Biochemistry Cumulative Examination
November 5, 2020, Total 100 points

1. Epidermal growth factor receptor1 (EGFR) contains multiple tyrosine residues in the cytosolic domain, which are phosphorylated by autophosphorylation (or by other kinases) upon epidermal growth factor (EGF) binding. Resulting phosphotyrosine (pY) residues are recognized by various adaptor proteins containing pY-specific SH2 or PTB domains (see Fig. 1), leading to a wide range of cellular signal transduction.

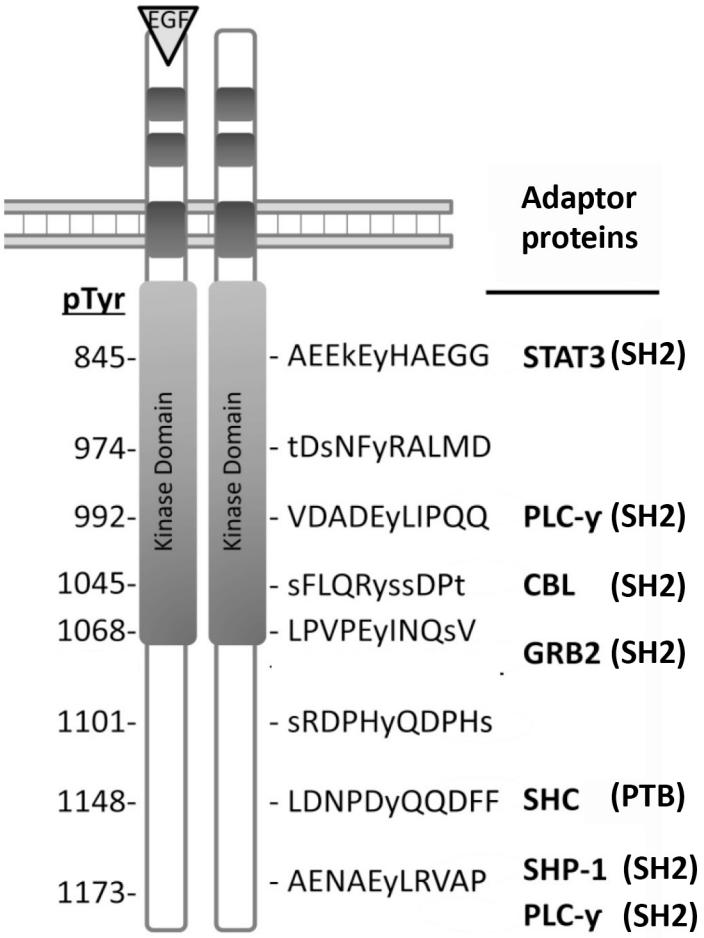

Fig. 1

1. Brian plans to develop a specific biosensor that detects the formation of pY1068 using the GRB2 SH2 domain as a template. He thus prepared several mutants of GRB2 SH2 with different affinity (see Table 1) for a peptide, LPVPE(pY)INQ, which corresponds to the sequence of EGFR that is recognized by GRB2-SH2 (see Fig. 1).

Table 1 Affinity of GRB2-SH2 wild type (WT) and mutants for LPVPE(pY)INQ

Grb2-SH2 proteins	WT	Mutant1	Mutant2	Mutant3
Affinity (μM)	0.5	2	0.1	0.001

(1) In terms of the affinity for the pY peptide alone, which of the four proteins serves as the best biosensor for detecting pY1068 in EGFR and explain why (20 points)

(2) Beside the affinity for the pY1068 peptide, described at least two other critical factors for successful preparation of a biosensor for pY1068 of EGFR (20 points)

(3) Describe how to construct a biosensor and "**quantitatively**" determine pY1068 of EGFR. Describe the design principle and preparation steps (30 points)

2. EGFR is often overexpressed or hyperactivated through mutation(s) in cancer and is thus a major target for cancer drug development. Propose at least two different strategies to inhibit EGFR in cancer and describe the most important factors in EGFR-directed drug development (30 points).