



Catalog Number:	MO25034	Host:	Mouse
Product Type:	Mouse Monoclonal IgM Purified. Clone: TRA-1-81.	Species Reactivity:	Human
Immunogen Sequence:	Human embryonal carcinoma cell line 2102Ep.	Format:	Liquid with Tris-glycine, 150mM NaCl and 0.05% Sodium Azide. Concentration 1 mg/ml.
Applications:	Immunohistochemistry: * Immunofluorescence: 1:50-1:200 Immunoprecipitation: * Western Blot: *		
	*Dilutions listed as a recommendation. Optimal dilution should be determined by investigator.		
Storage:	Store frozen. Aliquot as undiluted antisera and immediately place at -20°C. Antisera may have become trapped in top of vial during shipping. Centrifugation of vial is recommended before opening. Stable for at least 6 months at -20°C. Repeated freeze/thaw cycles compromise the integrity of the antibody.		

Application Notes

This antibody is useful for Immunofluorescence, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation and Western Blot.

Immunofluorescence:

Culture cells to appropriate density in 35 mm culture dishes or 6-well plates.

1. Remove culture medium and add 10% formalin to the dish. Fix at room temperature for 30 minutes.
2. Remove the formalin and add ice cold methanol. Incubate for 5-10 minutes.
3. Remove methanol and add washing solution (i.e. PBS). Be sure to not let the specimen dry out. Wash three times for 10 minutes.
4. To block nonspecific antibody binding incubate in 10% normal goat serum from 1 hour to overnight at room temperature.
5. Add primary antibody at appropriate dilution and incubate at room temperature from 2 hours to overnight at room temperature.
6. Remove primary antibody and replace with washing solution. Wash three times for 10 minutes.
7. Add secondary antibody at appropriate dilution. Incubate for 1 hour at room temperature.
8. Remove antibody and replace with wash solution, then wash for 10 minutes. Add Hoechst 33258 to wash solution at 1:25,000 and incubate for 10 minutes. Wash a third time for 10 minutes.
9. Cells can be viewed directly after washing. The plates can also be stored in PBS containing Azide covered in Parafilm™. Cells can also be cover-slipped using Fluoromount, with appropriate sealing.

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Image: TRA-1-81 staining of methanol-fixed NTERA-2 cl.D1 cells showing membrane localization.

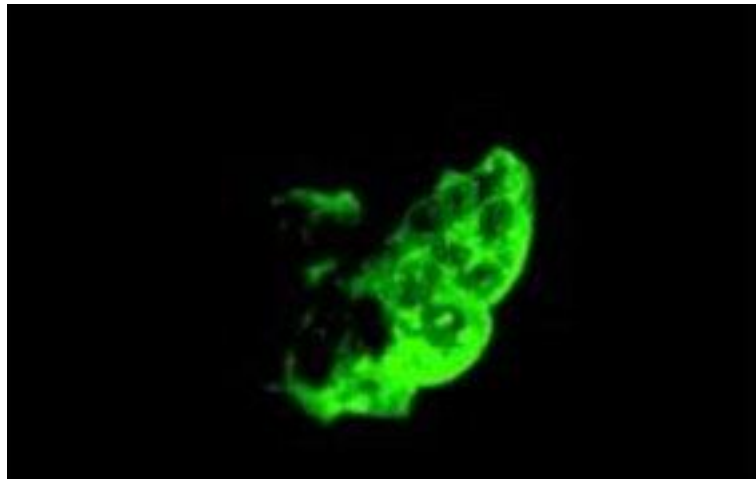
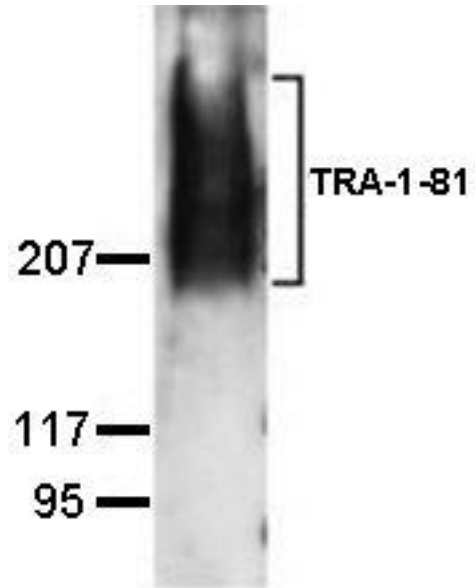


Image: TRA-1-81 western blot analysis of expression in NTERA-2 cl.D1 whole cell lysate.



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