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Automated Tissue Fixation with 10% NBF (Leica)

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ABSTRACT

Human tissue specimens are frequently formalin-fixed and paraffin-embedded (FFPE) for histological staining, pathological review, and archival purposes. This is the routine fixation protocol used by the Specialized Histopathology (SHP) Core and the Department of Pathology at Brigham and Women's Hospital. The protocol uses the Leica ASP300 S Fully Enclosed Tissue Processor to automate fixation.

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GUIDELINES

When fixatives other than 10% NBF are used, the case will be discussed with the investigator by management to ensure the best possible result.

MATERIALS TEXT

Materials & Equipment

- Leica ASP300 S Fully Enclosed Tissue Processor
- 10% Neutral buffered formalin
- Ethyl Alcohol
- Xylene
- Paraffin wax

BEFORE STARTING

Fixation times can range from 8 to 72 hrs depending on size, thickness, and type of the sample. It is not

recommended to fix the sample for longer time periods, as this can have deleterious effects for various assays.

Automated Tissue Fixation with 10% NBF (Leica)

- 1 Program the Leica ASP300 S Fully Enclosed Tissue Processor with either the short program or long program depending on size, thickness, and type of sample.

2 Formalin Short Program

A	B	C	D
Station/Reagent	Time	Temp	P/V
10% NBF	00:12 min	40	Y
70% Ethyl Alcohol	00:15 min	40	Y
95% Ethyl Alcohol	00:15 min	40	Y
95% Ethyl Alcohol	00:20 min	40	Y
100% Ethyl Alcohol	00:20 min	40	Y
100% Ethyl Alcohol	00:20 min	40	Y
Xylene	00:20 min	40	Y
Xylene	00:30 min	40	Y
Paraffin Wax 1	00:05 min	60	Y
Paraffin Wax 2	00:10 min	60	Y
Paraffin Wax 3	00:15 min	60	Y

Table 1. Formalin short program.

3 Formalin Long Program

A	B	C	D
Station/Reagent	Time	Temp	P/V
10% NBF	02:00 hr	40	Y
70% Ethyl Alcohol	00:45 min	40	Y
95% Ethyl Alcohol	00:45 min	40	Y
95% Ethyl Alcohol	00:45 min	40	Y
100% Ethyl Alcohol	00:30 min	40	Y
100% Ethyl Alcohol	01:00 min	40	Y
100% Ethyl Alcohol	30 min	40	Y
Xylene	30 min	40	Y
Xylene	30 min	40	Y
Paraffin Wax 1	30 min	60	Y
Paraffin Wax 2	30 min	60	Y
Paraffin Wax 3	30 min	60	Y

Table 2. Formalin long program.