

SUPPLEMENTARY ONLINE DATA

Differentiation of propeptide residues regulating the compartmentalization, maturation and activity of the broad-range phospholipase C of *Listeria monocytogenes*

Emily R. SLEPKOV, Alan PAVINSKI BITAR and Hélène MARQUIS¹

Department of Microbiology and Immunology, Cornell University, Ithaca, NY 14853, U.S.A.

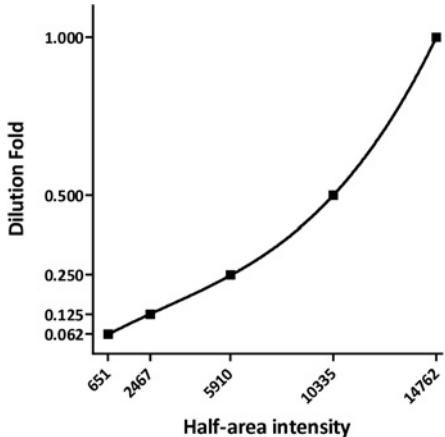


Figure S1 Regression curve of band intensity in function of protein concentration

PC-PLC was immunoprecipitated from pulse-labelled infected cells and 2-fold serial dilutions were resolved by SDS/PAGE. The autoradiograph was scanned with Image J. The half-area intensity was calculated for each lane and plotted as a function of the protein dilution fold. The curve follows a third-level exponential scale and the derived equation ($y = [2.897 \times 10^{-13}]x^3 - [2.68 \times 10^{-9}]x^2 + [4.176 \times 10^{-5}]x + 0.03561$) was used to normalize the experimental results obtained with the wild-type strain and the mutants.

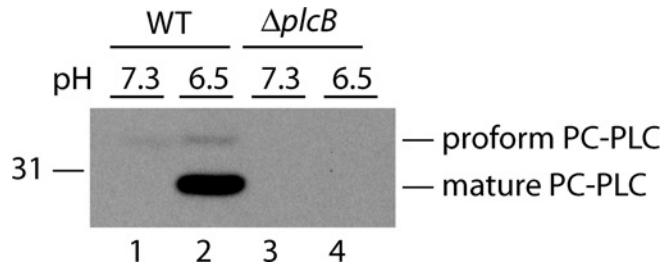


Figure S2 Compartmentalization and maturation of PC-PLC is regulated by pH in human epithelial HeLa cells

Infected HeLa cells were pulse-labelled with [³⁵S]methionine/cysteine, and bacterial protein synthesis was inhibited by the addition of chloramphenicol before chasing in nigericin-supplemented buffer at pH 7.3 or 6.5. Secreted PC-PLC was immunoprecipitated from host-cell lysates and detected by autoradiography. The position of a molecular-mass marker in kDa is indicated on the far left-hand side. Positions of the proform and mature form of PC-PLC are indicated on the far right-hand side. Lanes 1 and 2, cells were infected with strain HEL-1031 expressing wild-type (WT) PC-PLC; lanes 3 and 4, cells were infected with strain DP-L1935, which has a deletion in the gene encoding PC-PLC ($\Delta plcB$).

¹ To whom correspondence should be addressed (email hm72@cornell.edu).

Table S1 Sequences of DNA oligonucleotide primers used in the present study

The underlined sequences represent XbaI and PstI restriction sites in Marq 512 and Marq 515 respectively.

Primer	Sequence (5'→3')
Marq 512	CCCTCGAGTTAACAAATGTTAGAGAAAAATTAAATTCT
Marq 513	<u>CGTATCAGCAGGAGGGAGTATAAGTGAATGATATGAAATTCAAAATGTG</u>
Marq 514	CACATTITTAATTCATATCATTCACTTATCTCCCTCTCGTGATACG
Marq 515	<u>GGGCTGCAGTAAATATCCACCCGCTAACGA</u>
Marq 516	GTAACGATAAAAGCAATGCCAGCTGCTGAATACTTACAAACACCCGC
Marq 517	<u>GCGGCTTGTGTTAAGTATTGCACTGCGGCTTGTGCTTACGTTAC</u>
Marq 518	GCAAATGCGTGTGATCGAGCTGCACAAACACCCGAGCTCC
Marq 519	GGAGCTCGGGGTGTTGCACTGCTCATCACACAGGCATTG
Marq 520	GATGAATACTTAGCCGCAGCCGCAGCTCCGC
Marq 521	GCGGAGCTCGGGCTGCAGCTAACAG
Marq 522	CGGTAAACGATAAAAGCAATGCCATGATATTGACAGCAATTACCTC
Marq 523	<u>GAGGTAATTGCTGCAATATCATGGGCATTGCTTTATCGTTACCG</u>
Marq 524	TTACAAACACCCGAGCTCTCATAAACTTAGTTGGTCCCGGG
Marq 525	CCGCAGGACCAACTAAGTTATGAGGAGCTGCGGGTGTGAA
Marq 526	CTTACAAACACCCGAGCTGGCTGCTATTGACAGCAATTAC
Marq 527	<u>GGTAATTGCTGCAATAGCAGCCGCAGCTGGGTGTTGTAAG</u>
Marq 528	CTTACAAACACCCAAACACCGCATGATATTG
Marq 529	CAATATCATGGGTGTTGGGTGTTGTAAG
Marq 530	AGCTCCGCATGATGCTGCCAAATTACCAT
Marq 531	ATGTTGTAATTGGCCGCAGCATGCGGAGCT
Marq 532	TGATATTGACAGCGCAGCACATAAACTTAG
Marq 533	CTAAGTTATGCTGCTGCCGTGCAATATCA
Marq 534	CAGCAAATTACAGCTGCACTTAGTTGGTCT
Marq 535	GGACCAACTAAGTGCAGCTGGTAATTGCTG
Marq 536	ATTACACATAAAAGCTGCTGGTCAGCGATAA
Marq 537	TTATCCGCTGACCAAGCAGCTTATGTTGTAAT
Marq 538	GATGAATACTAAACACCCGAGCTC
Marq 539	GAGCTCGGGTGGTTAAGTATTCA
Marq 549	CGGTAAACGATAAAAGCAATGCCAAACACCCGAGCTCCGCAT
Marq 550	ATGCGGAGCTCGGGTGTGGCATTTGCTTTATCGTTACCG
Marq 551	AAATGCCCTTGTGATGAATACTACATGATATTGACAGCAATTACAC
Marq 552	GTGGTAATTGCTGCAATATCATGTAAGTATTCA
Marq 553	GATAAAAGCAATGCCGTGATGCGAGCTCCGCATGATATTGACA
Marq 554	TGCAATATCATGGGAGCTCACAAACAGGATTGCTTTATC
Marq 555	ATGCTGTTGCTGCACTAACAC
Marq 556	GTTTGTAAAGTATGCAAGCACACAGGCAT
Marq 570	CGGTAAACGATAAAAGCAATGCCCGCAGCTCCGCATGATATT
Marq 571	AATATCATGGGAGCTGGGGCATTTGCTTTATCGTTACCG
Marq 572	CGGTAAACGATAAAAGCAATGCCGCTCCGCATGATATTGACAGCA
Marq 573	TGCTGTCATATCATGGGAGGCCATTGCTTTATCGTTACCG
Marq 574	GATGAATACTAAACACCCGACTTAGTTGGTCCCGGGATAACC
Marq 575	GGTTATCCGCGACCAACTAAGTGCAGGTTGTAAGTATTCA
Marq 576	CGGTAAACGATAAAAGCAATGCCATTGCAAGCAATTACACATAAAC
Marq 577	GTTTATGTTGTAATTGCTGCAATGGCATTGCTTTATCGTTACCG
Marq 578	CGGTAAACGATAAAAGCAATGCCAGCAATTACACATAAACATTAGTTG
Marq 579	CAACTAAGTTATGTTGCAATTGCTGGCATTGCTTTATCGTTACCG
Marq 580	CGGTAAACGATAAAAGCAATGCCATTACACATAAACATTAGTTGGTCC
Marq 581	GGACCAACTAAGTTATGTTGTAAGGCAATTGCTTTATCGTTACCG
Marq 582	CGGTAAACGATAAAAGCAATGCCATTGCTGGCAGGCAATTGCTTTATCGTTACCG
Marq 583	GTCGGGTTATCCGCGGACCGAGCATTGCTTTATCGTTACCG
Marq 595	ATAAAAGCAATGCCGTGTTGATGCATACTAACACCCGAGCTC
Marq 596	GAGCTGCCGGTGTGTAAGTATGCAACACAGGATTGCTTTAT
Marq 597	TGCTGTCATATCATGGGAGGCCATTGCTTTATCGTTACCG
Marq 598	GATGAATACTAAACACCCGACTTAGTTGGTCCCGGGATAACC
Marq 599	GGTTATCCGCGACCAACTAAGTGCAGGTTGTTGTAAGTATTCA
Marq 600	CGGTAAACGATAAAAGCAATGCCATTGCAAGCAATTACACATAAAC
Marq 601	GTTTATGTTGTAATTGCTGCAATGGCATTGCTTTATCGTTACCG
Marq 602	CGGTAAACGATAAAAGCAATGCCAGCAATTACACATAAACATTAGTTG

Table S1 Continued

Primer	Sequence (5'→3')
Marq 603	GCAAATGCCCTGTTGTGATGAATAACGCACAAACACCCGCAGCTCCGCAT
Marq 604	ATGCGGAGCTGCGGGGTGTTGTGCGTATTCATCACAAACAGGCATTGC
Marq 609	CGGTAAACGATAAAAAGCAAATGCCAAACTAGTTGGCATTGCTTTATCGTTACCG
Marq 610	TTATCCCGGGACCAACTAAGTTGGCATTGCTTTATCGTTACCG
Marq 611	CGGTAAACGATAAAAAGCAAATGCCAGTTGGTCCGCGGATAACCCG
Marq 612	CGGGTTATCCCGGGACCAACTGGCATTGCTTTATCGTTACCG

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