

## 1 SUPPLEMENTARY FIGURE LEGENDS

2

3 Fig. S1. **The G496P mutation abolishes the translocation activity of OtpB.** HDB114  
4 transformed with pPC20 ( $P_{trc}$ -*otpA*) and either pPC11 ( $P_{BAD}$ -*otpB*) (A) or pPC117 ( $P_{BAD}$ -*otpB*) (B)  
5 were incubated with IPTG and arabinose and then subjected to pulse-chase labeling. Cultures  
6 were divided into cell (C) and secreted (S) fractions and half of the cells were treated with  
7 proteinase K (PK). OtpA and OtpB were immunoprecipitated from all samples. The S samples  
8 were derived from twice as much culture volume as each C sample. m, molecular weight  
9 markers.

10

11 Fig. S2. **Retention of a significant fraction of OtpA(N)cys in the periplasm.** HDB114  
12 transformed with pPC11 ( $P_{BAD}$ -*otpB*) and pPC91 [ $P_{trc}$ -*otpA(N)cys*] were incubated with IPTG and  
13 arabinose and then subjected to pulse-chase labeling. Cultures were divided into cell (C) and  
14 secreted (S) fractions, and the cell fraction was divided into three portions. One aliquot was  
15 untreated, the second was treated with proteinase K (PK), and the third was converted to a  
16 spheroplast suspension prior to PK treatment (sph). OtpA was immunoprecipitated from all  
17 samples. The S samples were derived from three times as much culture volume as each C and  
18 sph sample.

19

20 Fig. S3. **Retention of a significant fraction of OtpAcys in the periplasm.** HDB114  
21 transformed with pPC11 ( $P_{BAD}$ -*otpB*) and pPC105 ( $P_{trc}$ -*otpAcys*) were incubated with IPTG and  
22 arabinose, pulse labeled, and subjected to a 10 min chase. Cultures were divided into cell (C)  
23 and secreted (S) fractions, and the cell fraction was divided into three portions. One aliquot was  
24 untreated, the second was treated with proteinase K (PK), and the third was converted to a  
25 spheroplast suspension prior to PK treatment (sph). OtpA was immunoprecipitated from all

- 1 samples. The S sample was derived from three times as much culture volume as the C and sph
- 2 samples. m, molecular weight markers.

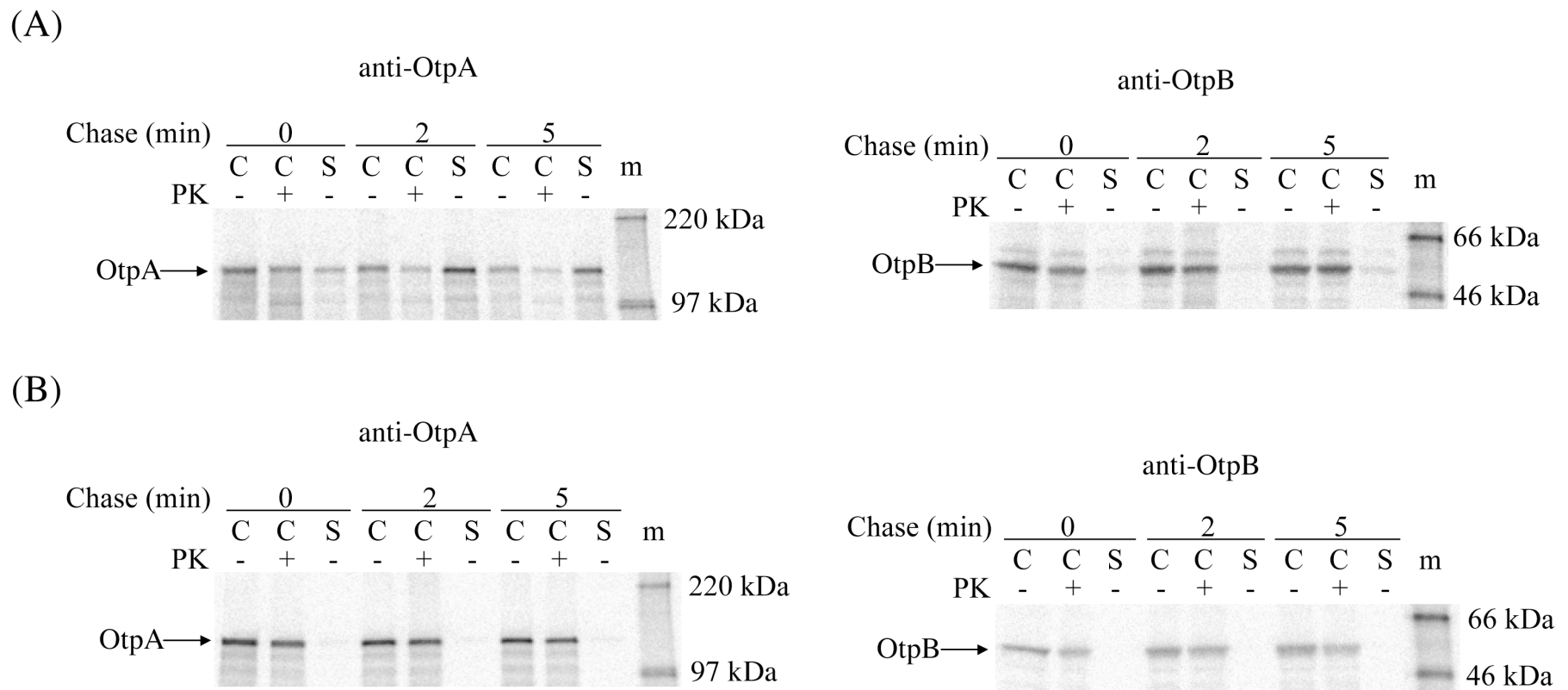


Fig. S1

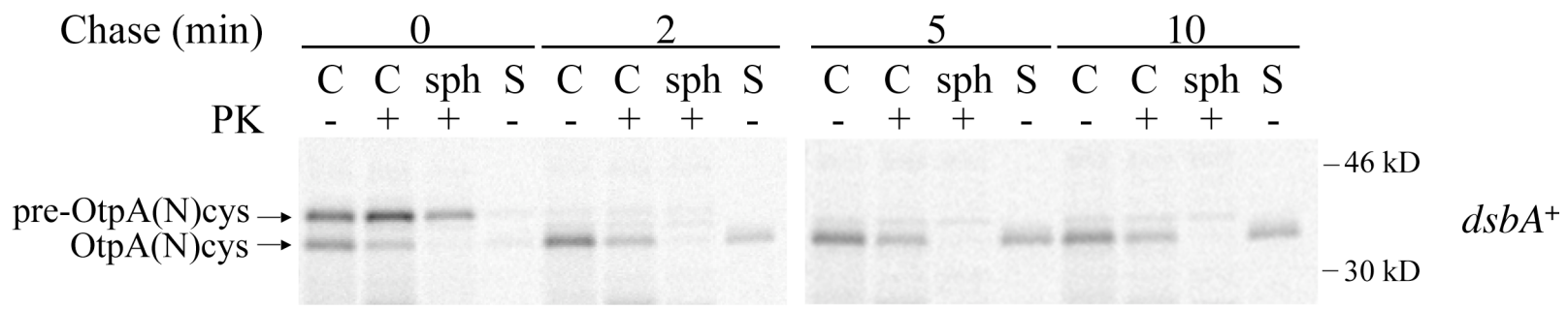


Fig. S2

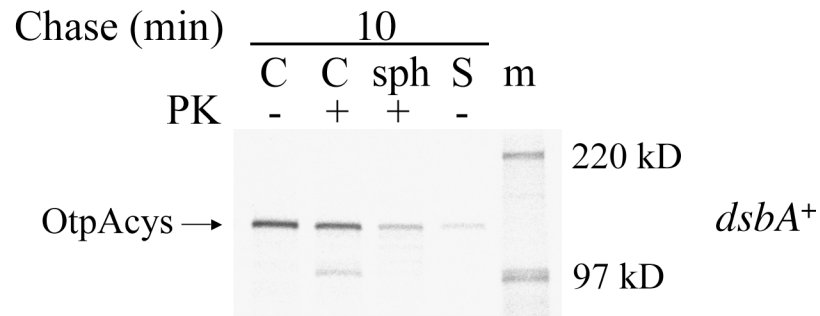


Fig. S3