

### Product Description

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Alcian Blue is a dye used to detect cell chondrogenesis and it stains the sulfated proteoglycan in cartilage tissue. The Alcian Blue Staining Kit contains 1% Alcian Blue Solution, 0.1% Nuclear Fast Red in convenient, ready-to-use solutions. At pH 2.5, Alcian Blue stains acid mucopolysaccharides and shows a blue color, while Nuclear Fast Red stains nuclei pink to red and the cytoplasm pale pink [1].

### Kit Components

Cat. No.	# of vials	Name	Quantity	Storage
8378a	1	1% Alcian Blue Solution	100 mL	Room temperature
8378b	1	0.1% Nuclear Fast Red	100 mL	Room temperature
8378c	1	3% Acetic Acid	100 mL	Room temperature
8378d	1	Xylene Substitute	100 mL	Room temperature

### Materials Supplied by User

Formaldehyde-fixed and paraffin-embedded tissue sections  
Ethanol (100%, 95%, 70%, 50%)  
Deionized H<sub>2</sub>O (diH<sub>2</sub>O)

### Product use

ABlue is for research use only. It is not approved for human or animal use, or for application in clinical or *in vitro* diagnostic procedures.

### Shipping

Room temperature.

### References

[1] Steedman, H.F. (1950) Alcian blue 8GS: A new stain for mucin. *Quarterly Journal of Microscopic Science*, Vol 91, p477-479.

### Procedures

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1. Deparaffinize and hydrate slides:

- 1) Deparaffinize the tissue sections in Xylene Substitute (Cat. #8378d), 3 changes of 5 minutes per change.
- 2) Hydrate in 100% ethanol, 2 changes of 2 minutes per change.
- 3) Hydrate in 95% ethanol, 2 changes of 2 minutes per change.
- 4) Hydrate in 70% ethanol for 2 minutes.
- 5) Hydrate in 50% ethanol for 2 minutes.

- 6) Rinse in diH<sub>2</sub>O for 5 minutes.
2. Incubate in 3% Acetic Acid (Cat. #8378c) for 3 minutes.
2. Stain in 1% Alcian Blue Solution, pH 2.5 (Cat. #8378a) for 30-60 minutes.
3. Wash in running tap water for 2 minutes and rinse in diH<sub>2</sub>O.
4. Stain in 0.1% Nuclear Fast Red (Cat. #8378b) for 10-20 minutes.
5. Wash in running tap water for 1 minute and rinse in diH<sub>2</sub>O for 2 minutes.
6. Dehydrate and clear slides:
  - 1) Dehydrate in 95% ethanol, 2 changes of 2 minutes per change.
  - 2) Dehydrate in 100% ethanol, 2 changes of 2 minutes per change.
  - 3) Clear the tissue sections in Xylene Substitute (Cat. #8378d), 2 changes of 2 minutes per change.
7. Mount the tissue sections and observe under microscope.

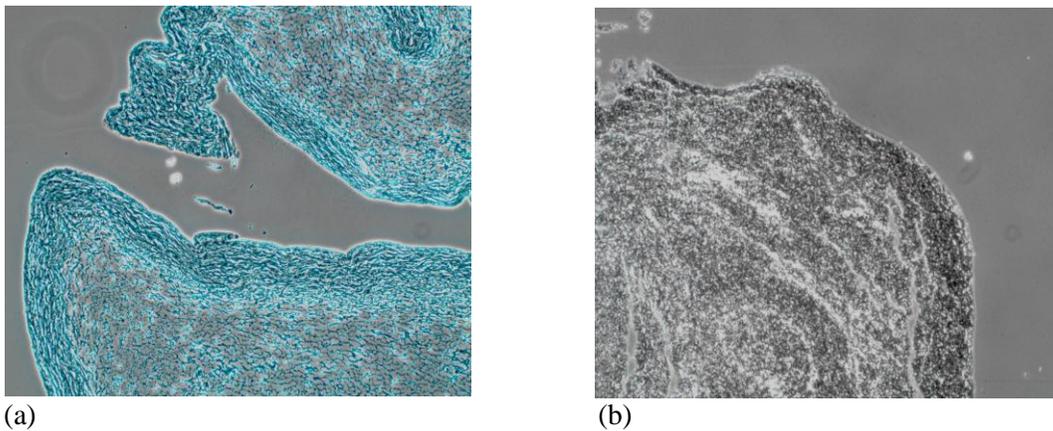


Figure 1. (a) HDF-f were cultured as pellets in complete MSC Chondrogenic Differentiation Medium (MCDM, Cat. # 7551) for 50 days. The pellets were fixed in 4% paraformaldehyde and sectioned. Alcian Blue staining demonstrated the presence of cartilage in cells (Magnification: 10X).

(b) Human Dermal Fibroblasts-fetal (HDF-f, Cat. #2300) were cultured as pellets in growth medium, complete Fibroblast Medium (FM, Cat. #2301) for 50 days. The pellets were fixed in 4% paraformaldehyde and sectioned. Alcian Blue staining was not detected (Magnification: 10X).