Table 4. Determination of Cla4-dependent phosphorylation sites onto GST-Swe1(K473A) *in vitro*

Peptide	Sequence	Amino acids
1	*ST*G*T*LNLSLSNTALSEANSK 1P (T or S)	63–82
2	W**S*** PFHENESVTTPITK	110–126
3	TNSPI**S*** LK	131–138
4	HNNQTNILSP*T*N*S*LVTNSSPQTLHSNK 1P (T or S)	276–302
5	ARN**S*** VILK	309–316
6	**S*** IIGATSQTHR	379–389
7	ESRPL**S*** LSSAIVTNTTSAETHSISSTDSSPLNSK	390–423
8	LSANPD**S*** HLFEK	432–443
9	EYIAPEII*S*DC*T*YDYK 1P (S or T)	618–633
10	SGDLSDAGRL*SST*DIHSESLFSDITK 1P (S or T)	664–689
11	LSSTDIHSESLFSDI**T*** KVDTNDLFDFER	673–700
12	LSSTDIH*S*E*S*LF*S*DI*T*KVDTNDLFDFER 2P (S and/or T)	673–700

Phosphorylation sites were determined as in Table 3. A Swe1 derivative bearing mutations in the Cla4 phosphorylation sites [Swe1(12A)] was generated by mutagenizing T66, S111, S136, S288, S312, S379, S395, S438, T629, T676, S682, and T688 to Alanine (A). In peptides 1, 2, 5, 6, 7, and 10–12, there is a Ser (or Thr) situated downstream of an Arg (removed by trypsin digestion) in a context that fits the Cla4 consensus motif (see text); however, other apparent Cla4 sites indicated in these and the other peptides appear to fit a looser consensus, S/T followed by a bulky nonpolar residue at positions +1 or +2. The phosphorylation site in peptide 2 is identical to one of the three *in vivo* phosphorylation sites determined so far (see footnote in Table 3).