

Immunofluorescence Labeling Protocol for Cells

STEP		TEMPERATURE	TIME
1	Warm-up PBS (Phosphate Buffered Saline), Cello™-IF, Primary Antibody Solution, Secondary Antibody Solution	37 °C	10 min
2	Aspirate medium, Fix the sample (i.e., 4% Paraformaldehyde)	37 °C	10 min
3	Incubate with Cello™-IF	37 °C	3X, 5min
4	Incubate with Primary Antibody Solution in Cello™-IF	37 °C	1 hr-1,5 hr
5	Incubate with Cello™-IF	37 °C	3X, 5 min
6	Incubate with Secondary Antibody Solution in Cello™-IF Keep in dark	37 °C	30 min-1hr
7	Incubate with PBS , Keep in dark	37 °C	3X, 5 min
8	Cover with Mounting Medium*, Keep in dark	RT	
	Refrigerate until microscopic examination	4 °C	

TOTAL : 2,5-3,5 hrs.

**Mounting Medium: 1,2 uL Hoechst or DAPI+675 uL PBS + 675 uL Glycerol

NOTES:

-It is highly recommended to use glass coverslips or high-optical quality cell culture vessels (i.e., glass-bottomed dish, multi-chamber slide, Cello™-M) for growing, labeling, and examining the cells under the microscope.

-For long-term storage of the labeled specimen, seal the coverslip with nail polish, surround the glass bottomed-dish with parafilm, or close tightly the lid of Cello™-M.

-Glycerol can also be used as mounting medium without nuclear stain (i.e., Hoechst, DAPI, etc.)