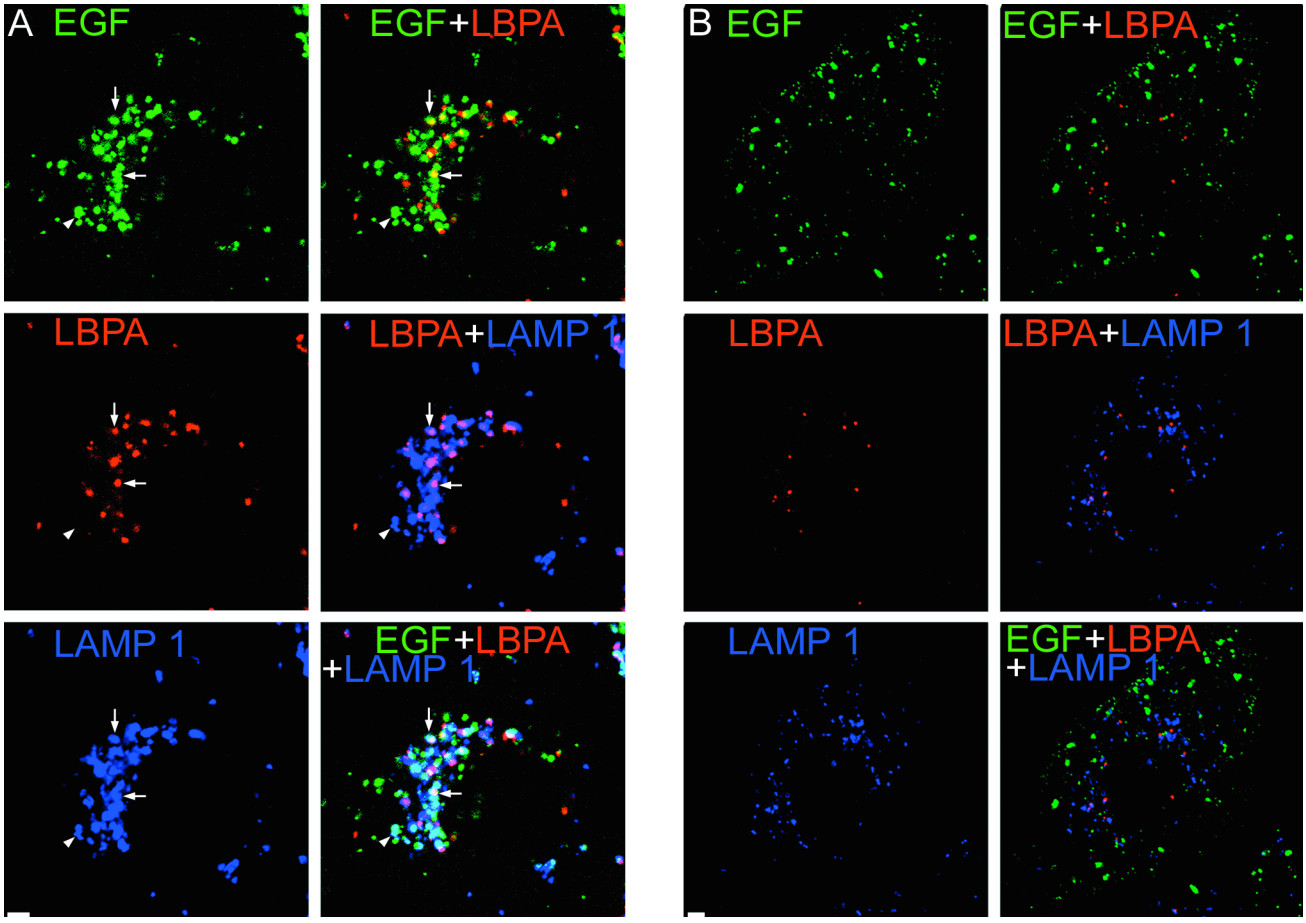
Supplementary Figure.

LBPA compartments that fail to crosslink do not contain EGF

HEp 2 cells were loaded with HRP in preparation for cross-linking. Non cross-linked cells treated without DAB/H_20_2 (A) or those that were cross-linked with DAB/H_20_2 (B) were stimulated with fluorescent EGF at 37ºC for 90 min in the presence of leupeptin. Cells were fixed prior to permeabilisation then stained with antibodies raised against LBPA and LAMP 1. Colocalisation between EGF and LBPA appears as yellow in the merged images, LBPA and LAMP 1 as magenta, EGF and LAMP 1 (example shown by arrowheads) as turquoise, and any colocalisation between all three (examples shown by arrows) as white.

Compartments showing colocalisation of EGF and LBPA in non cross-linked cells is always accompanied by LAMP 1 staining. Cross-linking of lysosomes abolishes both the majority of LBPA staining, and any colocalisation between EGF, LBPA, and LAMP 1. Bars = 5μm.


A EGF
EGF+LBPA
LBPA
LBPA+LAMP 1
LAMP 1
EGF+LBPA
+LAMP 1
B EGF
EGF+LBPA
LBPA
LBPA+LAMP 1
LAMP 1
EGF+LBPA
+LAMP 1