**SUPPLEMENTAL**

**Table S1.** List of *E. coli* strains and plasmids used.

| Identification | Description | Reference |
|---|---|---|
| BL21 (DE3) | Reductive cytoplasm<br>Known for high levels of recombinant protein expression<br>Does not promote disulfide bond formation in the cytoplasm | 10 |
| Origami 2 | K-12 derivative<br>Oxidative cytosolic environment<br>Promotes disulfide bond formation in the cytoplasm | 11 |
| SHuffle® Express Competent *E. Coli* (C3028) | BL21 derivative<br>Oxidative cytosolic environment<br>Expresses prokaryotic disulfide bond isomerase, DsbC Promotes disulfide bond formation in the cytoplasm | 13, 14 |
| pMAL-c2x<br>(Empty Vector) | Contains the *malE* gene that encodes for the maltose binding protein (MBP)<br>Mutation in the *malE* gene signal sequence allows for protein expression in the cytoplasm | 17 |
| pMAL-c2x-PI2 | Contains the *pi2* gene sequence designed by Fogarty *et al*. inserted between the *malE* and the *lacZα* gene in the pMAL-c2x vector | 7 |

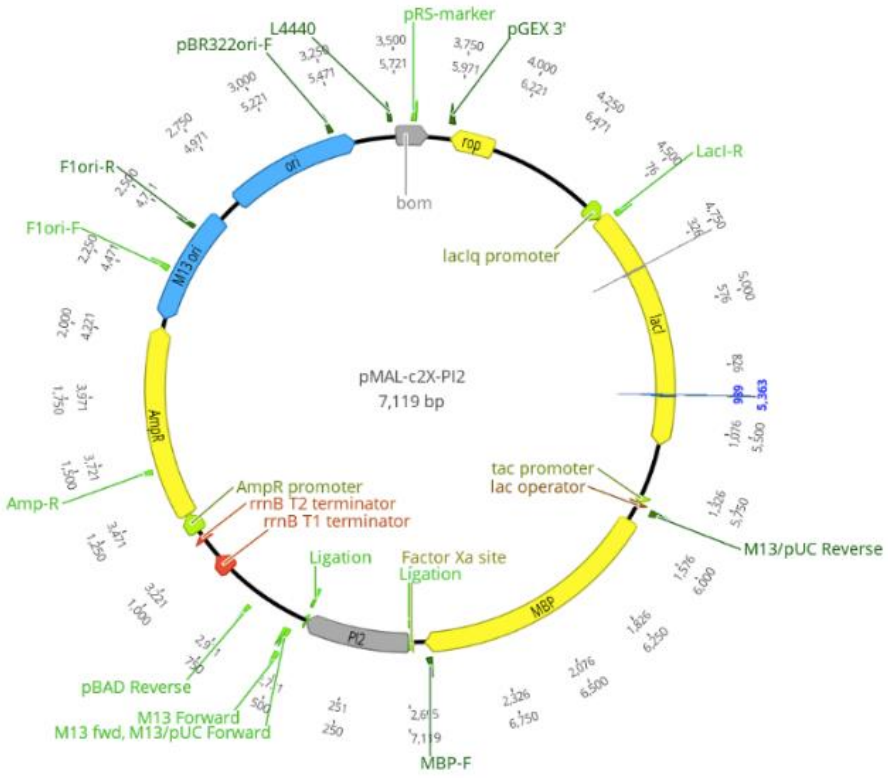


**Figure S1. Map of expression vector pMAL-c2x-PI2 cloned by Lapointe *et al*.** The vector map was generated using Geneious Prime. The *pi2* gene is shown to be inserted in the correct orientation downstream of the lac operon promoter, following the *mbp* gene. Ligation sites are highlighted in green, flanking the *pi2* gene.

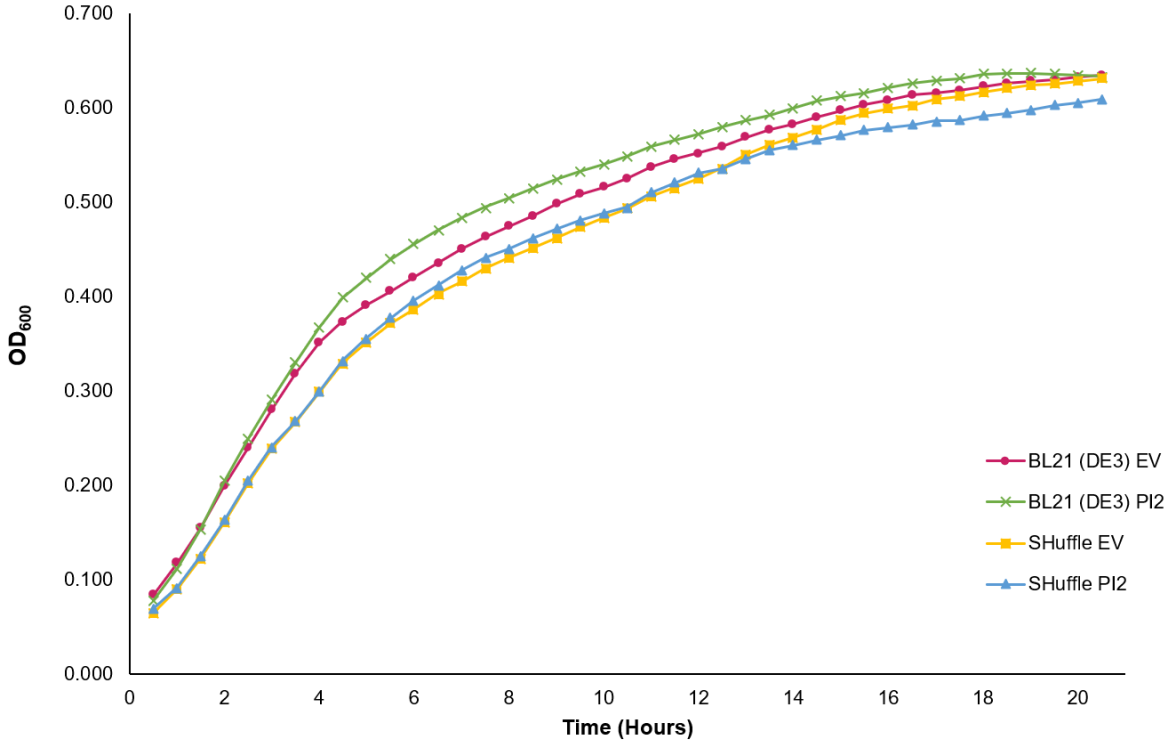


**Figure S2. No significant difference in the growth of SHuffle and BL21 (DE3) with EV and MBP-PI2 was observed.** Each strain was respectively inoculated into LB broth with ampicillin (100 µg/ mL) and grown overnight at 30˚C for 20 hours. The cultures were diluted to $OD_{600}$ of 0.2 in LB broth with ampicillin (100 µg/ mL) and plated in triplicate at a volume of 200 µL/well. LB broth + amp blank was also plated in triplicate to the same volume. The BioTek Epoch 2 Microplate Reader was set to read at $OD_{600}$ every 30 minutes for 20 hours at 30˚C. The plate reader was set to orbital shake continuously at a frequency of 282 CPM.

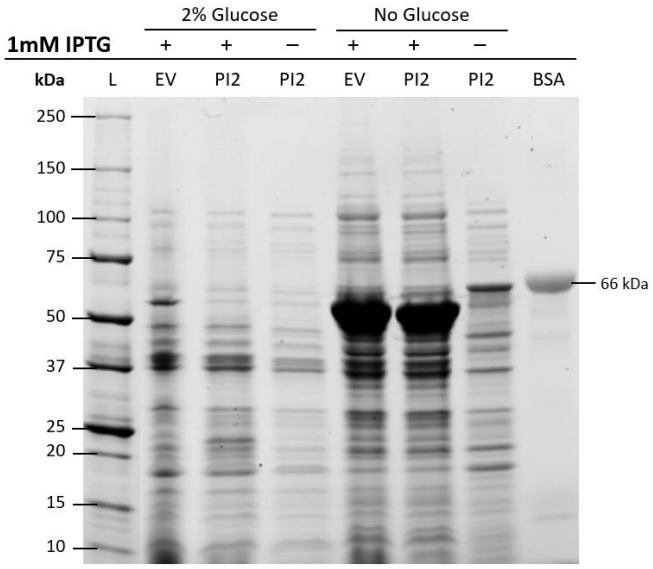


**Figure S3. Glucose content of induction media altered protein expression in Origami 2.** Origami 2 cells transformed with pMAL-c2X (EV) or pMAL-PI2 (PI2) were grown in LB media containing 2 % glucose supplement and not containing glucose supplement. Cultures were then induced overnight with 1 mM IPTG at 30°C. The resulting cell lysates were analyzed by SDS-PAGE. A reference ladder (L) and bovine serum albumin (BSA) served as molecular weight standards. Protein bands were fluorescently visualized using the Bio-Rad Gel Doc EZ System. Bands were absent at the expected molecular weight of MBP-PI2 (66.5 kDa) from the induced PI2-transformed Origami 2 in both 2% glucose and no glucose supplement media conditions. However, the induction of both the EV and PI2-transformed Origami 2 grown without glucose supplement resulted in expression of an unknown protein at 50 kDa. Uninduced PI2-transformed Origami 2 grown without glucose supplement expressed an unknown protein at 66 kDa.