

# ThinPrep® Stain Protocol for the LEICA ST5020 Multistainer using 95% alcohol fixed slides

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

**Caution:** Change the Distilled / Instrument Feed Water bath at Station 32 after every **four** racks of slides. Maintain bath heights to completely cover the slides at full immersion. (Note: Full bath volume is 450 ml.)

<b>Required Setup Parameters:</b> Dips = 9, Dip Amplitude = 2, Lift speed = 1					
Step	Station	Reagent	Duration	Exact	Dip
----	Load	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) <sup>1</sup>	00:01:00	no	yes
3	33	<b>ThinPrep Nuclear Stain</b>	00:07:00	yes	yes
4	32	Distilled or Instrument Feed Water <sup>1</sup> (Replace after every 4 racks.)	00:00:10	yes	yes
5	31	<b>ThinPrep Rinse Solution</b>	00:01:00	yes	yes
6	30	Distilled or Instrument Feed Water <sup>1</sup>	00:00:30	no	yes
7	29	<b>ThinPrep Bluing Solution</b>	00:00:30	yes	yes
8	28	Distilled or Instrument Feed Water <sup>1</sup>	00:00:30	no	yes
9	27	50% Reagent or Ethyl alcohol	00:00:30	no	yes
10	24	95% Reagent or Ethyl alcohol	00:00:30	no	yes
11	23	<b>ThinPrep Orange G Solution</b>	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	<b>ThinPrep EA Solution</b>	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 <sup>2</sup>	00:01:00	no	yes
21	13	Xylene or other Hologic approved clearing agent <sup>2</sup>	00:03:00	no	yes
22	Exit	Xylene or other Hologic approved clearing agent <sup>2</sup>	----	----	----

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

<sup>1</sup> Hologic specification for IFW:  $\geq 1.0$  megohm-cm Resistivity OR  $\leq 1.0$   $\mu$ Siemens/cm Conductivity (Refer to Clinical and Laboratory Standards Institute (CLSI) document C3-A4, 2006)

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



Hologic, Inc. • 250 Campus Drive • Marlborough, MA 01752 USA • 1-800-442-9892 • www.hologic.com  
Hologic BV • Da Vincilaan 5 • 1930 Zaventem • Belgium

**Solution Stations**  
**Leica ST5020 Multistainer**  
 For staining slides fixed with 95% alcohol

Bath Station Number→	27
Solution→	<b>50% Alcohol</b>
Step Number→	<b>9</b>

								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
<b>Xylene</b>	<b>Xylene</b>	<b>100% Alcohol</b>	<b>100% Alcohol</b>	<b>100% Alcohol</b>	<b>95% Alcohol</b>	<b>95% Alcohol</b>	<b>ThinPrep EA</b>	<b>95% Alcohol</b>	<b>95% Alcohol</b>	<b>ThinPrep Orange G</b>	<b>95% Alcohol</b>
<b>22</b>	<b>21</b>	<b>20</b>	<b>19</b>	<b>18</b>	<b>17</b>	<b>16</b>	<b>15</b>	<b>14</b>	<b>13</b>	<b>12</b>	<b>11</b>
25	26	27	28	29	30	31	32	33	34	35	36
<b>Xylene (Unload)</b>	<b>Xylene (Unload)</b>	<b>50% Alcohol</b>	<b>Distilled Water</b>	<b>ThinPrep Bluing</b>	<b>Distilled Water</b>	<b>ThinPrep Rinse</b>	<b>Distilled Water</b>	<b>ThinPrep Nuclear</b>	<b>Distilled Water</b>	<b>50% Alcohol</b>	<b>70% Alcohol (Load)</b>
<b>23</b>	<b>23</b>	<b>10</b>	<b>9</b>	<b>8</b>	<b>7</b>	<b>6</b>	<b>5</b>	<b>4</b>	<b>3</b>	<b>2</b>	<b>1</b>