

Table I

MS:MS analysis of the signal at 658.3597 identifying a conjugate comprising residues 21-23 and 1037-1042 of the PhK regulatory β subunit.

Lower and upper case letters denote ions arising from amide fragmentation of the peptide backbone and the GMBS cross-link, respectively. C', C and B refer respectively to fragments of the intervening cross-link that result from c and b type cleavages after the maleimido nitrogen, butyryloxy carbonyl and ϵ -amine nitrogen of K22. Intact cross-links (:) or covalent attachment of a fragment of the cross-link (·) are so noted under the ion heading. The immonium ion of K22 is denoted by a_{3-3} .

m:z Experimental	Relative intensity	ion ^{a,b,c}	sequence	m:z Calculated	error ppm
217.0755	0.80	b_{2-3}^+ or a_2^+	ES or DE	217.0819	30.0
245.0763	1.79	b_2^+	DE	245.0768	5.30
262.0418	3.89	c_2^+	DE	262.1034	234
284.0638	0.77	$(b_4^0 \cdot C')^{2+}$	DESR	284.1118	169
341.1742	0.94	$(c_2 : b_{4-5})^{2+}$	TK : RL	341.2059	93.0
409.3294	1.74	$(a_2^* : b_{2-5}^0)^{2+}$	TK : ESRL	409.2124	286
425.4752	1.45	$y_2^* : y_3^{*2+}$	KR : RLK	425.4752	0.127
430.364	19.44	$(y_6^* \cdot B-2H_2O)^{2+}$	DESRLK	430.2010	378
470.2997	0.71	$(a_2 : a_5)^{2+}$	TK : DESRL	470.2483	109
477.6212	0.99	$(y_2 : b_{2-5})^{2+}$	KR : ESRL	477.2620	753
493.3414	1.01	$(a_2 : b_5 + H_2O)^{2+}$	TK : DESRL	493.2530	179
512.3757	2.32	$(y_2 : a_5^*)^{2+}$	KR : DESRL	512.2645	217
518.7748	3.02	$(y_3 : b_{2-5}^0)^{2+}$	TKR : ESRL	518.7783	10.0
528.0253	100.00	$(y_3^0 : c_4)^{2+}$	TKR : DESR	528.2650	453
536.4355	1.25	$(y_3 : y_4)^{2+}$	TKR : SRLK	536.3171	221
539.1461	2.17	$(x_2 : b_5^0 - 17)^{2+}$	KR : DESRL	539.2885	264
544.1265	2.28	$(b_2^0 : y_6^0 - H_2O)^{2+}$	TK : DESRLK	544.3705	448
585.5363	4.34	$(y_3 : b_5)^{2+}$	TKR : DESRL	585.2991	405
587.6885	6.71	$(y_3^* : x_5 - 17 - H_2O)^{2+}$	TKR : ESRLK	587.8097	206
590.5178	3.52	$(y_2^* : y_6^0)^{2+}$	KR : DESRLK	590.3095	353
601.3137	2.72	$(a_2^* : b_{4-5}^0)^+$	TK : RL	601.356	71.0
615.2781	1.61	$(y_5^*)^+$	ESRLK	615.3466	111
623.2832	2.02	$(a_{2-5}^* \cdot C)^+$	ESRL	623.3154	51.6
635.7613	3.23	$(a_{3-3} : a_{2-4})^+$	K : ESR	635.3142	703
640.4674	11.05	$(y_3^0 : y_6^0)^{2+}$	TKR : DESRLK	640.3233	197
641.4561	11.18	$(y_3^* : y_6^*)^{2+}$	TKR : DESRLK	641.3253	204
646.4799	6.89	$(b_2^0 : b_{4-5})^+$	TK : RL	646.3667	175
649.6328	10.07	$(y_3^* : y_6)^{2+}$	TKR : DESRLK	649.8385	316
670.5092	1.36	$(b_4 \cdot C)^+$	DESR	670.2791	343
717.6456	0.78	$(b_2^* : b_{3-5}^*)^+$	TK : SRL	717.3567	400
761.4265	1.49	$(y_3^* : b_{3-4}^* - 17)^+$	TKR : SR	761.3570	91.0
777.2460	0.42	$(y_3^* : b_{3-4}^0)^+$	TKR : SR	777.3997	198
789.6215	0.57	$(y_2^* : b_{3-5}^0)^+$	KR : SRL	789.4415	228
795.6428	1.10	$(y_2^* : a_{2-4})^+$	KR : ESR	795.4100	292
801.0935	1.25	$(b_5 \cdot C + H_2O)^+$	DESRL	801.3730	350
874.5126	5.95	$(a_{3-3} : b_5^0)^+$	K:DESRL	874.4714	108
897.4690	0.44	$(b_2 : y_4)^+$	TK : SRLK	897.4690	61.0
909.6128	2.59	$(y_2 : a_4 - H_2O)^+$	KR : DESR	909.4621	166
940.5364	0.82	$(c_2 : x_4)^+$	TK : SRLK	940.5294	7.50
957.6760	1.23	$(z_2 : b_4 + H_2O)^+$	KR : DESR	957.4145	273
968.6324	0.78	$(y_3 : z_3)^+$	TKR : RLK	968.5845	50.0
985.4358	2.28	$(a_2 : b_5 + H_2O)^+$	TK : DESRL	985.4948	60.0
1039.615	0.46	$(y_3^* : b_4^*)^+$	TKR : DESR	1039.4886	121
1054.669	17.52	$(y_3 : y_4^*)^+$	TKR : SRLK	1054.6086	58.0
1151.717	0.34	$(y_3 : b_5^0)^+$	TKR : DESRL	1151.5887	111
1169.831	2.64	$(y_3 : b_5)^+$	TKR : DESRL	1169.5992	198
1180.738	1.07	$(y_2^* : y_6^*)^+$	KR : DESRLK	1180.6040	113
1187.445	0.32	$(y_3 : b_5 + H_2O)^+$	TKR : DESRL	1187.6010	131

^aThe MH⁺ ions of TKR and DESRLK are denoted, respectively, as y₃ and y₆.

^bFor singly charged ions, it should be noted that one of the two peptide ions covalently attached to either functional group of GMBS is a neutral product of the indicated backbone amide cleavage.

^cInternal fragments and internal fragments-28 Da are respectively labeled b_{1-x} and a_{1-x}.