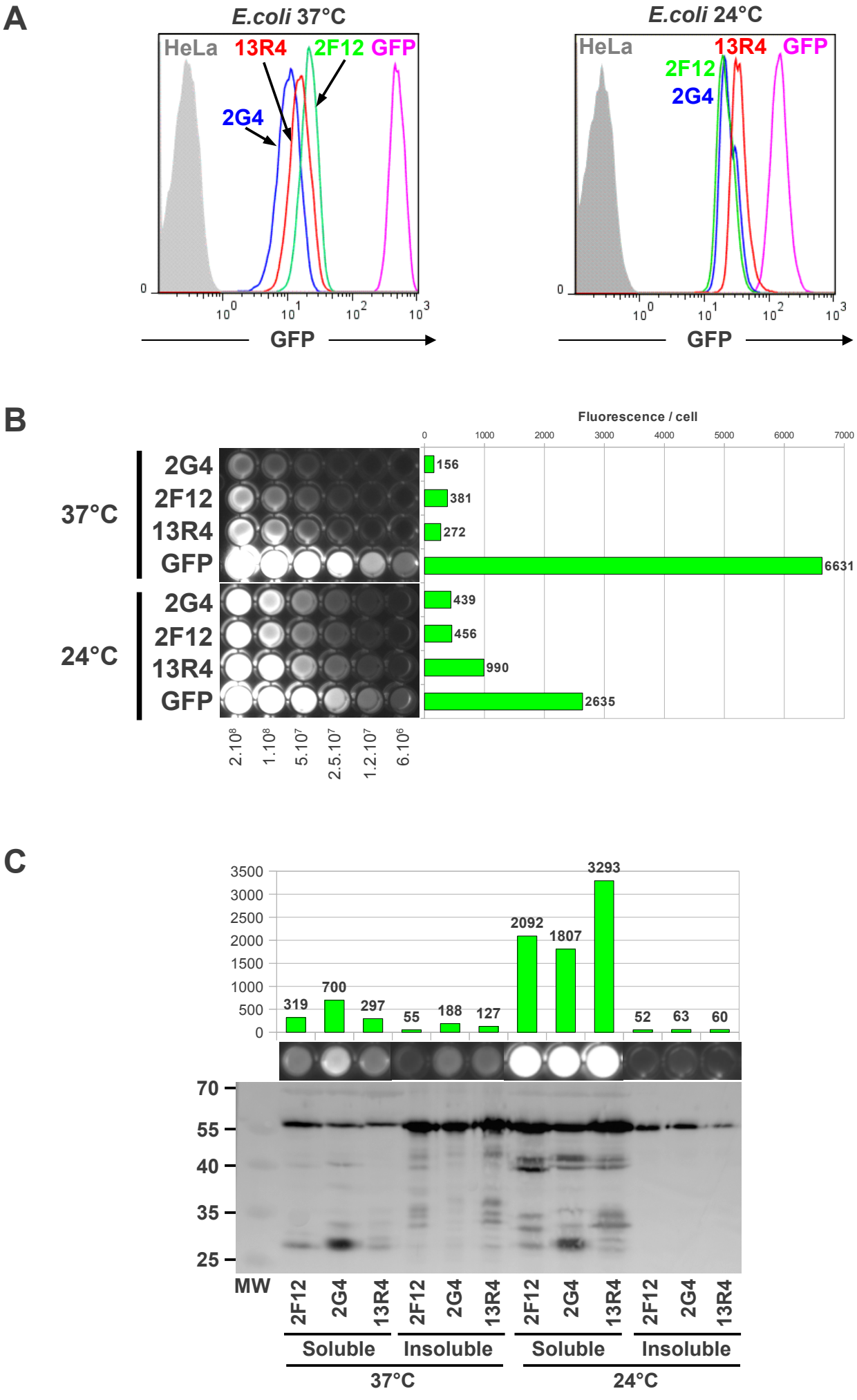

Expression and activity of scFv-GFP fusions in *E. coli*

ScFv 2F12, 2G4 and 13R4 were expressed in BL21(DE3) as a fusion with the GFP folding reporter (Waldo *et al.*, 1999) under the control of the T7 promoter. Expression plasmid was obtained by inserting the GFP folding reporter gene in the *Not*I site of pET23NN plasmid (Philibert and Martineau, 2004). Bacteria were grown in auto-inducible medium at 37°C and 24°C. A) Bacteria were diluted 5000-fold in PBS and at least 10,000 events were analysed using a Beckman-Coulter FC500 cytometer (488 nm excitation, 525 nm emission). B) Bacteria were pelleted, washed in PBS, then resupended in 1/4th of the initial culture volume. Two-step dilutions were prepared in a 96-well microtiter plate and the fluorescence of 100 µl of each dilution was measured using a Molecular Device Gemini XS spectrofluorometer (485 nm excitation, 538 nm emission, 9 nm band-pass). The signal was normalised using the OD600nm of the same plate. The picture of the plate was obtained using a Safe Imager™ blue light transilluminator (Invitrogen). C) Soluble and insoluble fractions were prepared and analysed by Western blot using a rabbit polyclonal serum against GFP. The GFP fluorescence of each fraction, displayed at the top, was measured as in B.