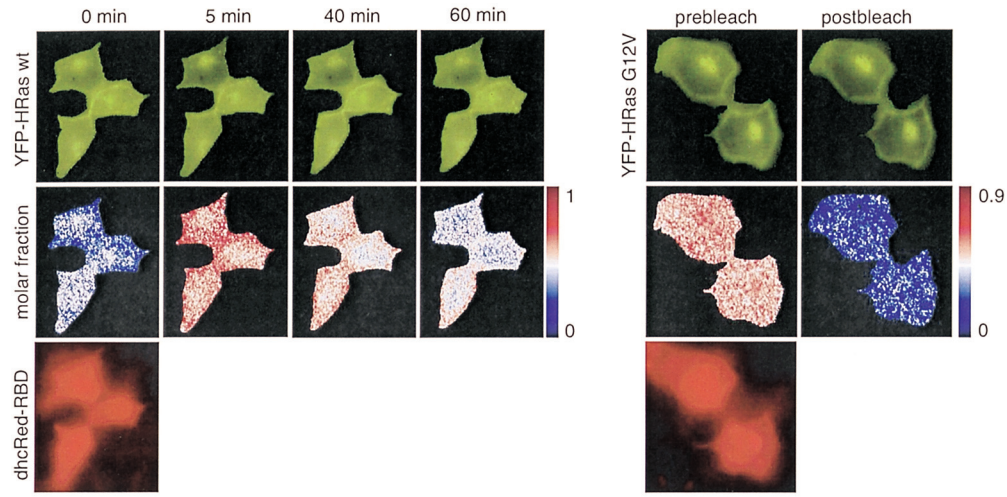

FIGURE 6. FLIM images of live cells showing the activation status of H-ras. (*Left*) Molar fractions of active H-ras in MDCK cells coexpressing RafRBD-dhcRed and YFP-H-ras after stimulation with 200 μM zinc at the indicated time points. (*Right*) The measured molar fractions of activated H-ras in MDCK cells coexpressing the constitutively activated mutant YFP-H-ras G12V and RafRBD-dhcRed, before and after photobleaching of the acceptor fluorophore. The photobleaching is a control to show that the fraction of H-ras molecules that undergo FRET is reduced to zero after destroying the acceptor (signal is due to FRET). In effect, the active H-ras can no longer be measured, but is presumed to remain.

Protein–Protein Interactions: A Molecular Cloning *Manual*, 2nd Ed., © 2005 by Cold Spring Harbor Laboratory Press, Chapter 32, Figure 6.