Perioperative Considerations for Tumor Tissue Procurement to Obtain Tumor-Infiltrating Lymphocytes for Adoptive Cell Therapy

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MOFFITT CANCER CENTER SARCOMA

SPOT Lab™
We investigate TIL cancer is cured.

Disclosures

- Sponsored research agreement: Intellia Therapeutics
- Consultant: Merit Medical, Lyell Therapeutics, and Iovance Biotherapeutics
- Moffitt Cancer Center has licensed Intellectual Property (IP) related to the proliferation and expansion of tumor infiltrating lymphocytes (TILs) to Iovance Biotherapeutics. I am an inventor on such Intellectual Property.
- Research support: NIH-NCI (K08CA252642), Ocala Royal Dames, and V Foundation

Perioperative Considerations: Patient Factors

TIL Therapy Challenges: Patient Factors

- Co-morbidities
  - H&N SCC, NSCLC: pulmonary function, tobacco-related health concerns
  - Bladder/RCC: Renal function
- Prior systemic therapy regimens
  - Doxorubicin-based systemic therapy:
    - Cardiac dysfunction (depressed EF)
  - Multiple prior cytotoxic chemotherapy regimens:
    - Marrow suppression
- Site of tumor resection

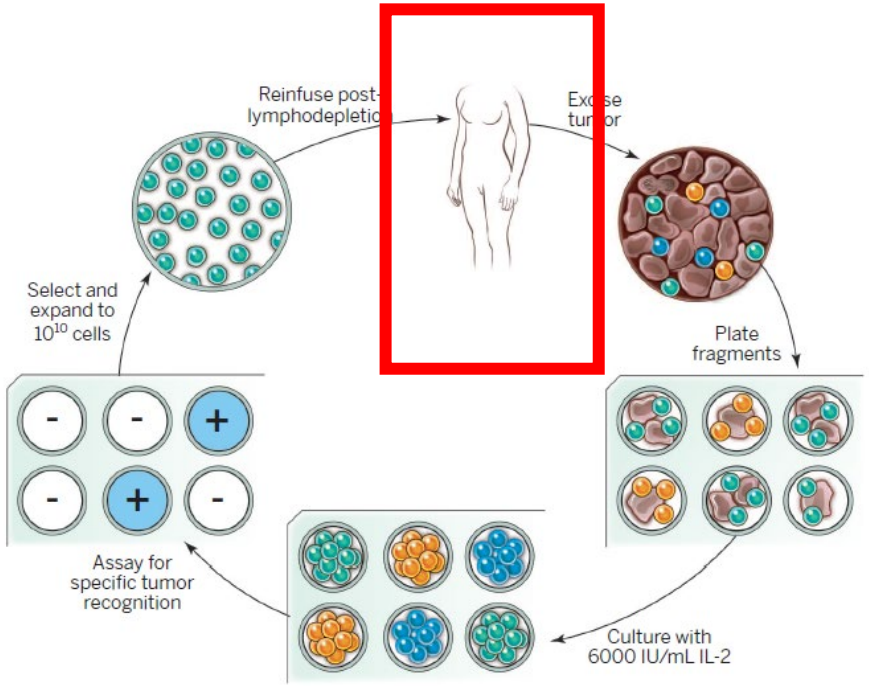

Figure: Rosenberg, Restifo. Science 348(6230); 62-68, 2015.

NCI Surgery Branch Review & Editorial

Ann Surg Oncol (2018) 25:565–572
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CONTINUING EDUCATION– TRANSLATIONAL RESEARCH AND BIOMARKERS

Metastasectomy for Tumor-Infiltrating Lymphocytes: An Emerging Operative Indication in Surgical Oncology

Joseph G. Crompton, MD, PhD[1], Nicholas Klemen, MD[2], and Udai S. Kammula, MD[3,4]

TABLE 2 Patient selection for adoptive cell transfer (ACT) immunotherapy

Relative contraindications	Age < 18 or > 70 years
	ECOG performance status > 1
	Unacceptable risk of sepsis or bleeding during 7–10 days of neutropenia and thrombocytopenia
	Inability to tolerate interleukin-2 administration due to cardiopulmonary or renal insufficiency (some ACT protocols use low-dose or no IL-2)
	Current treatment with corticosteroids or immunosuppressive agents
Absolute contraindications	Primary immunodeficiency or chronic viral disease (e.g., HIV, HBV, HCV)
	Pregnancy
Other considerations	Large, symptomatic, or bleeding CNS lesions should be treated before ACT.
	Although trial eligibility may necessitate treatment with standard-of-care therapy before ACT, metastasectomy for TIL harvest can be performed first and the T cells frozen for later use.
	Patients in trials may require a radiographically evaluable target lesion for measurement of response to ACT.

ECOG Eastern Cooperative Oncology Group, *HIV* human immunodeficiency virus, *HBV* hepatitis B virus, *HCV* hepatitis C virus, *CNS* central nervous system, *TIL* tumor-infiltrating lymphocyte

TABLE 3 Operative considerations in isolating tumor-infiltrating lymphocytes (TILs)

Consideration	Details
Tumor size	Tumor size does not correlate with TIL efficacy, but tumors should be at least 2 cm in largest diameter to obtain adequate yield of tissue for processing.
Irradiated tumors	Avoid harvesting TILs from a tumor site that has previously been irradiated.
Tumor site	Because TILs can be procured from a variety of tumor sites, favor surgical sites that result in minimal morbidity and consider laparoscopic approach.
Margins	Wide surgical margins and major organ resection are not typically necessary unless the tumor resection is being performed for curative intent. Avoid cutting through tumor to minimize risk of seeding tumor site.
Wound healing	Avoid harvest of superficial lesions if wound healing may be compromised.
Contamination	Ulcerating tumors and those with high suspicion for bacterial colonization can result in contamination of cultures. Isolation of TILs from bowel lesions is possible but may be associated with an increased risk of contamination.
Splenic lesions	Splenic tumors are not optimal for TILs because of theoretical concern that they may be enriched in bystander lymphocytes that are not tumor-reactive.
CNS lesions	Tumors metastatic to CNS have not been adequately assessed as a source of TILs for treatment.
Harvest	Refer to institutional guidelines for instructions on handling, processing, and labeling of tumor specimens.
Confirmation	Confirmation that the metastasectomy specimen contains malignant cells will ensure that benign or nodal tissue has not been inadvertently collected.

CNS central nervous system

SITC Guidelines for Tissue Procurement

Journal for ImmunoTherapy of Cancer

Defining best practices for tissue procurement in immuno-oncology clinical trials: consensus statement from the Society for Immunotherapy of Cancer Surgery Committee

TIL Harvest Recommendations

1. Fit patient with non rapidly progressive disease
2. Close collaboration between surgeon (if not PI) and treating physician regarding optimal site for harvest relative to target lesions
3. Sufficient tissue (>2cm^3) with minimal morbidity using "least invasive technique"
4. Placement into media immediately after resection to ensure sterility
5. Partition of specimen, if needed, should be performed in sterile biologic safety cabinet within pathology

Table 5 Major considerations in biospecimen collection and handling reported by the Society for Immunotherapy of Cancer Surgery Committee

Prior to biopsy	During collection	After collection	Therapeutic immune cell collection
Early consultation with surgical or interventional physician expertise	Ensure that all supplies and collection containers are available	Ensure rapid delivery to laboratory, if appropriate	Sterile conditions must be used throughout
Early consultation with surgical pathologist to plan specimen allocation and testing (ie, SOC vs research)	Once tissue is obtained, processing should be as rapid as possible	Rapid shipping with proper labels and addresses	Ensure all processing and shipping SOPs are in place
Ensure IRB approval and written informed consent are obtained prior to the procedure	Ensure enough tissue is obtained, especially if required for SOC	Monitor the temperature of collected specimens prior to processing and avoid excessive heat	Work with clinical immunotherapy experts to ensure appropriate patients and lesions are selected
Establish SOPs for specimen collection at institution	If sample not fixed immediately, consider vacuum sealing, placing in sterile gauze with preservative fluid, or on ice in sterile system	If processing is delayed, keep specimen on ice unless otherwise indicated	Confirm days and times open for specimen receiving prior to procedure
Ensure that all personnel involved in tissue collection are trained in local SOPs	If a biosafety cabinet is not available, establish a "clean" area for initial specimen handling		
Consider the number of cells and viability status needed from tissue; consult with immunology experts to define	Avoid contact between different specimens		
Consider preservatives needed to process and store tissue once collected	Use new supplies and containers for each new specimen		
Consider if matched specimens are needed (ie, PBMC and tumor) at each time point			
Determine the type of biopsy (eg, core needle, incisional, excisional, etc.) to be done and what instruments and reagents (eg, needles, collection bottles, preservatives, etc.) are needed			
Understand institutional policies and regulations, including coordination with pathology for SOC			
Ensure pre-labeling of all specimen containers and patient materials			
Consider using a time tracking process with documentation			
Consider collecting normal tissues as control			

IRB, institutional review board; PBMC, peripheral blood mononuclear cell; SOC, standard of care; SOP, standard operating procedure.

Gastman, *Journal of Immunotherapy for Cancer*, 2020

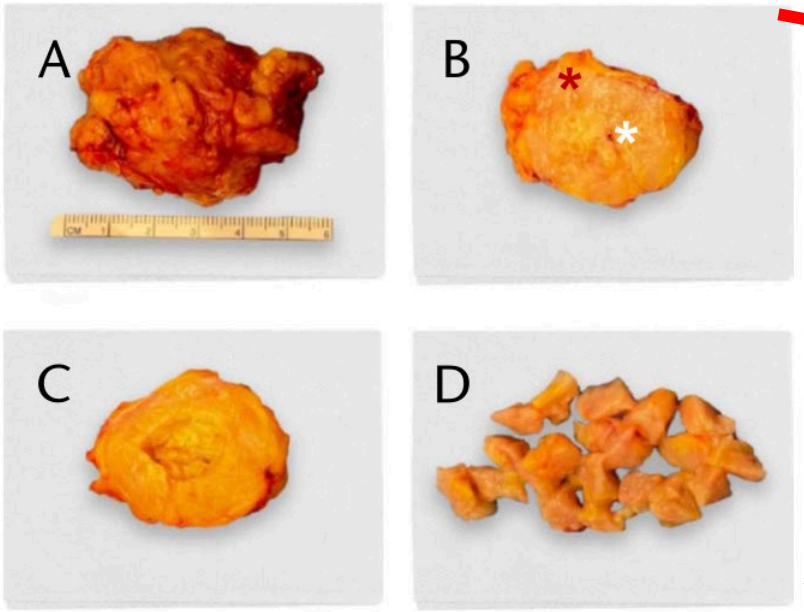
Multi-institutional Review of Tissue Procurement Principles

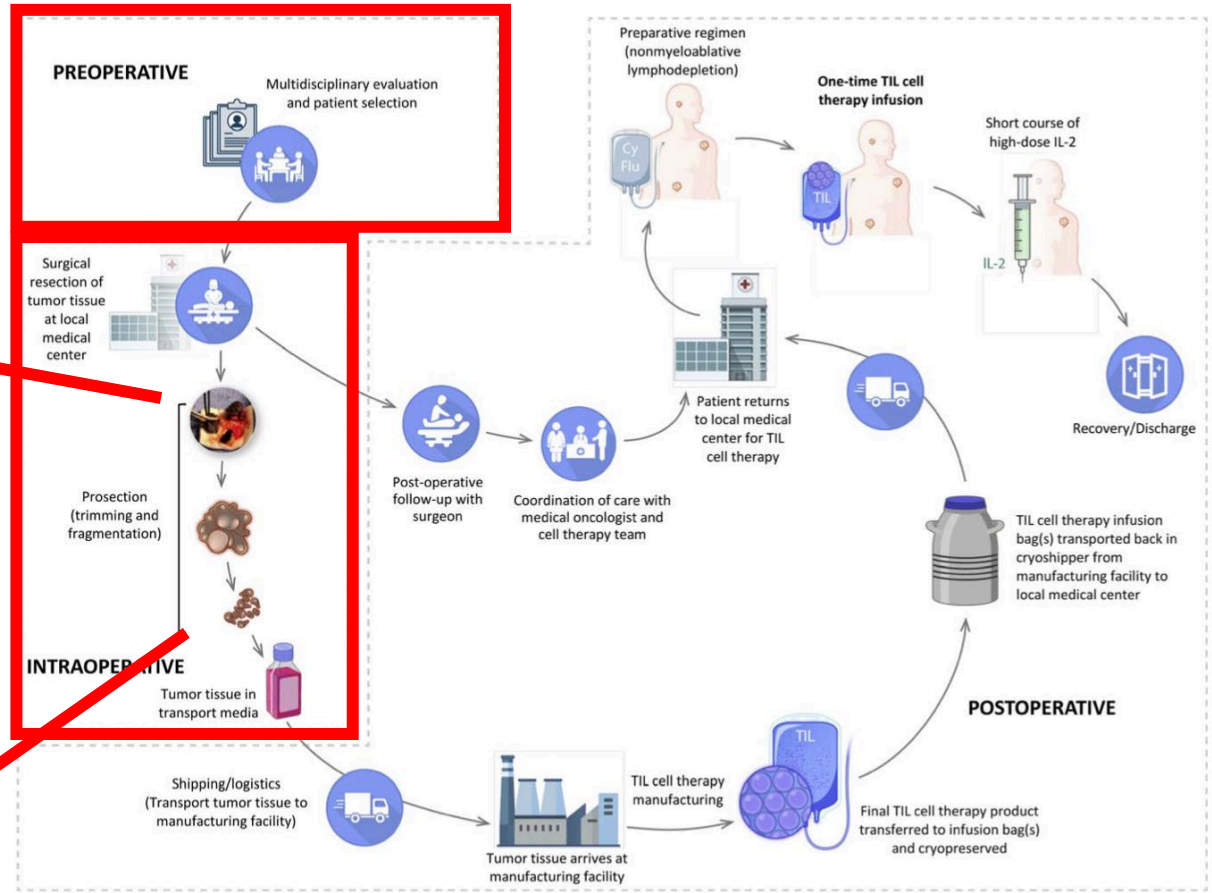
REVIEW ARTICLE

OPEN

Surgical Considerations for Tumor Tissue Procurement to Obtain Tumor-Infiltrating Lymphocytes for Adoptive Cell Therapy

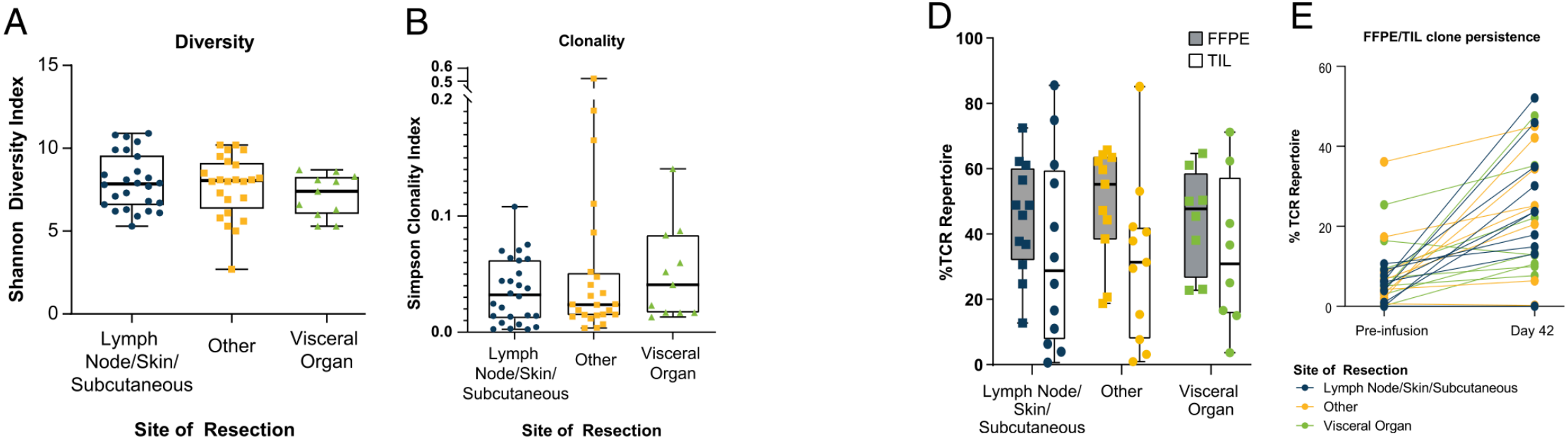
John E. Mullinax, MD,†‡ Michael E. Egger, MD, MPH,§ Martin McCarter, MD,|| Bradley J. Monk, MD,¶ Eric M. Toloza, MD, PhD,†‡# Susan Brousseau, RT, MBA,** Madan Jagasia, MD,** and Amod Sarnaik, MD†‡††*





Mullinax et. al, *Cancer Journal*, 2022

Site of TIL harvest does not impact TCR repertoire of infusion product



Mullinax et. al, *Cancer Journal*, 2022

Perioperative Considerations:
Team Engagement and Tissue Workflow

Fundamental Intraoperative Principles

- Operating room layout
  - Location of tumor after extirpation
- Sterility
  - Preclinical: ensure sterile process that allows for SOC margin assessment
  - Clinical trial (ACT patient): Prepare specimen in OR, place in media or shipping container
- Control of the tissue
  - Part of the surgical time out before case begins
  - Detailed team instructions
- Inclusion of all stakeholders
  - Dedicated team of interested perioperative staff
  - Pathology engagement

Cell Therapy Clinical Program

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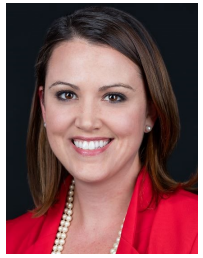
Tissue Core



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Pathology

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Vice Chair, Research Pathology

Janis de la Iglesia, PhD
Path Research Services Manager

Cell Therapy Facility

Cheryl Cox
Director, CTF Operations

Coordinating the Team

- Laminated OR placard
- Research blood collection kits
  - Include tubes, instructions
  - Disconnect from standard clinical process
- Prepare dedicated research/clinical trial supplies
  - Disposable scalpel, forceps, specimen cups

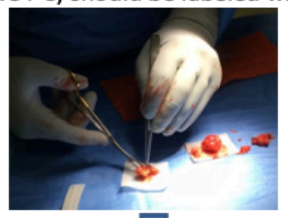
MCC Protocol 19837, PI: Dr. John Mullinax (Sarcoma TIL Trial)

Contacts

Tissue Core: 256-4665 — PI, Dr. John Mullinax: — (Office) : 745-8736; (Pager) : 256-5784
Cell Thrpy Core: 256-5079 — Coordinator, Brook Olmo: — (Office) : 745-5221; (Pager) : 256-4498

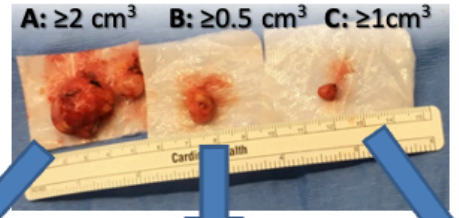
Instructions:

- This is a clinical trial, it is not part of TCC, but it is managed by Tissue Core.
- Tissue Core will carry in blue and orange cups containing sterile media where the **largest piece** of tumor to be placed.
- Surgeon is to cut the tumor tissue into three pieces (in order of importance) as follows in the OR:
  - **A.** ≥2 cm^3 collected in sterile media (blue cup prepared by Cell Therapy Core)
  - **B.** ≥0.5 cm^3 (Green dot, collected and frozen by Tissue Core)
  - **C.** ≥1 cm^3 collected in sterile media (orange cup prepared by Mullinax Lab)
- Samples A and B (NOT C) should be labeled with PHI



1. Surgeon prosects adjacent tissue and allocates tumor into 3 portions under sterile conditions.

2. **Page Tissue Core**. Tissue core tech will arrive with labeled cups.

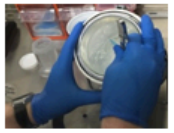


3. **Transfer** samples to correct containers based on size.

STERILE TRANSFER

A. Tissue Core brings **blue cup w/ media.**

B. Standard Anatomic Pathology.



C. Weighed & Snap-frozen by Tissue Core. Send to Mullinax Lab

Stored in 4°C Frig by Tissue Core, notifies Cell Therpy Core for pick up.
DO NOT FIX OR FREEZE

Uniform Specimen Labeling Ensures Consistency

Fresh and / or sterile tissue collection required

GREEN DOT
Purpose: for biobanking

KEY POINTS

- **MAY be recovered** by pathologist for clinical care
- **Must be documented** on OR pathology requisition as separate specimen with unique letter
- **Documented, received, processed** by Tissue Core

BLUE DOT
Purpose: for research special collection

KEY POINTS

- **NOT Recoverable** by pathologist for clinical care
- **IF ACCOMPANIED BY ROUTINE PATHOLOGY SPECIMEN, then must be documented** on OR pathology requisition as separate specimen with unique letter; otherwise **Not Included** on OR pathology requisition
- **Documented, received, and transported** by Tissue Core for immediate release to research lab

WHITE DOT
Purpose: for Cell Therapies Facility

KEY POINTS

- **NOT Recoverable** by pathologist for clinical care
- **IF ACCOMPANIED BY ROUTINE PATHOLOGY SPECIMEN, then must be documented** on OR pathology requisition as separate specimen with unique letter; otherwise **Not Included** on OR pathology requisition
- **Immediate handoff** to Cell Therapy Facility staff for transport (present in OR)

If a concern arises regarding the appropriateness of tissue for allocation with respect to histopathologic assessment, please page on-call frozen section pathologist.

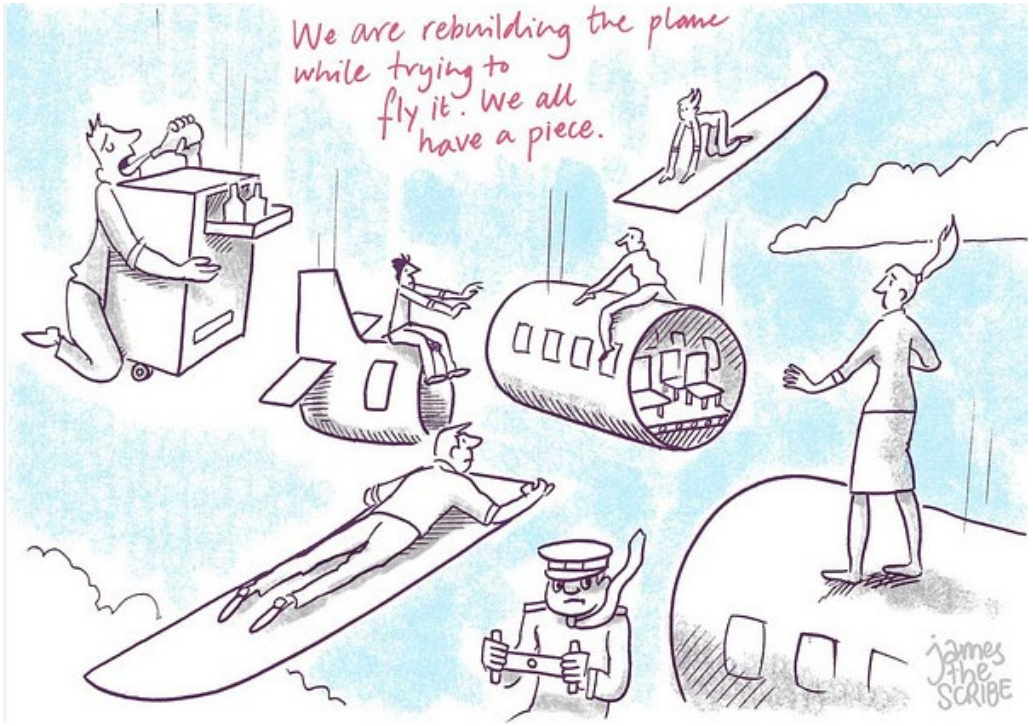
Frozen Room: #. *MCC:* 813-745-2931 *MKC*: 813-745-7106 Cell Therapy: page 813-256-5176

Moffitt Cancer Center TIL Harvest Workflow

Surgical Timeout
Pre-incision

Emerging considerations

- Pathology involvement
  - Ownership of specimen
  - Documentation
- Surgeon volume
  - Scheduling restrictions related to manufacturing
  - Accommodation with clinical practice
- Informed consent
  - Non-therapeutic operation
  - Acceptance of risk
- Billing
  - Reimbursement of procurement operations



Summary

- TIL harvest from non-LN tissue is safe and effective with minimally invasive surgical approach (i.e. laparoscopic, robotic-assisted)

- Non-melanoma solid tumors require greater consideration
  - Patient co-morbidities
  - TIL harvest site

- Team involvement is crucial for ACT clinical trial and preclinical research success
  - From surgical consultation to tissue in media
  - Early involvement of surgeon

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