exchange for purified Rab9–GDI complexes, late endosome membranes, or mixtures of these components, using a filtration assay (Fig. 2*a*). As expected, GDI blocked GTP-$\gamma$S binding to $Rab9_{GDP}$-GDI complexes over a 60-min incubation. MPR-enriched membranes showed some binding of GTP-$\gamma$S, probably due to other GTP-binding proteins. However, addition of purified Rab9-GDI complexes to MPR-enriched membranes led to a significant increase in membrane-associated $^{35}$S-GTP-$\gamma$S. The overall kinetics of Rab9 membrane association and Rab9-dependent $^{35}$S-GTP-$\gamma$S binding were very similar (Fig. 2*b*). Nevertheless, at the earliest time points $^{35}$S-GTP-$\gamma$S binding lagged behind the association of $Rab9_{GDP}$ with membranes. In addition, some GDI was initially detected on the membranes. These data suggest that $Rab9_{GDP}$-GDI complexes associate transiently with membranes; after a short lag, GDI is released, GTP binds and Rab9 becomes stably associated with the membrane.

Rab proteins may possess distinct, saturable, target membrane-specific receptors[1]. Alternatively, a protein catalysing nucleotide exchange could be localized to specific membrane compartments; after the release of GDI, a doubly prenylated Rab protein could jump into the adjacent membrane in its GTP-bound conformation. To distinguish between these possibilities, we examined whether the initial rate and/or extent of Rab recruitment were saturable.

Both the initial rate (Fig. 3*a*) and the overall extent (Fig. 3*b*) of Rab recruitment were saturable, but only at a ~50-fold excess of Rab protein relative to endogenous levels. Recruited Rab proteins were stably associated with the targets because, unlike a significant fraction of membrane-recruited ADP ribosylation factor (ARF)[19], they could not be dislodged by up to 4 mM phosphatidylcholine liposomes at 0 °C (~750-fold excess over endosomal membrane lipid). The high capacity of membranes for Rab9 protein was consistent with the finding that 50-fold overexpression of Rab proteins does not lead to their mislocalization[4,11]. These data strongly suggest that Rab proteins associate with a relatively abundant downstream target protein; nucleotide exchange/GDI displacement appear to be rate-limiting for targeting, and the saturable initial rate may reflect this membrane-catalysed process.

We have shown that prenylated $Rab9_{GDP}$ can be transferred to its specific membrane target when presented by GDI. Rab9 recruitment is accompanied by nucleotide exchange. Comparison of the rates of GDP release and GTP binding observed here with those measured for free Rab9 protein[20] suggest that Rab recruitment may involve a catalytic GDI-displacement factor (GDF), rather than just a conventional nucleotide exchange factor (GDS)[16,21,22] that would enhance the intrinsic rate of GDP release from Rab9 protein. Rab proteins, with GTP bound, would then associate stably with another downstream protein, perhaps a component of the SNARE complex[23], to catalyse vesicle targeting and/or fusion. □

Received 6 January; accepted 14 March 1994.


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ACKNOWLEDGEMENTS. We thank members of our laboratory for their comments, and R. Roth for anti-protein disulphide isomerase antibodies. This research was funded by a research grant to S.R.P. from the NIH, a postdoctoral fellowship of the Swiss NSF to T.S., and a predoctoral fellowship of the US NSF to A.D.S.

## ERRATUM

# Dynamic response of Jupiter's atmosphere to the impact of comet Shoemaker–Levy 9

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*Nature* **368,** 525–527 (1994)

IN this letter, Fig. 2*a* and *b* was printed upside-down; the figure is correctly displayed here.