Exercise walk-through

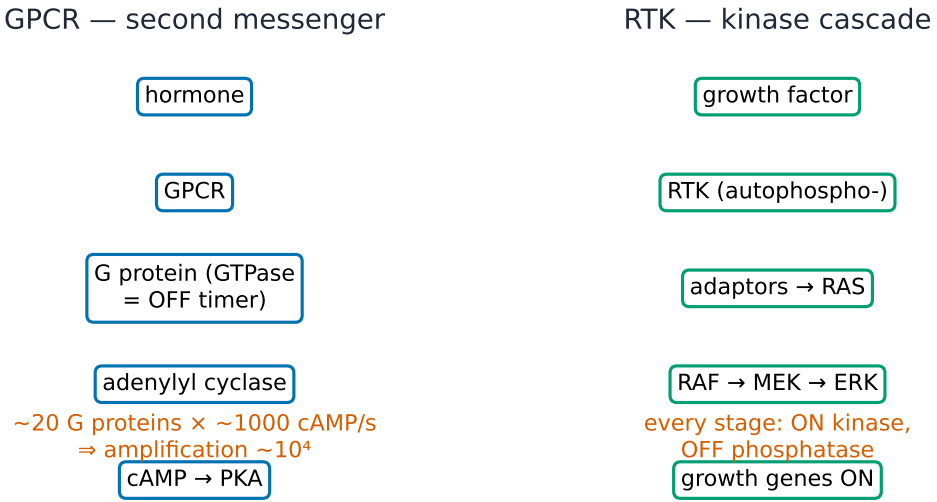
1.6
Cell signalling

University Medicine

Signalling: amplify and reset

1. **Ledger** Resting cAMP $\approx$ 12 000 molecules per hepatocyte; ten cyclases at 1000/s replace the pool in a second - OFF matters as much as ON.
2. **Switch** cAMP switches 10 nM $\rightarrow$ 1 µM (a hundredfold) in seconds - a maximal hormone pulse.
3. **Welded** Cholera toxin disables the Gαs GTPase: cyclase runs constitutively, Cl^- and water pour out - the OFF enzyme IS the story.

Two cascades



GPCR-cAMP and RTK-RAS side by side: $\sim 10^4$ amplification, every stage reset-table.

Exercises 2.1·2.3

2.1 Name the OFF timer of the Gα subunit. [3]

2.2 Compute the cAMP fold-change from 10 nM to 1 µM. [3]

2.3 Why do GPCR ligands not enter the cell? [3]

Exercises 2.1·2.3

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2.1

- Its intrinsic GTPase: GTP $\rightarrow$ GDP, seconds.

2.2 Compute the cAMP fold-change from 10 nM to 1 µM. [3]

2.2

- $10^{-6}/10^{-8} = 100\times$.

2.3 Why do GPCR ligands not enter the cell? [3]

2.3

- The message is relayed by the transmembrane receptor and second messengers; hydrophilic ligands cannot cross the bilayer (only lipophilic steroids enter, by a different mechanism).

Exercise 3.1

3.1 A maximal hormone pulse.

Compute the cAMP molecules at 1 µM in a 2×10^{-12} L cell (compare the 12 000 resting). Explain the amplification chain that produces them. State two OFF mechanisms and their timescale. [8]

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3.1

- $10^{-6} \times 2 \times 10^{-12} \times N_A \approx 1.2 \times 10^{6}$ molecules – 100× the 12 000 resting ✓.
- Hormone → receptor → ~20 Gs → cyclases → cAMP: stage gains multiply (~10^4).
- OFF: Gα GTPase (seconds); PDE destroying cAMP (seconds).

Exercise 3.2

3.2 Cholera toxin (fictional vignette for the mechanism).

State the molecular modification and its direct effect on Gαs. Trace the cascade to luminal Cl^- and water loss. Explain why therapy is fluid replacement rather than an inhibitor. [7]

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3.2

- ADP-ribosylation of Gαs; GTPase disabled $\rightarrow$ Gs permanently on.
- Constitutive cyclase $\rightarrow$ maximal cAMP $\rightarrow$ PKA $\rightarrow$ Cl^- channel open $\rightarrow$ Cl^- and water follow into the lumen.
- No inhibitor can unweld an already-modified protein pool; support while proteins turn over.