Table S1. Electrospray ionization mass spectrometric analysis of purified  $\text{Ero1}\alpha$  complex.  $\text{Ero1}\alpha$  co-purifies with PDI and both species, as well as the mixed disulfide species, are observed by mass spectrometry.  $\text{Ero1}\alpha$  is produced with (phospho)gluconoylation of the N-terminal His tag (Geoghegan et al, 1999), with the gluconoylated species being the major state. For clarity, the other states of the N-terminal His tag (no modification and phosphogluconoylation) are not shown. The data are indicative that 14 of the 15 Cys in  $\text{Ero1}\alpha$  are in a disulfide state and that a mixed disulfide forms between  $\text{Ero1}\alpha$  and PDI in the inactive complex.

| NEM | Species<br>observed                   | Expected mass (Da) | Experimental mass (Da) | Δmass  | Explanation              |
|-----|---------------------------------------|--------------------|------------------------|--------|--------------------------|
| -   | Gluconoylated<br>Ero1α                | 53255              | 53240                  | -15Da  | 7 disulfides             |
|     | PDI                                   | 55425              | 55423                  | -2Da   | 1 disulfide              |
|     | Gluconoylated<br>Ero1α-PDI<br>complex | 108680             | 108663                 | -17Da  | 8 disulfides             |
| +   | Gluconoylated<br>Ero1α                | 53255              | 53365                  | +110Da | One NEM<br>7 disulfides  |
|     | PDI                                   | 55425              | 55546                  | +121Da | One NEM, 2<br>disulfides |
|     | PDI                                   | 55425              | 55670                  | +245Da | Two NEM, 2<br>disulfides |