Multiplexed Gel-Eluted Liquid Fraction Entrapment Electrophoresis: A Liquid-Phase Method for Mass Separation and Analysis
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OVERVIEW
- A multiplexed GELFrEE device is reported for proteome prefractionation
- 8-fold increase in loading capacity & throughput
- LC-MS/MS of 16 resulting fractions gave 420 unique yeast proteins
- Separation is highly correlated to mass of identified proteins

INTRODUCTION
GELFrEE\(^1\) affords rapid mass-based protein separation over a range 10-150 kDa. Here, we demonstrate a multiplexed design enabling increased loading capacity and throughput. We demonstrate comprehensive analysis of the yeast proteome using GELFrEE coupled to LC-MS/MS analysis.

\(^1\)Tran & Doucette, Anal. Chem. 2008, 80, 1568-1573.

METHODS
- Instrument design: 8 gel columns are coupled in parallel to independent collection chambers
- All buffers and gels were prepared according to Laemmli protocol
- 150 µL of sample is loaded per gel column
- Fractions were analyzed by 1D gels or precipitated, digested and subject to LC-MS/MS (1.5 hr gradient 5-40% ACN / 0.1% formic acid) on an LTQ linear ion trap MS

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CONCLUSIONS
- Multiplexed GELFrEE affords high throughput & loading
- Compatible with LC MS/MS analysis
- Collected fractions highly correlate to molecular weight in MS