

Online supplemental material. Figure S1 shows the probability based Mowse score used in the identification of Kar3 as a protein that co-purifies with Gpa1 in pheromone-treated cells.

Figure S2 provides representative images that were used to simultaneously score actin (top row) and microtubule (bottom row) polarity defects in shmooing cells. Cells expressing Kar3-GFP were stained with rhodamine phalloidin. Figure S3 provides recovery curves representative of 10 cells and a representative image of 1 cell for each strain and condition analyzed by FRAP (Table 4). The digital data were not manipulated in any way prior to performing the analysis. However, the contrast of the images shown here was uniformly increased to facilitate visualization of the Kar3-GFP signal. Videos 1 and 2 show the microtubule dynamics in pheromone-treated wild type and Gpa1-deficient cells, respectively.

Supplemental Figure 1

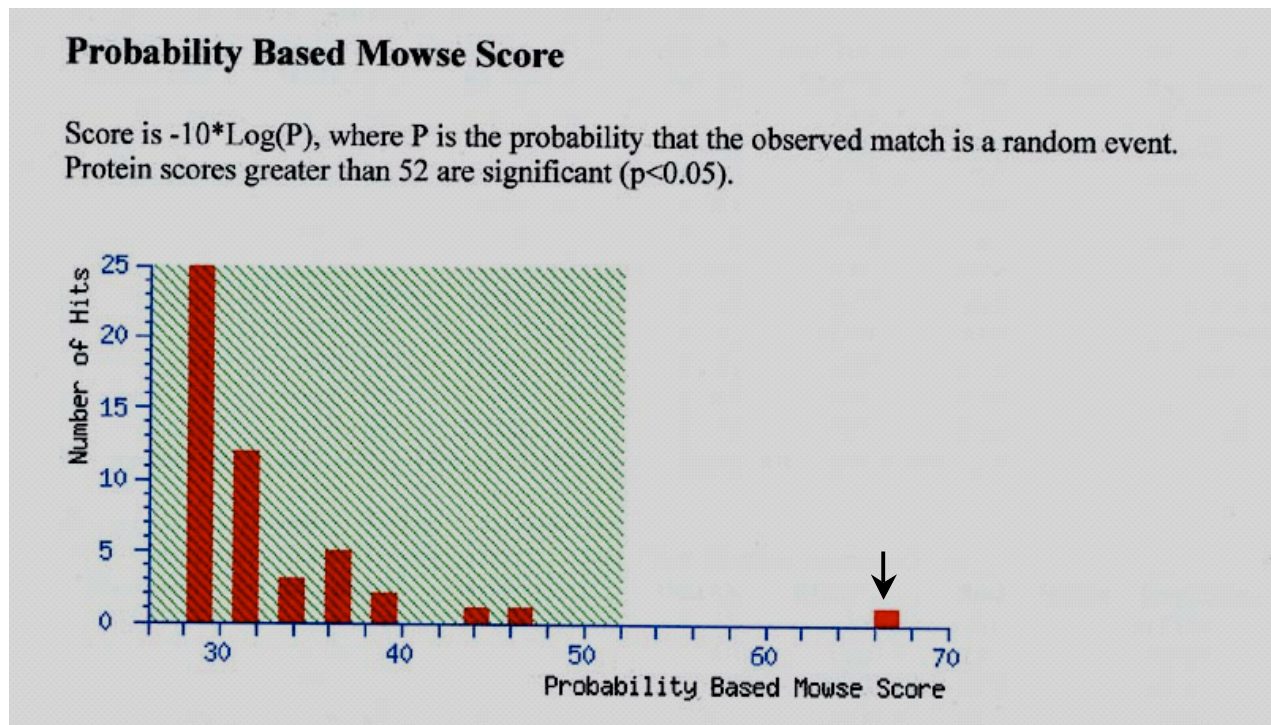
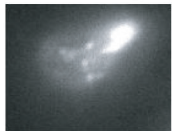
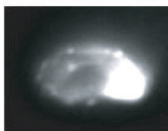
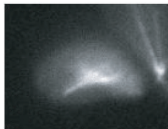


Figure S1. Identification of Kar3 as a protein that co-purifies with Gpa1 in pheromone-treated cells. Proteins were eluted from the Gpa1 affinity column and separated on two-dimensional gels. Spots that were seen only after pheromone treatment were then subjected to in-gel trypsin digestion, MALDI MS, and peptide fragment mass fingerprint analysis. The bar graph shows the number of potential matches as a function of probability score, as identified by MASCOT search engines. Hits outside the green area (scores greater than 52) are significant. Kar3 (arrow) was the only high-confidence hit for the protein spots shown in Fig. 1A.

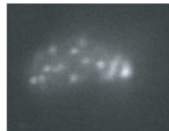
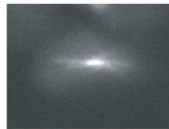
Supplemental figure 2



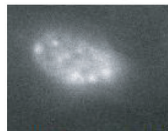
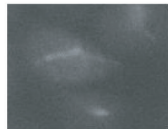
no defect



microtubule
polarity defect



actin
polarity defect



microtubule and actin
polarity defect

Supplemental Figure 3

