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Thermo Scientific Richard-Allan Scientific Cytology Specimen Management System Instructions for Use

For in vitro diagnostic use.

For staining cytology specimens.

Thermo Scientific™ Richard-Allan Scientific™ Cytology Specimen Management System (C-SMS) is a new approach to cytology staining. Color intensity and hue are under complete control of the user, for both gynecological and non-gynecological specimens. The system requires fewer stations and less time than most cytology staining programs. In addition, the cytoplasmic stain is a single solution of unique formulation that produces colors identical to those achieved with conventional Papanicolaou stains. All reagents are quality controlled to assure consistent results.

Specimen Collection and Preservation

Optimally stained slides can only be achieved with properly collected and preserved material. Specimens must be fresh, applied in a thin film over glass or membrane filter and fixed immediately. Fixation may be accomplished at the time of collection.

Spray fixatives, such as Thermo Scientific™ Richard-Allan™ Scientific Fix-Rite™ 2 should be applied before the slide becomes air-dried. Excessive amounts of spray may cause uneven staining if not completely removed.

Basic Instruction for Staining

The general format for staining with C-SMS is given in Table 1. The five stations with variable times are critical steps that allow control over hue and intensity.

Nine different staining programs are provided in Table 2 to accommodate varied personal preferences among cytologists for intensity and hue. Because basic names for colors are not precisely defined nor uniformly applied, these programs are to be used as relative guidelines only.

Once a basic staining program has been selected from Table 2 and tried, alterations in the results can be made by changing one or more of the times given for the five critical steps in Table 2. Please refer to Table 3 for detailed instructions.

Thermo Scientific Richard-Allan Scientific Reagents

The Cytology Specimen Management System (C-SMS) consists of the following reagents:

Thermo Scientific™ Richard-Allan Scientific™ Fix-Rite 2
Cytology fixative is a non-aerosol, spray packaged in a pump container which emits no fluorocarbons. Consistent chemical composition guaranteed. Carbowax fixatives must be fully removed prior to staining by immersing fixed slides in 95% alcohol for 10-15 minutes.

Thermo Scientific™ Richard-Allan Scientific™ Hematoxylin 1
A rapid, progressive nuclear stain; when used with Bluing Reagent, it produces extremely sharp contrast and exceptional nuclear detail. The dye powder used in formulating this stain is certified by the Biological Stain Commission.

Thermo Scientific™ Richard-Allan Scientific™ Hematoxylin 7211
Hematoxylin 7211 is a unique product producing results similar to that of Gill or Harris Hematoxylins. A distinct characteristic of 7211 is that there is no affinity for acid mucopolysaccharide (mucin) staining. Slides stained with Hematoxylin 7211 are crisp and there is clear nuclear chromatin delineation.

Thermo Scientific™ Richard-Allan Scientific™ Clarifier 1
Clarifier 1™ renders cells more transparent and achieves more transparent green and red hues in cytoplasm.

Thermo Scientific™ Richard-Allan Scientific™ Bluing Reagent
Significantly enhances nuclear detail when used in conjunction with Thermo Scientific Richard-Allan Scientific Hematoxylin.

Thermo Scientific™ Richard-Allan Scientific™ Cyto-Stain
Possesses a unique chemical formulation developed for the Cytology Specimen Management System producing the full range of familiar colors; offers increased convenience in a single solution counterstain while providing complete control of cytoplasmic staining intensity. All dyes used in this stain are certified by the Biological Stain Commission. A minor difference between Cyto-Stain™ conventional pap staining is that the cilia in bronchial washings stains blue-green with Cyto-Stain rather than red as seen in conventional pap staining.

Thermo Scientific™ Richard-Allan Scientific™ Alcohols
Allows rapid removal of fixative and controls differentiation of Cyto-Stain.

Thermo Scientific™ Richard-Allan Scientific™ Xylene or Clear-Rite 3™
Allows rapid clearing of specimens prior to coverslipping.

Thermo Scientific™ Richard-Allan Scientific™ Mounting Medium
Quick-drying, neutral mounting medium with a refractive index of 1.5.

Mode of Action

Thermo Scientific Richard-Allan Scientific Hematoxylin 1 and 7211 are oxidized to form hematein, a dye lake is formed with Aluminum Sulfate or Aluminum Ammonium Sulfate to produce cationic complexes, $[Al\ OH]^{++}[Al(OH)_2]^{+}$, in solution. The mordant (Aluminum Sulfate or Aluminum Ammonium Sulfate) attaches itself to phosphoryl groups in nucleic acids and possesses little affinity for anionic groups of the cytoplasmic protein. Thermo Scientific Richard-Allan Scientific Clarifier 1 acts as a selective, gentle reagent to render cells more transparent. Additionally, it may be used to achieve transparent green and red hues in cytoplasm. Reducing or eliminating exposure of slides to Clarifier 1 produces greater blue cytoplasmic hues.

Thermo Scientific Richard-Allan Scientific Cyto-Stain acts as a single-solution counterstain. Selective staining is accomplished through the action of a mordant and through differential affinities of the dyes for various tissue components. Final cytoplasmic colors are influenced by prior action of Clarifier 1.

The two 95% alcohol rinses after the Cyto-Stain function to remove Eosin from certain components, allowing greater differentiation of red and orange colors.

Table 1

Generalized Staining Program

Station	Solution	Time
1	95% Alcohol	3 minutes
2	95% Alcohol	2 minutes
3	Deionized or distilled water	30 seconds with agitation
4	Deionized or distilled water	30 seconds
5	Hematoxylin 1 or 7211	Select time from Table 2
6	Deionized or distilled water	15 seconds with agitation
7	Clarifier 1	Select time from Table 2
8	Deionized or distilled water	30 seconds
9	Bluing Reagent	30 seconds
10	Deionized or distilled water	30 seconds with agitation
11	95% Alcohol	30 seconds
12	Cyto-Stain	Select time from Table 2
13	95% Alcohol	Select time from Table 2
14	95% Alcohol	Select time from Table 2
15	100% Alcohol	30 seconds with agitation
16	100% Alcohol	30 seconds
17	100% Alcohol	30 seconds
18	Xylene	30 seconds with agitation
19	Xylene	30 seconds
20	Xylene	30 seconds

Table 2

The Five Critical Steps of the Cytology Specimen Management System

Hue	Intensity	Step 5 Hematoxylin 1 or 7211	Step 7 Clarifier 1	Step 12 Cyto-Stain	Step 13 95% Alcohol	Step 14 95% Alcohol
Blue	Dark	120	0	120	0	0
	Medium	90	0	75	30	30
	Pastel	60	0	45	60	60
Blue-Green	Dark	60	60	120	0	0
	Medium	45	45	75	30	30
	Pastel	30	30	45	60	60
Green	Dark	30	120	120	0	0
	Medium	20	90	75	30	30
	Pastel	15	60	45	60	60
	Time in Seconds					

Table 3

Fine Tuning the Cytology Specimen Management System

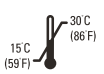
Desired Trait	Step 5 Hematoxylin 1 or 7211	Step 7 Clarifier 1	Step 12 Cyto-Stain	Step 13 95% Alcohol	Step 14 95% Alcohol
Nuclei lighter, cytoplasm unchanged	-	(-)			
Nuclei lighter, cytoplasm more green	-	+			
Nuclei lighter, cytoplasm blue	-	-			
Nuclei unchanged, cytoplasm more green		+			
Nuclei unchanged, cytoplasm more blue		-			
Nuclei darker, cytoplasm unchanged	+	+			
Nuclei darker, cytoplasm more green	+	(+)			
Nuclei darker, cytoplasm more blue	+	-			
Cytoplasm lighter, more red			-	-	-
Cytoplasm lighter, same hues			-		
Cytoplasm lighter, less red			-	+	+
Cytoplasm more red				-	-
Cytoplasm less red, more orange				+	+
Cytoplasm darker, more red			+	-	-
Cytoplasm darker, same hues			+		
Cytoplasm darker, less red			+	+	+
More red nucleoli	-	+		-	-
Less blue on membrane filter	-	+			
Less red on membrane filter				+	+
Less blue on mucus	-	+			
Less stain in thick areas	-	+	-	+	+

Key
+ = Increased Time - = Decreased Time ( )= Slight Change

Order Information

Product	Size	Qty.	REF
Clear-Rite 3	1 gal.	4/cs.	6901
Clear-Rite 3	5 gal.	1/cs.	6905
Clear-Rite 3	55 gal.	1/cs.	6955
Eosin Y	5 L	1/cs.	7111L
Hematoxylin 7211	1 pt.	4/cs.	7211
Hematoxylin 7211	5 L	1/cs.	7211L
Hematoxylin 7212	5 L	1/cs.	7212L
Hematoxylin 1	1 pt.	4/cs.	7221
Hematoxylin 1	5 L	1/cs.	7221L
Hematoxylin 2	5 L	1/cs.	7231 L
Bluing Reagent	1 gal.	1/cs.	7301
Bluing Reagent	5 L	1/cs.	7301L
Bluing Reagent	1 gal.	4/cs.	7341
Clarifier 1	1 gal.	1/cs.	7401
Clarifier 2	5 L	1/cs.	7402L
Clarifier 1	1 gal.	4/cs.	7441
Cyto-Stain	1 gal.	1/cs.	7501R
Cyto-Stain G	1 gal.	1/cs.	7501G
Cyto-Stain	1 pt.	4/cs.	7511
Fix-Rite 2 (pump spray)	4 oz.	12/cs.	76150

Anatomical Pathology



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