Supporting information

Label-free quantification of cell-penetrating peptide translocation into liposomes

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Figure S1- Dynamic light scattering measurements of POPG and POPC liposomes in absence and presence of RW₉, penetratin and RFWF₉.

Figure S2- Turbidity measurements of POPG and POPC liposomes in absence and presence of RW₉, penetratin and RFWF₉.

Figure S3 - Maximum fluorescence intensity of RFWF₉ alone, after 30 min incubation with LUVs loaded or unloaded with acrylamide and following acrylamide addition.

Table S1 - Calcein leakage upon addition of 1 M acrylamide to POPC and POPG liposomes (loaded or unloaded with acrylamide).

Table S2 - Average size of the POPG and POPC LUVs loaded or unloaded with acrylamide before and after incubation with 1 M acrylamide measured by DLS.

Table S3 - Turbidity of POPG and POPC LUVs (50 μ M) loaded or unloaded with acrylamide before and after 60 min incubation with 1 M acrylamide. Turbidity was measured by the absorbance at 436 nm.



Fig. S1. Dynamic Light Scattering measurements of the POPG and POPC LUVs (50 μ M) loaded or unloaded with acrylamide before (black) and after incubation with RW₉ (red), Penetratin (blue) and RFWF₉ (green) at 1 μ M (P/L 1/50). Bars represent the average value obtained from 3 different measurements (each averaged out as they were performed in triplicate) and error bars represent the standard error of the mean (SEM). Polydispersity index values obtained are below 0.2.



Fig. S2. Turbidity of POPG and POPC LUVs (50 μ M) loaded or unloaded with acrylamide before (black) and after incubation with RW₉ (red), Penetratin (blue) and

RFWF₉ (green) at 1 μ M (P/L 1/50). Bars represent the mean obtained from three experiments and error bars represent the standard error mean (SEM).



Figure S3. Maximum fluorescence intensity of RFWF₉ alone (0.5 μ M; black), after 30 min incubation with LUVs that encapsulate acrylamide (red) and that do not encapsulate acrylamide (yellow) (P/L 1/100) and following acrylamide addition (1 mM) to LUVs encapsulating (blue) or not acrylamide (green). Panel A was performed with POPG and panel B with POPC. The difference between the fluorescence maximum intensity values of the peptide following acrylamide addition to the liposomes encapsulating (blue) or not (green) acrylamide provides the amount of internalized peptide into LUVs. Difference between blue and green data is not significant for both panels. The data was normalized to the peptide concentration. The present data was performed three times and each in triplicate. Bars represent the average value obtained from the average values of three different experiments (each in triplicate) and error bars represent the standard error of the mean (SEM).

Table S1. Calcein leakage (shown in percentage) upon addition of 1 M acrylamide to POPC and POPG liposomes (loaded or unloaded with acrylamide) following 60 min incubation at room temperature.

	POPC (acryl)	POPC (no acryl)	POPG (acryl)	POPG (no acryl)
No	0.52 ± 0.05	0.62 ± 0.06	0.50 ± 0.03	0.64 ± 0.06
acrylamide				
With 1M	0.62 ± 0.03	0.65 ± 0.04	0.48 ± 0.20	0.68 ± 0.08
acrylamide				

Note: The data presented corresponds to the average value and corresponding standard deviation (SD) from 3 independent measurements.

Table S2. Average size (in nm) of the POPG and POPC LUVs (50 μ M) loaded or unloaded with acrylamide before and after 60 min incubation with 1 M acrylamide measured by DLS.

	POPC (acryl)	POPC (no acryl)	POPG (acryl)	POPG (no acryl)
No	150 ± 19	130 ± 14	160 ± 15	120 ± 12
acrylamide				
With 1M	145 ± 21	140 ± 18	152 ± 21	112 ± 15
acrylamide				

Note: The data presented corresponds to the average value and corresponding standard deviation (SD) from 3 independent measurements.

Table S3. Turbidity of POPG and POPC LUVs (50 μ M) loaded or unloaded with acrylamide before and after 60 min incubation with 1 M acrylamide. Turbidity was measured by the absorbance at 436 nm.

	POPC (acryl)	POPC (no acryl)	POPG (acryl)	POPG (no acryl)
No	0.003 ± 0.0004	0.004 ± 0.0003	0.001 ± 0.0004	0.003 ± 0.0002
acrylamide				
5				
With 1M	0.005 ± 0.0005	0.006 ± 0.0004	0.009 ± 0.0008	0.005 ± 0.0005
1 . 1				
acrylamide				

Note: The data presented corresponds to the average value and corresponding standard deviation (SD) from 3 independent measurements.