

ThinPrep® Stain Protocol using Rinse II and Bluing II Solutions on the LEICA ST5020 Multistainer

Use this protocol ONLY when ThinPrep Rinse II and Bluing II Solutions (P/Ns ASY-04875, ASY-04876) are components of the ThinPrep Stain.

Use this protocol for staining slides fixed with 95% alcohol.

Maintain bath heights to completely cover the slides at full immersion. (Note: Bath fill volume = 450 ml)

Caution: Change the Distilled / Instrument Feed Water bath at Station 32 after every **four** racks of slides.

Required Setup Parameters: Dips = 9, Dip Amplitude = 2, Lift speed = 1					
Step	Station	Reagent	Duration	Exact	Dip
LOAD	36	70% Reagent or Ethyl alcohol	----	----	----
1	35	50% Reagent or Ethyl alcohol	00:01:00	no	yes
2	34	Distilled or Instrument Feed Water (IFW) ¹	00:01:00	no	yes
3	33	ThinPrep Nuclear Stain	00:07:00	yes	yes
4	32	Distilled or Instrument Feed Water ¹ (Replace after every 4 racks.)	00:00:10	yes	yes
5	31	ThinPrep Rinse II Solution	00:01:00	yes	yes
6	30	Distilled or Instrument Feed Water ¹	00:00:30	no	yes
7	29	ThinPrep Bluing II Solution	00:00:30	yes	yes
8	28	Distilled or Instrument Feed Water ¹	00:00:30	no	yes
9	27	Distilled or Instrument Feed Water ¹	00:00:30	no	yes
10	24	95% Reagent or Ethyl alcohol	00:00:30	no	yes
11	23	ThinPrep Orange G Solution	00:02:00	yes	yes
12	22	95% Reagent or Ethyl alcohol	00:00:15	yes	yes
13	21	95% Reagent or Ethyl alcohol	00:00:15	yes	yes
14	20	ThinPrep EA Solution	00:04:00	yes	yes
15	19	95% Reagent or Ethyl alcohol	00:01:00	yes	yes
16	18	95% Reagent or Ethyl alcohol	00:01:00	yes	yes
17	17	100% Reagent or Ethyl alcohol	00:00:30	no	yes
18	16	100% Reagent or Ethyl alcohol	00:00:30	no	yes
19	15	100% Reagent or Ethyl alcohol	00:00:30	no	yes
20	14	Xylene or other Hologic approved clearing agent ²	00:01:00	no	yes
21	13	Xylene or other Hologic approved clearing agent ²	00:03:00	no	yes
UNLOAD	25 & 26	Xylene or other Hologic approved clearing agent ²	----	----	----

Remove slides to a separate clearing bath then coverslip with the appropriate Hologic approved mountant. ²

¹ Hologic specification for IFW: ≥ 1.0 megohm-cm Resistivity OR ≤ 1.0 μ Siemens/cm Conductivity (Refer to Clinical and Laboratory Standards Institute (CLSI) document C3-A4, 2006)

² See ThinPrep Stain User's Manual, Section 1, "STAINING" or contact Hologic for current list of Hologic approved clearing agents and mounting media.



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Solution Stations

Leica ST5020 Multistainer

Using ThinPrep Rinse II and Bluing II Solutions

For slides fixed with 95% alcohol

Bath Station Number→ 27

Solution→ 50% Alcohol

Step Number→ 9

								O37	O38	O39	O40
1	2	3	4	5	6	7	8	9	10	11	12
13	14	15	16	17	18	19	20	21	22	23	24
Xylene	Xylene	100% Alcohol	100% Alcohol	100% Alcohol	95% Alcohol	95% Alcohol	EA Solution	95% Alcohol	95% Alcohol	Orange G Solution	95% Alcohol
21	20	19	18	17	16	15	14	13	12	11	10
25	26	27	28	29	30	31	32	33	34	35	36
Xylene	Xylene	Distilled Water	Distilled Water	Bluing II Solution	Distilled Water	Rinse II Solution	Distilled Water	Nuclear Stain	Distilled Water	50% Alcohol	70% Alcohol
UNLOAD	UNLOAD	9	8	7	6	5	4	3	2	1	LOAD