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KAPP-Sen TMC: Whole Pancreas Preparation

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Cellular Senescence Net...

KAPP-Sen TMC



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We use this protocol and it's working

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Abstract

Whole pancreas and tissue blocks preparation

Troubleshooting

Overview

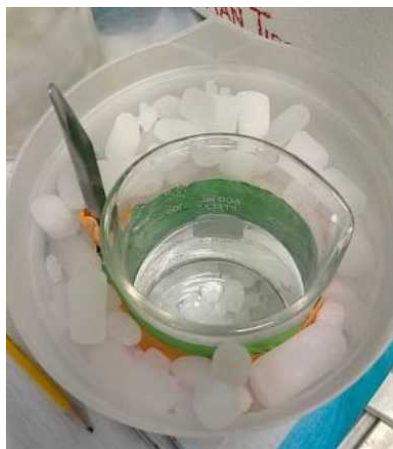
- 1 All the whole pancreases were procured by the **University of Texas Health San Antonio** and shipped to **Joslin Diabetes Center, University of Harvard Medical**. The pancreases were cut into the Head, Body, and Tail of the locations along the guideline (1), cut into 4mm thickness, and placed into order paraffin or frozen cassettes. The paraffin cassettes were fixed in 4% paraformaldehyde for 24 hours and processed for paraffin embedding at **Joslin Histology Core**. The frozen cassettes were molded with O.T.C. compound (*Fisher Scientific No.23-730-571*) and frozen in the cold 2-Methylbutane (*Fisher Scientific No.60-048-072*) bath followed by dried and stored at -80°C . At least three paraffin cassettes per each location of Head, Body, and Tail were shipped to **The Jackson Laboratory**; all the procedures were done at 4°C and avoided exposure to direct light.

Preparation of Human Pancreas

- 2 **30 minutes before the pancreas arrives:**

1. Prepare 2-Methylbutane bath

- Fill the bottom of the plastic big beaker with dry ice (use tip, not powder dry ice)
- Place a glass beaker inside a larger plastic beaker
- Add dry ice around the glass beaker up to 400.
- Pour 100% EtOH onto dry ice until it appears "slushy" - keep adding dry ice chips and ethanol as needed. Should be bubbling.
- Fill the glass beaker about half-way with 2-Methylbutane and let it chill for 15-20 minutes before using.



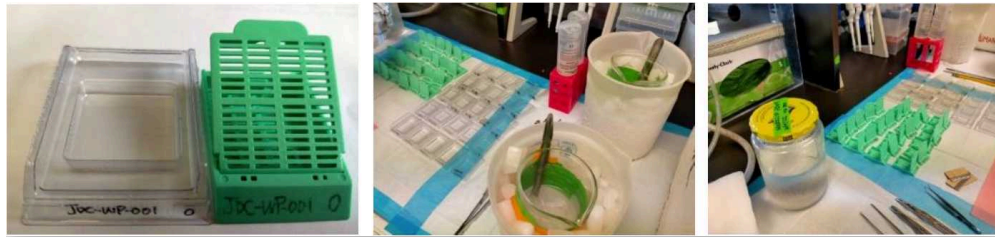
2. Prepare Cassettes

- Name the cassette "JDC-WP-####-X" to both green paraffin cassettes and clear frozen mold. #### should be the experimental number. X should be alphabetical from a to z.

After z, create aa, ab, up to ae.

-Add OCT to cover the bottom of a labeled frozen mold without bubbles.

-Set everything like the pictures



3. When the Pancreas Arrives

-Get ice and place between two trays.

-Pour PBS into the top tray until the pancreas is completely immersed.



4. Test isopentane to make sure that it is cold enough - with long forceps, put mold with OCT medium into chilled isopentane (if it doesn't turn within 5 seconds, wait longer, and add more dry ice and more ethanol).

5. After removing fat from the pancreas, spread out pancreas on wax, and cut into Head, Body, and Tail (see Section "**Examination of Specimens from Patients with Carcinoma of the Pancreas**" for the definition of anatomical locations), and measure the weight.

-Paraffin: put it in the green cassettes from 'a'

-Frozen: put it onto the mold and add another OCT. Dry it by placing it on dry ice, and standing it up at an angle.



6. Be sure to bleach everything for over 30 minutes. ***Always treat it as contaminated with an infectious disease.***



Examination of Specimens from Patients with Carcinoma of the Pancreas

3 From "Pancreas Exocrine 4.0.0.1" page 8, explanatory note **B. Definition of Location**

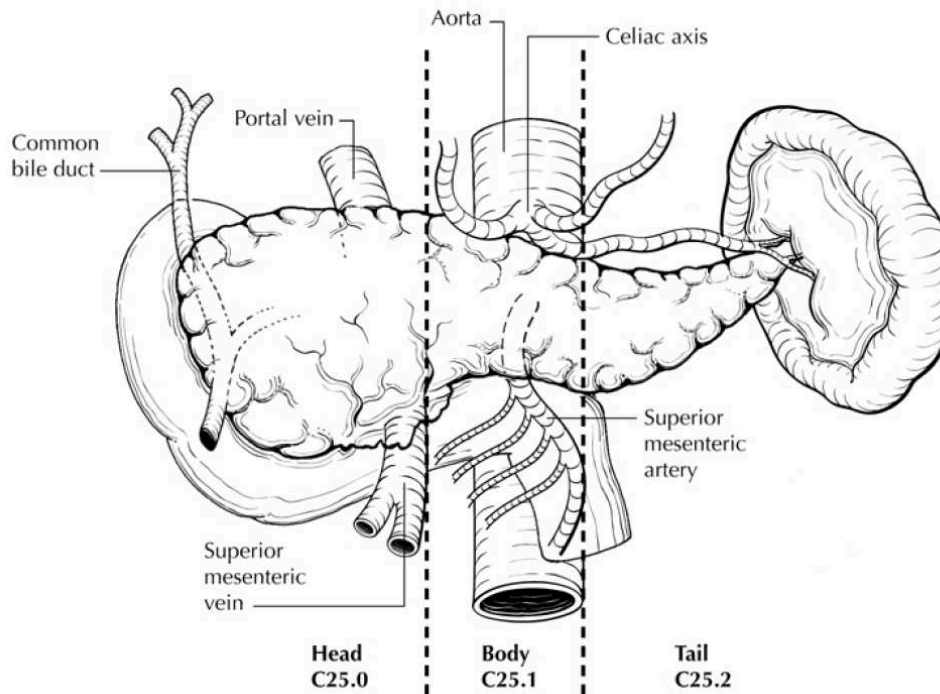


Figure 1. Anatomic subsites of the pancreas. From Greene et al.³⁰ Used with permission of the American Joint Committee on Cancer (AJCC), Chicago, Illinois. The original source for this material is the AJCC Cancer Staging Atlas (2006) published by Springer Science and Business Media LLC, www.springerlink.com.

Citation

Sanjay Kakar, Chanjuan Shi, N. Volkan Adsay, Patrick Fitzgibbons, Wendy L. Frankel, David S. Klimstra, Alyssa M. Krasinskas, Mari Mino-Kenudson, Timothy Pawlik, Jean-Nicolas Vauthey, Mary K. Washington (2017)

. Protocol for the Examination of Specimens From Patients With Carcinoma of the Pancreas. Pancreas Exocrine 4.0.0.1: Includes pTNM Requirements From The. 8th Edition AJCC Staging Manual.

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Citations

Step 3

Sanjay Kakar, Chanjuan Shi, N. Volkan Adsay, Patrick Fitzgibbons, Wendy L. Frankel, David S. Klimstra, Alyssa M. Krasinskas, Mari Mino-Kenudson, Timothy Pawlik, Jean-Nicolas Vauthey, Mary K. Washington. Protocol for the Examination of Specimens From Patients With Carcinoma of the Pancreas. Pancreas Exocrine 4.0.0.1: Includes pTNM Requirements From Theh

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