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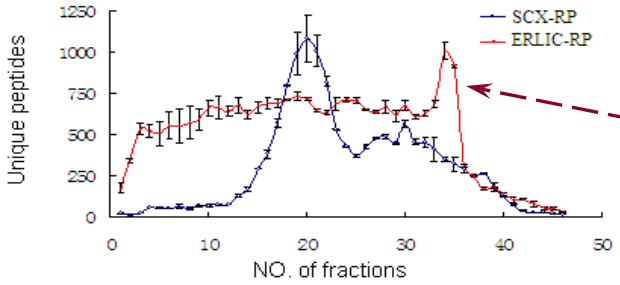
Superior Proteomics Fractionations with ERLIC*-RPC

*(Electrostatic Repulsion-Hydrophilic Interaction Chromatography) March 2010

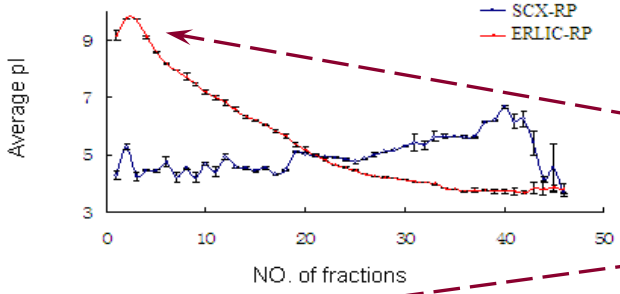
In bottom-up proteomics, more peptides of lower abundance are identified if complex digests are fractionated via two dimensions of chromatography. The most widely used combination at present is SCX-RPC. There are significant advantages to substitution of the new ERLIC mode for SCX. ERLIC of peptides is performed with an anion-exchange column operated in the HILIC mode at low pH. With a decreasing organic solvent gradient, hydrophilic interaction becomes so weak that electrostatic repulsion causes the peptides' elution. Volatile solvents can be used!

Example: 2 mg. peptides from tryptic digest of rat kidney homogenate



<u>Distribution of peptides</u>:

Much more uniform in ERLIC than in SCX

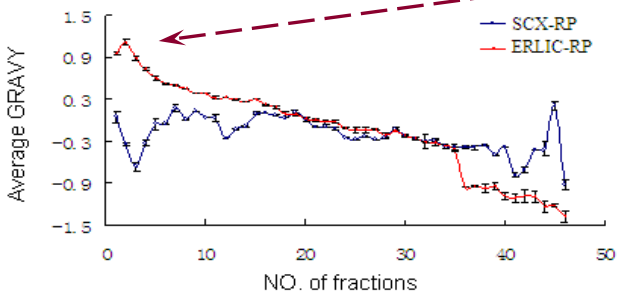


<u>Basis of separation in ERLIC</u>:

pI of peptide (high to low)

and

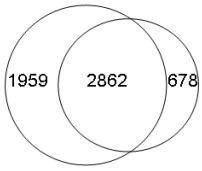
polarity (low to high)



- both variables are complementary to selectivity in RPC -

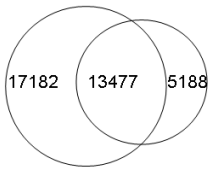
The number of identifications is significantly higher with ERLIC than with SCX

Protein id's

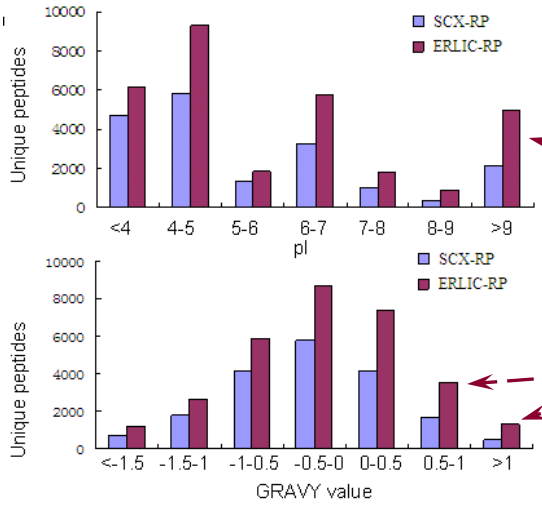


ERLIC -RPC SCX-RPC

Unique Peptide id's



ERLIC -RPC SCX-RPC

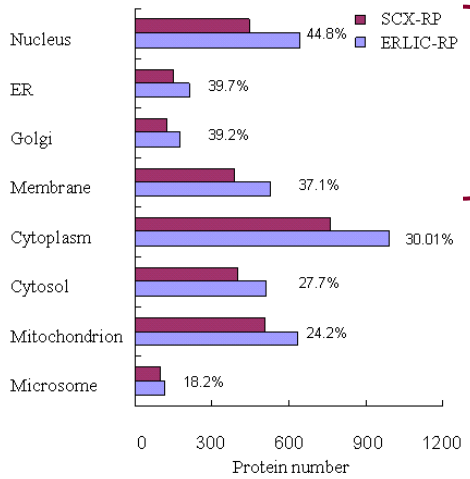


ERLIC affords > 120% more id's than does SCX with peptides that are:

very basic

or

very hydrophobic



This makes it more effective at identifying peptides from membrane-rich organelles

- *data courtesy of S.K. Sze, Nanyang Technological Univ.* -

References on ERLIC:

1) A.J. Alpert, *Anal. Chem. 80* (2008) 62
2) C.S. Gan , T. Guo, H. Zhang, S.K. Lim, and S.K. Sze, *J. Proteome Res. 7* (2008) 4869
3) U.L. Lewandrowski, K. Lohrig, R. Zahedi,. D. Walter, and A. Sickmann, *Clin. Proteom 4* (2008) 12
4) H. Zhang, T. Guo, X. Li, A. Datta, J.E. Park, J. Yang, S.K. Lim, J.P. Tam, and S.K. Sze, *Mol. Cell Proteomics, online Jan. 2010*
5) P. Hao, T. Guo, X. Li, S. Adav, J. Yang, W. Meng, and S.K. Sze, *submitted for publication*

Buying products: Contact The Nest Group, Inc., 800-347-6378, www.nestgrp.com

ERLIC: PolyWAX LP column, 200 x 4.6mm, 5µm, 300Å (item# P204WX0503)

SCX: PolySULFOETHYL A column, 200 x 4.6mm , 5µm, 200Å (item# P204SE0502)