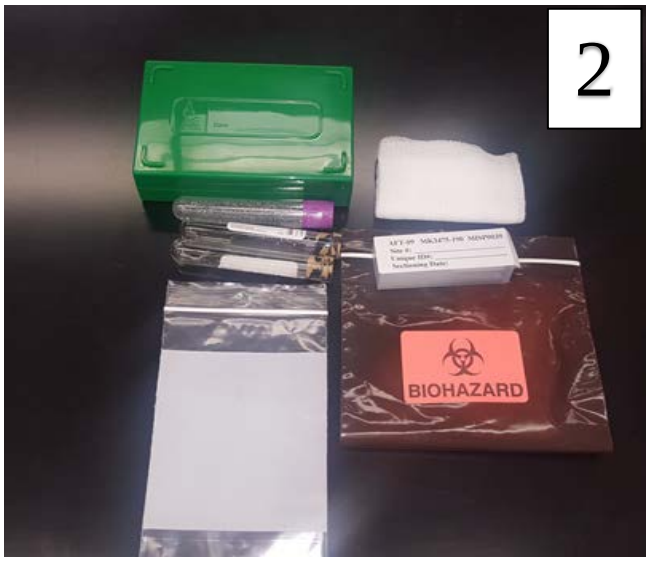
Collection Protocol Title: Randomized Phase II Trial Evaluating the Optimal Sequencing of PD-1 Inhibition with Pembrolizumab (MK-3475) and Standard Platinum-based Chemotherapy in Patients with Chemotherapy naïve stage IV Non-small Cell Lung Cancer

Collection Protocol Short Title: AFT-09

Collection Protocol PI: AFT Administrator

Collection Time Points: **(1)** Screening **(2)** Cycle 4 **(3)** Cycle 8

Biospecimen Collection Kit Contents: Single Shipper



Set #	Component	QTY	Time Points
1	Single shipper (Outer box w/ pre-printed Biological Substance, Category B,UN 3373 Markings, Insulated Chest)	1	(1) (2) (3)
	FedEx Airbill for return shipment	1	(1) (2) (3)
	Disposable Secondary Pressure Vessel (Medium) (Outer envelope, Inner leak proof polybag)	1	(1) (2) (3)
	Saf-T-Rap® Sticky Side Bubble Wrap - 6 x 12 Inches	1	(1) (2) (3)
	100 mL Absorbent Strip	1	(1) (2) (3)
	Saf-T-Temp controlled room temperature (CRT) PCM packs - 500G *(Not Pictured)*	1	(1) (2) (3)
2	Bag, specimen, minigrip reclosable (4” x 6”)	1	(1)
	Vacutainer® K_2 EDTA Tube (Lavender top, 10ml)	1	(1)
	Streck Cell-Free DNA BCT® Tube (Black/Brown Marble top, 10mL)	2	(1) (2) (3)
	Slide box, holds 25 slides, plastic green	1	(1)
	Gauze, sterile cotton (2” x 2”)	3	(1)
	Charged Capillary Gap (ProbeOn Plus™) Microscope Slides (in holder)	5	(1)

See backside for Collection Site Instructions

Collection Site Packaging Instructions

Follow packaging instructions to ensure kit performance and shipment compliance with IATA Packaging Instructions 650 for biological specimens that are being shipped and packaged. Properly packed, this kit will ensure ambient specimens remain at appropriate temperatures for up to 24hr. Do *NOT* freeze or refrigerate the Saf-T-Temp (CRT) PCM pack included in the kit.

Specimen Processing

1) Label slides, microcentrifuge tubes, specimen bag, K_2 EDTA Vacutainer® tube and Streck Cell-Free DNA BCT® tube with patient study ID, patient initials and date of specimen collection using a permanent marker.
   a. Slides for PD-L1 staining must be labeled with patient study ID, institutional surgical pathology number, institutional block ID, and serial cut number (i.e. 1-5).
2) Collect whole blood into K_2 EDTA Vacutainer® and Streck Cell-Free DNA BCT® tubes according to protocol instructions; wrap tubes in bubble wrap.
3) Cut paraffin block sections to 5-Charged Capillary Gap (ProbeOn Plus™) Microscope Slides according to protocol instructions. Transfer into the 5-slot slide holder provided, place piece of blue foam on top of slides, tape holder lid closed, wrap with bubble wrap, insert slide holder into Amber UV Biohazard Bag and close bag.
   a. The slide mailer containing the slides for PD-L1 must also be labeled with patient study ID, site number, and sectioning date.
4) If collected, wrap the paraffin block with 1 piece of gauze and place into the labeled 4x6 minigrip specimen bag.
5) *Optional Specimens-Unstained slides:* Cut unstained paraffin block sections to microscope slides according to protocol instructions and transfer them into the 25-slot slide box provided. Place two pieces of gauze on top of the slides before securing the lid of the slide holder.
6) *Optional Specimens-H&E and Tissue curls*: Place H&E slide into the 25-slot slide box provided. Cut tissue curls according to protocol instructions and transfer into labeled 1.5-2.0ml microcentrifuge tubes or similar *(not provided with kit)* and place tubes inside a small biohazard bag *(not provided with kit).*

Specimen Packaging

1) Place bubble-wrapped K_2 EDTA Vacutainer® and Streck Cell-Free DNA BCT® tubes, along with the paraffin block in the minigrip bag, inside the leak-proof inner poly bag (biohazard) of the disposable secondary pressure vessel along with the 100mL absorbent strip. Seal the poly bag by removing the red tape, place inside the white Tyvek envelope and seal the Tyvek envelope.
2) If optional microscope slides (unstained/H&E) are collected, wrap the 25-slot slide box with bubble wrap provided.
3) Place all specimens inside the insulated chest of shipping container and secure the chest lid.

Preparing the Shipping Container

1) Register specimen collection information in the AFT BioMS tracking database, which **is accessible via the AFT portal (https://alliancefoundationtrials.org)**. Place a copy of the AFT BioMS packing slip inside of the shipping container.
2) If applicable, include a de-identified copy of the institutional surgical pathology report pertaining to the tissue/slides sent.
3) Close the outer corrugated box and secure with packaging tape
4) A completed FedEx Air Bill is included with the kit to return to the biorepository. (Please do not interchange FedEx air-bills between kits).
   a. Ship to: AFT Biorepository at Washington University, c/o Siteman Cancer Center Tissue Procurement Core, 425 S. Euclid Ave., Rm 5120, St. Louis MO, 63110-1005; Phone (314-454-7615).
   b. Do not ship on Friday, Saturday, Sunday or day before a nationally recognized holiday.

Requesting Additional Biospecimen Collection Kits

1) All Biospecimen Collection Kits provided by the AFT Biorepository must be requested through the AFT Biospecimen Management System (AFT BioMS); **do not call, email or fax the biorepository.** Please contact the AFT BioMS Helpdesk (aftbiomshelp@bmi.wustl.edu or 855-642-4667) for questions regarding kit requests through AFT BioMS.
2) Kit requests will be processed and distributed within 10 working days of request receipt.
   a. The number of kits distributed per request may be limited based upon accrual rate of the trial or due to kit availability. Requesting sites will be notified in advance if the number of kits received will differ from what was requested.
   b. Kits distributed will have a shelf life of at least 90 days, unless precluded by the stability of a particular component.
   c. In the event a kit has expired components, the **entire kit** must be returned to the AFT Biorepository.