48 hours

SAMPLE NO.	T0	T24	T48
1590	growth	no growth	no growth
1592	growth	growth	growth
1608	growth	growth	growth
1609	growth	growth	growth
1610	growth	no growth	no growth

Table 2: Growth of tumor implant in mice up to 96 hours

SAMPLE NO.	T0	T24	T48	T72	T96
1613	growth	growth	no growth	growth	growth
1626	growth	growth	growth	growth	growth
1630	growth	growth	growth	growth	growth
1635	growth	growth	growth	no growth	growth
1638	growth	growth	growth	growth	growth

growth
no growth

In the second set of 5 cases, tumor fragments grew within 20 days of implantation showing the viability of tumor tissue stored vacuum refrigerated for up to 96 hours (Table 2 and Fig. 4).

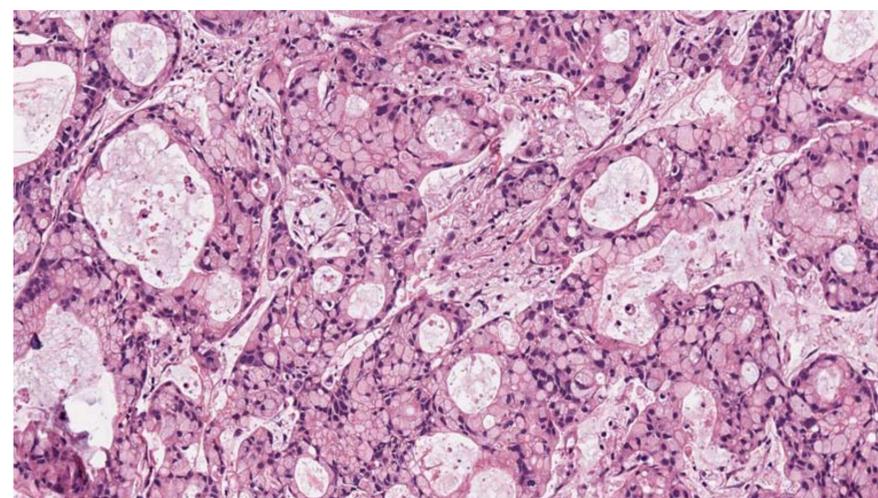


Fig.4: H&E of PDAC xeno-graft from human tissue vacuum refrigerated for 96 hours

CONCLUSIONS

Samples can be maintained fresh for up to 96 hours and still guarantee cellular vitality. This permits the possibility to produce cell cultures even after prolonged delays from tissue sampling. Furthermore it facilitates xenograft production by maintaining cellular viability for implantation and growth. Perhaps most important of all, it provides options for long distance transport of fresh tissue with less stringent transport conditions.