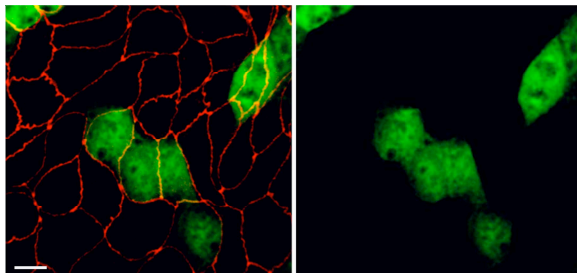
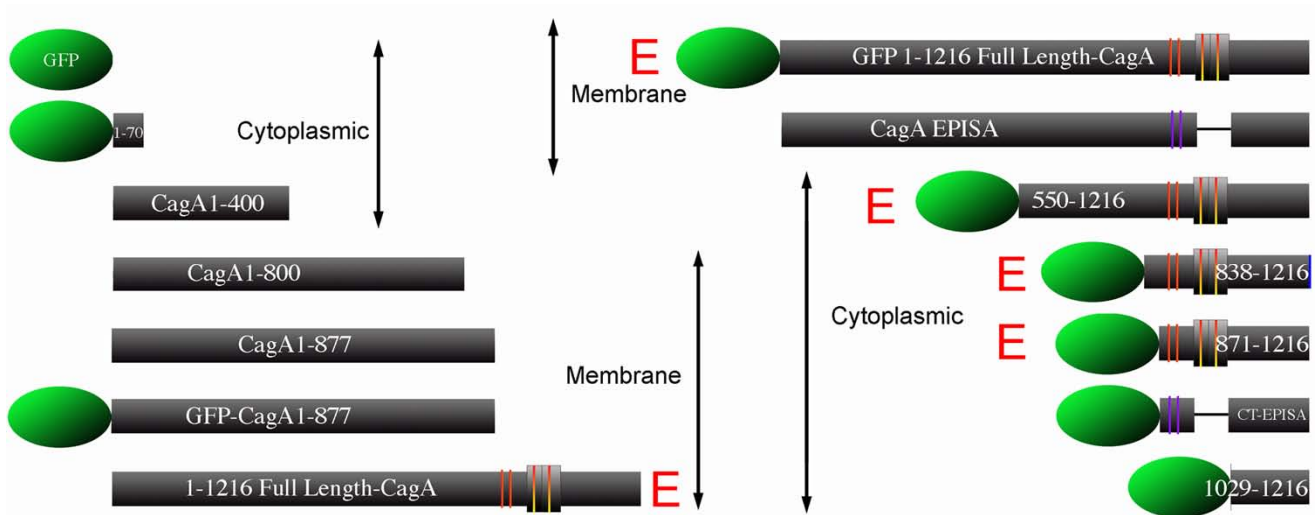
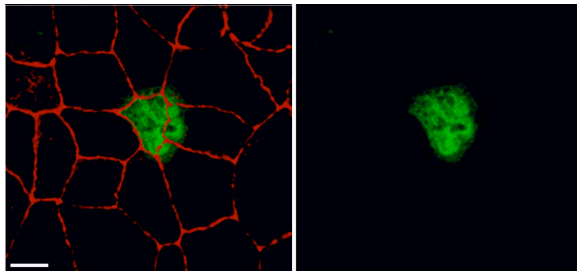


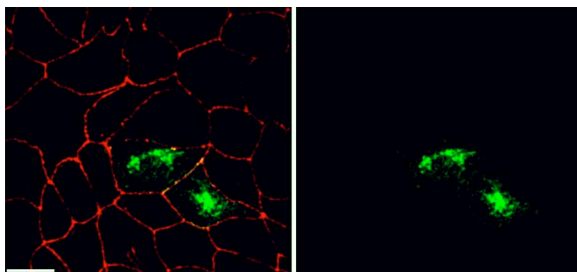
CagA fragments, their localization and effect on MDCK or AGS cell morphology (E = elongated).



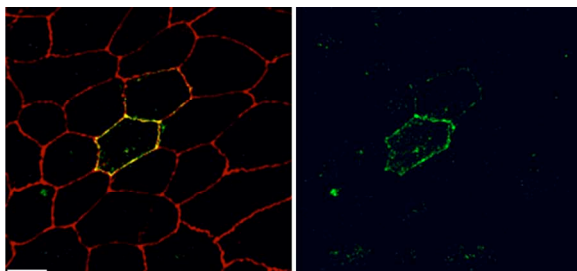
Construct: GFP
Vector: EGFP N3- Clontech
Localization: Diffuse, cytoplasmic
Cell Type: MDCK
Cell Morphology: Normal
Visualization: GFP fluorescence
Notes: GFP also diffuses into the nucleus



Construct: GFP-CagA1-70
Vector: EGFP N3- Clontech
Localization: Diffuse, cytoplasmic
Cell Type: MDCK
Cell Morphology: Normal
Visualization: GFP fluorescence
Notes: The N-terminal 70aa fragment does not have a localization domain

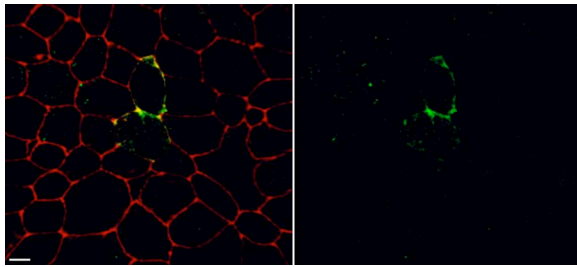


Construct: CagA1-400
Vector: pCI - Promega
Localization: Cytoplasmic near the nucleus
Cell Type: MDCK
Cell Morphology: Normal
Visualization: anti-CagA antibodies
Notes: Recognized by anti-CagA antibodies



Construct: CagA1-800
Vector: pIRES- Clontech
Localization: Peripheral near the plasma membrane with enhancement at the intercellular junctions
Cell Type: MDCK
Cell Morphology: Normal
Visualization: anti-CagA antibodies
Notes: Smaller than the NT-CagA fragment studied in the manuscript, yet it contains the membrane localization information.





Construct: CagA1-877 or NT-CagA

Vector: pIRES- Clontech

Localization: Peripheral near the plasma membrane with enhancement at the intercellular junctions

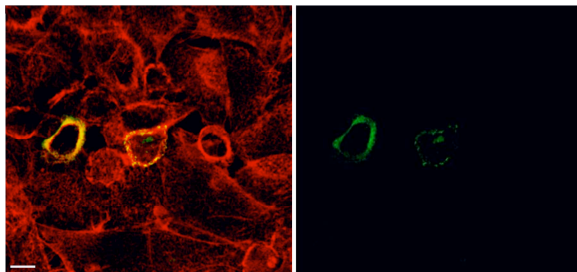
Cell Type: MDCK

Cell Morphology: Normal

Visualization: anti-CagA antibodies

Notes: NT-CagA has the same localization whether visualized by antibodies or through the GFP tag. See Fig. 4.

CagA1-877



Construct: GFP-CagA1-877 or GFP-NT-CagA

Vector: EGFP C3- Clontech

Localization: Peripheral near the plasma membrane

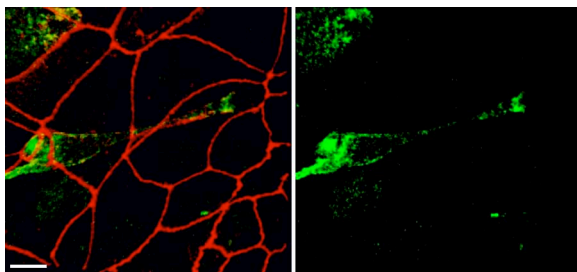
Cell Type: AGS

Cell Morphology: Normal

Visualization: GFP fluorescence

Notes: The NT-CagA fragment localizes near the membrane in unpolarized AGS cells.

GFP-CagA1-877



Construct: CagA1-1216 or Full Length CagA

Vector: pIRES- Clontech

Localization: Peripheral near the plasma membrane with enhancement at the intercellular junctions. Causes junction disruption

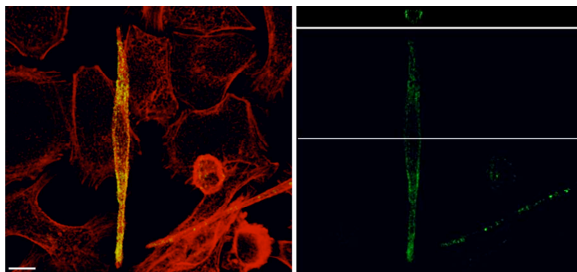
Cell Type: MDCK

Cell Morphology: Elongated. Loss of polarity

Visualization: anti-CagA antibodies

Notes: The GFP tagged version described in the manuscript has identical properties.

1-1216 Full Length-CagA



Construct: GFP-CagA1-1216 or Full Length GFP-CagA

Vector: EGFP C3- Clontech

Localization: Peripheral near the plasma membrane.

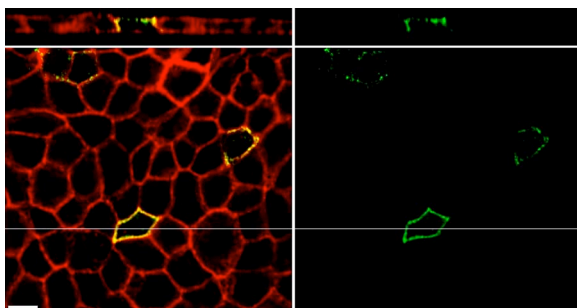
Cell Type: AGS

Cell Morphology: Elongated

Visualization: GFP fluorescence

Notes: This is the same construct used in the manuscript but expressed in AGS cells. The cross section visualizes the protein near the membrane. Compare to GFP-CT-CagA.

GFP 1-1216 Full Length-CagA



Construct: CagAEPISA

Vector: pIRES-Clontech

Localization: Peripheral near the plasma membrane with enhancement at the intercellular junctions.

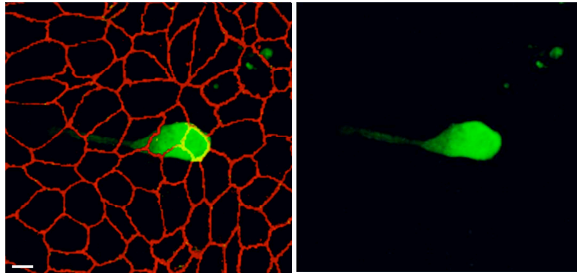
Cell Type: MDCK

Cell Morphology: Normal

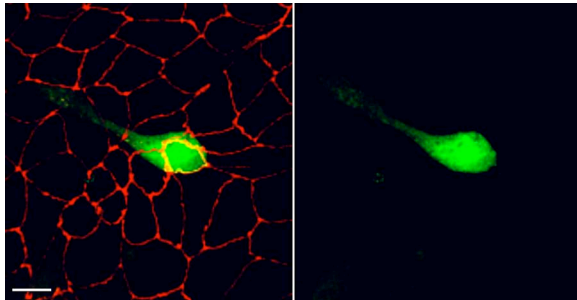
Visualization: anti-CagA antibodies

Notes: This construct lacks the last two EPIYA repeats and in the first two, the tyrosines have been replaced by serine. The protein is not tyrosine phosphorylated. Cross sections at the top. This monolayer is counterstained with phalloidin.

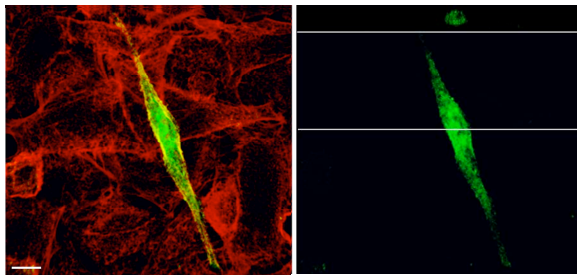
CagA EPISA



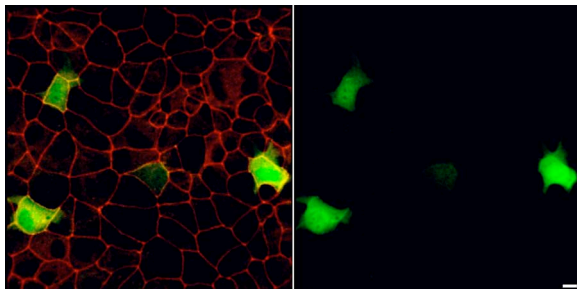
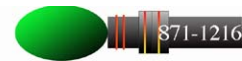
Construct: GFP-CagA550-1216
Vector: pIRES- Clontech
Localization: Diffuse throughout the cell cytoplasm.
Cell Type: MDCK
Cell Morphology: elongated
Visualization: GFP fluorescence
Notes: Larger construct than CT-CagA. It causes the same phenotypes as GFP-CT-CagA



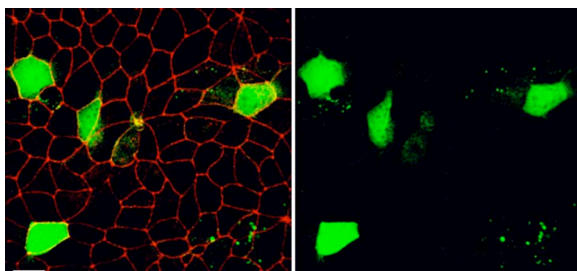
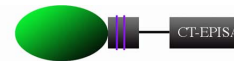
Construct: CT-CagA838-1216-FLAG
Vector: EGFP C3- Clontech
Localization: Diffuse throughout the cell cytoplasm.
Cell Type: MDCK
Cell Morphology: elongated
Visualization: GFP fluorescence
Notes: This construct behaves like the CT-CagA fragment and also has a C-terminal FLAG tag for immunoprecipitation.



Construct: GFP-CagA871-1216 or GFP-CT-CagA
Vector: pIRES - Clontech
Localization: Diffuse throughout the cell cytoplasm.
Cell Type: AGS
Cell Morphology: elongated
Visualization: GFP fluorescence
Notes: CT-CagA expressed in AGS cells causes elongation. Cross section shown at top of GFP image shows cytosolic distribution.



Construct: GFP-CT-EPISA
Vector: pIRES- Clontech
Localization: Diffuse throughout the cell cytoplasm.
Cell Type: MDCK
Cell Morphology: Normal
Visualization: GFP fluorescence
Notes: Based on CT-CagA, it lacks the last two EPIYA repeats and has the tyrosines mutated to serine in the first two EPIYA motifs.



Construct: GFP-CagA1029-1216
Vector: pIRES- Clontech
Localization: Diffuse throughout the cell cytoplasm.
Cell Type: MDCK
Cell Morphology: Normal
Visualization: GFP fluorescence
Notes: This construct lacks all the EPIYA motifs.

