NIDDK Liver Transplantation Database
MANUAL OF OPERATIONS (MOOP) DEFINITION

Purpose: To document the histological evaluation of a specimen of the transplanted liver, and the pathologic evaluation based on these findings. The rejection grade, as determined by the LTD grading scheme, should also be recorded.

Person(s) Responsible: LTD pathologist; LTD Clinical Coordinator.

Source(s) of Information: Liver specimen; pathology report.

General Instructions: This form is to be completed in the event a liver biopsy (protocol or otherwise) was performed, in the case of retransplantation for a failed allograft, or in the event the patient died and an autopsy was performed. In the case of a failed allograft or death, the PG form (Pathology for Failed Allografts and Autopsies) must also be completed. Because of the specific, technical nature of the information requested, both forms should be completed by the pathologist examining the specimen. However, the information requested in section IV of the PP Form can be provided by the data collector or clinical coordinator.

SURGICAL #

The surgical number is the number assigned to the tissue specimen at the transplant center. This number is not entered in the computer.

Completing Form: Enter the clinical center code (01, 02, etc.) before the hyphen, and the surgical number after the hyphen.

PATIENT ID

This is the number assigned to the patient upon entry in the LTD, and may not be changed.

Completing Form: Enter the clinical center code (01, 02, etc.) before the hyphen, and the assigned patient number after the hyphen. This information should be provided on the form by the clinical coordinator before the pathologist fills out the form.

TRANSPLANT NUMBER

This is the most recent transplant that the patient has received.

Completing Form: Enter the number of the most recent transplant for this patient.

DATE OF SPECIMEN

This is the date the specimen was obtained, not the date it was sent to pathology.

Completing Form: Enter the date the specimen was obtained as month, day and year.

I. SOURCE OF SPECIMEN

The source of specimen may be from a liver biopsy, protocol or otherwise; from a failed allograft or from an autopsy.

Completing Form: Check the appropriate category to indicate the source of the specimen.

II. HISTOLOGICAL EVALUATION

The histological evaluation section is meant to contain qualitative and semi-quantitative information on the histopathologic findings based on a subjective impression of the pathologist examining the post-transplant liver allograft specimens. Ideally, this form should be filled out by the single pathologist designated to interpret the post-transplant liver biopsies at the time that he is reviewing the specimens. It is designed in such a manner as to assist in the identification of rejection and other common post-

transplant complications. The following are meant to be a guideline to the integration of this form with biopsy interpretation.

II.1-2 The information regarding the presence of 4 or more portal tracts must be available, as well as the adequacy of the specimen.

II.3.1 Inflammation intensity in the portal tract is graded as to the overall relative intensity based on the most intensely involved portal tract. "None" is self-explanatory. "Severe" should be checked when the portal tracts are markedly expanding and there is inflammatory cell bridging (bridging necrosis) between adjacent portal tracts.

II.3.2 Bile duct inflammation/damage refers to the presence of inflammatory cells, if any, present within the basement membrane and/or lumen of bile ducts and/or bile duct damage. Damage should be manifest as biliary epithelial cell vacuolization, pyknosis, eosinophilia or frank luminal disruption.

II.3.3 Bile duct loss occurs when a portal tract is identified with an artery(iole) and contains no bile duct of a similar diameter within a surrounding area of 2-3 times the diameter of the artery. This finding is based on the observations of Nakanuma (Gastroenterology 76:1326-1332, 1979). The total number of portal tracts and those without bile ducts should be recorded.

II.3.4 Bile duct/cholangiolar proliferation refers to ducts (interlobular and septal) located within the connective tissue of the portal tract, or structures located at the edge of the portal tract.

II.4-5 Presence and type of arteritis is to be listed, as well as the presence of intimal fibrosis or xanthomatous inflammation of arterial or venous channels.

II.6 Subendothelial inflammation refers to mononuclear cells beneath the portal or central vein endothelium.

II.7-12 Lobular architectural and hepatocellular alterations: fibrosis, architectural disarray, inflammation, cholestasis and steatosis are to be noted and recorded in the appropriate categories.

Completing Form:

II.1-2 Check whether there were more than 4 portal tracts and whether the specimen was adequate.

II.3 For each of the listed categories of portal tract description, check the appropriate response. For "bile duct loss", if "yes" was checked, record also the total number of portal tracts and the number without ducts.

II.4-12 For each item, check the appropriate response.

III. PATHOLOGIC DIAGNOSIS(ES)

The diagnosis sections should be completed after gross (if applicable) and histologic evaluation of the specimen. More than one gross and microscopic histopathologic diagnosis may be (and often is) appropriate. In this case, the diagnoses are listed in their perceived order of importance as determined by the examining pathologist and numbered "1" through "4". "1" is perceived to be the most significant diagnosis followed by the other diagnoses in order of importance. It is recognized that in some instances a subjective judgement is necessary. Diagnoses listed in Section III are evident after microscopic examination alone.

The following were added to the form on February 21, 1991. Records prior to this date may have missing data for these questions.

- Preservation injury (III.2.2)
- Acute, chronic persistent, and chronic active categories were added to possibly/probably viral (III.3.1) and definitely viral (III.3.2) hepatitis.
- Sepsis (III.5.2), Possible recurrent disease (III.5.4), and possible drug reaction (III.5.6) were added to other diagnoses.

Completing Form: Record "1" through "4" in the perceived order of importance all the appropriate diagnosis(es) up to four diagnoses. If only one diagnosis is appropriate, record only "1" in the designated space. If the diagnosis is not listed, record the number beside "Other" and specify the type in 30 characters or less in the space provided. For "viral hepatitis" record the designated ranking under possibly/probably viral or definitely viral and check "acute", "chronic persistent" or "chronic active" for each if appropriate. If definitely viral hepatitis, check the appropriate virus based on morphology (ref. Appendix I).

IV.1 REASON FOR EVALUATION

This is the reason for which the biopsy was performed. If it was done for any reason other than a protocol biopsy, but coincided with the protocol biopsy schedule, it should be considered done for the other reason rather than for the protocol schedule.

Allowable windows for protocol timepoints changed several times throughout the course of the LTD. Timepoints are as follows:

	Prior to 2/12/91		On/After 2/12/91	
Day 0/1	0-1	days	0-1	days
Day 3	2-4	days	2-4	days
Week 1	4-14	days	4-12	days
Week 2	8-20	days	8-20	days
Week 3	15-28	days	13-35	days
Week 4	24-32	days	24-32	days
Week 5	31-39	days	31-39	days
Week 6	38-46	days	38-46	days
Month 4	90-180	days	90-180	days
Year 1	10-14	months	10-14	months (changed in March, 1992 to 9-18 months)

Completing Form: Check only one reason for this biopsy. If "protocol biopsy" was checked, also check the appropriate protocol timepoint. If it was not done at a specified protocol timepoint, check "other".

IV.2 WAS THIS SPECIMEN A BIOPSY?

If the specimen obtained was from a biopsy, information on type of specimen, route of biopsy, and occurrence of complications is requested.

Completing Form: Check whether the specimen was from a biopsy. If "yes", check the type of specimen obtained and the route of the biopsy. If the type and/or route are not listed, check "Other" and specify in the space provided. Also check whether there was a complication as a result of the liver biopsy. If "yes", check the appropriate items from the list. If not included in the list, check "Other" and specify. Also check whether a transfusion, operation and/or hospitalization were necessary for this liver biopsy.

V. COMMENTS

Use this space for any other information that is pertinent to this histological evaluation that has not been recorded elsewhere in this form.

Completing Form: Check whether there are any comments to be made. If "yes" write in the comments that are pertinent to this evaluation process.

VI. REJECTION GRADE

The rejection grade was added to the form in October, 1991. Records prior to this time may have missing data for this question.

Grade according to LTD grading scheme.

A0 No Rejection

A Rejection without bile duct loss

1. Rejection infiltrate in some, but not all of the triads, confined within the portal spaces.
2. Rejection infiltrate involving all of the triads, with or without spillover into the lobule. No evidence of centrilobular hepatocyte necrosis, ballooning or dropout.
3. Infiltrate in some or all of the triads, with or without spillover into the lobule, w/wo inflammatory cell linkage of the triads, associated with centrilobular hepatocyte ballooning or necrosis and dropout.

B Rejection with Bile Duct Loss

1. Bile duct loss present, without centrilobular cholestasis, perivenular sclerosis or

hepatocyte ballooning or necrosis and dropout.

2. Bile duct loss present, with at least one of the following four findings: centrilobular cholestasis, perivenular sclerosis or hepatocellular ballooning or necrosis and dropout.
3. Bile duct loss present, with at least two of the following four findings: centrilobular cholestasis, perivenular sclerosis or hepatocellular ballooning or necrosis and dropout.

C Rejection, indefinite for bile duct loss

1. No complicating lobular changes.
2. Lobular changes including one of the following three findings; centrilobular cholestasis, perivenular sclerosis or hepatocellular ballooning or necrosis and dropout.

D Biopsy insufficient or unsuitable for diagnosis

Completing Form: Refer to the LTD grading scheme.

VII. PATHOLOGIST ID

Record the two digit site number and the initials of the pathologist.

APPENDIX I

VIRAL STUDIES: the presence of a virus within the graft should be documented by characteristic morphologic alterations combined with the presence of diagnostic inclusion bodies and/or immunoperoxidase studies.

The following is a source list of antibodies used to stain for the presence of viral antigens in tissue section.

Virus	Tissue Prep[1]	Source	(C)[2]	CAT #	Comments
Adenovirus	FR	Chemicon	M	MAB805	wide specificity
CMV	FX	Chemicon[3]	M	MAB810	early antigen
		Polyscience	P	15460	late cytoplasmic
Hepatitis B	FX				
-core		DAKO	P	B586	
-surface		DAKO	P	B560	
Herpes Simplex	FX				
-Type I		DAKO	P	B114	
-Type II		DAKO		B116	

1 FR = Requires frozen tissue in OCT compound.
FX = Formalin fixed paraffin embedded tissue.
2 (C) = clonality
3 Requires tissue digestion protease K Type XI (Sigma) for 15 minutes
Color development for immune staining is achieved using an Avidin-Biotin Complex with DAB or AEC chromogen.

DIGESTION PROCEDURES

Protease K (Sigma) - Used for CMV only.

Use 1.0 mg. protease K per 10 ml Tris buffer (or PBS) (with/out EDTA).
Put 3-4 drops on sections for 5 minutes at room temperature.
Wash off with cool running tap water for 5 minutes.