HALT-C Trial Q x Q

Specimen Collection

Form # 14 Version B: 03/19/2001

Purpose of Form #14: The Specimen Collection form is used to record the collection of liver tissue. The form records the collection date, specimen size, and other specifics about the liver specimen.

When to complete Form #14: This form should be completed for all patients when a liver biopsy is done, usually at the following study visits:

- Screening phase: Screening (S00).
- Randomization phase: Month 24 (M24) and Month 48 (M48).

Form #14 is data entered at each clinical site and should never be set to missing in the DMS.

- ► In the DMS, Form # 14 is expected even if a patient misses the M24 or M48 visit. You will be reminded when you run an Outstanding Forms Report that you must complete this form as outlined below. You can complete and data enter Form #14 when the patient is willing to have the biopsy after a missed M24 or M48 visit.

- ► If at M24 or M48, the biopsy is not done, but is scheduled for a later date, leave Form #14 as expected in the DMS. Complete the paper copy when the biopsy is done and data enter in the DMS under the M24 or M48 visit.

- ► If you know for certain that the M24 or M48 biopsy will never be done, complete and data enter Form #14. Answer "NO" on Question B1 asking if liver tissue was collected at this visit. Add a written comment to the form explaining why the biopsy could not be completed (for example, "PI discretion because of low platelets"). The data entry person should add this written statement as a form level comment in the DMS.

SECTION A: GENERAL INFORMATION

A1. Affix the patient ID label in the space provided.
- If the label is not available, record the ID number legibly.

A2. Enter the patient's initials exactly as recorded on the Trial ID Assignment form.

A3. Enter the three-digit code corresponding to this visit.

A4. Record the date of the visit in MM/DD/YYYY format.

A5. Enter the initials of the person completing the form.

NOTE: Liver Tissue Collection Protocol Changes

The HALT-C Steering Committee approved a change in the protocol for handling of liver biopsy specimens. Effective June 15, 2004, all HALT-C Clinical Sites should follow the revised liver biopsy protocol. For sites participating in the Immunology/Virology Ancillary Study, the liver biopsy protocol was updated again on August 16, 2004.

PRIORITIES:

1. The **top priority** for use of the biopsies will continue to be at least a 20 mm core for histological interpretation for the Main HALT-C trial.

2a. If there is excess liver tissue from a patient not participating in the Immunology-Virology AS:

- Next priority: continue the current practice, whenever possible, to place 3.0 mm (or larger) cores of biopsies into cryovials, flash-frozen in liquid nitrogen at the bedside, and later transferred to dry ice (-80^{0}C or colder) for long-term storage.

- Last priority: whenever possible, embed any remaining liver tissue (preferably $\geq$ 4.0 mm cores) from HALT-C biopsies at the bedside into Tissue-Tek OCT prior to freezing in liquid nitrogen. A detailed procedure on how to collect liver in OCT is provided in Numbered Memo 27.

2b. If there is excess liver tissue from a patient who consented to the Immunology/ Virology Ancillary Study, the M24 and M48 VCS or upcoming procedures report in the DMS specifies whether the patient is in the CTL and/or Replication substudy:

  - If the patient had fresh liver tissue collected at S00, collect fresh liver at M24 and at M48 for both the CTL and Replication studies as per the 12/2002 protocol and MOO.

  - If the patient had **no** fresh liver tissue collected at S00, collect fresh liver at M24 and at M48 for the CTL study only, not the Replication study.

  - If the patient had **no** fresh liver tissue collected at S00 and **no** fresh tissue collected for CTL at M24, do not collect fresh liver for the CTL study or the Replication study at M48.

- The next priority is 5 mm fresh tissue for the CTL substudy.

- For patients participating in the Replication study at M24 and M48 (only those who had fresh tissue collected at S00), the next priority is 5 mm fresh tissue processed at bedside for the Replication substudy.

- The next priority is to place 3 mm (or larger) cores of biopsies into cryovials, flash-frozen in liquid nitrogen at the bedside, and later transferred to dry ice (-80^{0}C or colder) for long-term storage.

- The last priority is to embed any remaining liver tissue (preferably 4 mm) into Tissue-Tek OCT prior to freezing in liquid nitrogen.

SECTION B: LIVER TISSUE SPECIMENS

B1. Record whether liver tissue was collected for the S00, M24, or M48 study visit.

- If the biopsy was performed at the clinical center, the answer is YES. Circle "1" and continue to question B2.

- If the biopsy was performed at a hospital or institution that is not affiliated with HALT-C, the answer is YES. Circle "1" and add a written comment stating, "collected at an non-HALT-C facility". The data entry person should add this written statement as a field level comment in the DMS. In the medical record notes, indicate the particulars of name of the facility and accession # of liver biopsy for reference. To maintain HIPAA compliance, hospital name or accession # should not be data entered in the DMS.

- If you know for certain that this liver biopsy will never be done either at the clinical site or an outside hospital, the answer is NO. Circle "2" and the form is complete and should be data entered in the DMS. Add a written comment to the form explaining why the biopsy could not be completed (for example, "PI discretion because of low platelets"). The data entry person should add this written statement as a form level comment in the DMS.

B2. Record the actual date of the liver biopsy procedure using MM/DD/YYYY format.

- If the M24 or M48 biopsy is collected at a time point outside of the M24 or M48 visit window, you should still record the actual date of the liver biopsy procedure. Form #14 should be entered under the M24 or M48 visit.

  - Example: A patient missed the M24 study visit in December 2003. The patient completed a liver biopsy on October 1, 2004 during the M33 visit window. Form # 14 should be completed with Question A3 (visit number) = M24 and Question B2 (date of biopsy) = 10/01/2004. Form #14 should be data entered in the DMS under the M24 visit, not under the M33 visit.

B3. Record the size in millimeters of the entire liver specimen.

- Enter the 2-digit number for the millimeters in the two spaces provided.
- One millimeter = "01", two millimeters = "02", one centimeter = "10".

B4-8. The entire liver sample will be used for a variety of study related "Purposes". Collection and handling of each specimen will depend on the purpose for that specimen. Use one row (B4 through B8) for each liver sample purpose and/or collection method.

In Columns a-d, record the following information on each liver specimen.

Column a: Size of Specimen
Record the size of this section of the liver specimen.
- Enter the 2-digit number for the millimeters in the two spaces provided.
- One millimeter = "01", two millimeters = "02", one centimeter = "10".

Column b: Purpose of Specimen
Record the purpose of this section of the liver specimen. Use one of the codes for Purpose listed in the code box below.

Column b code	*Purpose of Specimen*
1	Pathology for Main Trial
2	Repository
3	Replication AS
4	Iron AS
5	Immunology AS
6	Serum Fibrosis Marker AS

Column c: How Collected / Handled
Record how this section of the liver specimen was collected and handled. Use one of the codes for How Collected/Handled listed in the code box below.

Column c code	*How Collected / Handled*
1	Room temperature - fresh
2	Room temperature - for pathology
3	Snap frozen in liquid nitrogen
4	Snap frozen in OCT medium and liquid nitrogen

Column d: Initials
Record the initials of the physician performing the biopsy and collecting this section of the liver specimen.

Example A. Patient participating in Main Trial only:

	a.	b.	c.	d.
	Size of Specimen	**Purpose of Specimen**	**How Collected/Handled**	**Initials**
B4.	20 mm	1 = Path for Main Trial	2 = Room temp - for path	A B C
B5.	4 mm	2 = Repository	3 = Snap frozen liquid nitrogen	A B C
B6.	4 mm	2 = Repository	4 = Snap frozen in OCT	A B C
B7.				
B8.				

Example B. Patient participating in Main Trial and Immunology CTL AS and Replication AS:

	a.	b.	c.	d.
	Size of Specimen	**Purpose of Specimen**	**How Collected/Handled**	**Initials**
B4.	20 mm	1 = Path for Main Trial	2 = Room temp - for path	A B C
B5.	4 mm	2 = Repository	3 = Snap frozen liquid nitrogen	A B C
B6.	4 mm	5 = Immunology AS	1 = Room temp - fresh	A B C
B7.	4 mm	3 = Replication AS	4 = Snap frozen in OCT	A B C
B8.	4 mm	2 = Repository	4 = Snap frozen in OCT	A B C

Example C. Patient participating in Main Trial and Iron AS:

	a.	b.	c.	d.
	Size of Specimen	**Purpose of Specimen**	**How Collected/Handled**	**Initials**
B4.	20 mm	1 = Path for Main Trial	2 = Room temp - for path	A B C
B5.	4 mm	2 = Repository	3 = Snap frozen liquid nitrogen	A B C
B6.	4 mm	4 = Iron AS	1 = Room temp - fresh	A B C
B7.	4 mm	2 = Repository	4 = Snap frozen in OCT	A B C
B8.				

Labels and Sequence Numbers:

Sequence Number	**Purpose of Specimen**	**How Collected/Handled**	**Patients**
No labels	1 = Path for Main Trial	2 = Room temp - for pathology	All patients: stained and unstained slides
Labels #130 and #131	2 = Repository	3 = Snap frozen in liquid nitrogen	All patients
Label #132	2 = Repository	4 = Snap frozen in OCT	All patients (Note: for patients in Replication AS, seq. #320 has higher priority than seq. #132)
Label #320	3 = Replication AS	4 = Snap frozen in OCT	Sites 11, 12, 16, 17: Patients enrolled in Replication AS
Label #600	4 = Iron AS	1 = Room temp - fresh	Sites 11 & 15
Label #630	5 = Immunology AS	1 = Room temp - fresh	Sites 11 & 12: Patients enrolled in Immunology CTL AS
Label #633	5 = Immunology AS	1 = Room temp - fresh	Sites 16 & 17: Patients enrolled in Immunology CTL AS
No label (seq. #708)	6 = Serum fibrosis AS	3 = Snap frozen in liquid nitrogen	Site 18