**Kongres XI dan Seminar Nasional XIX**
**Perhimpunan Biokimia dan Biologi Molekuler Indonesia**
**(PBBMI)**

# SERTIFIKAT

*Diberikan Kepada:*

## Prof. Dr. Ernawati Sinaga, MS. Apt

*Sebagai:*

# PEMBICARA

Dalam kegiatan seminar dengan tema:

***"Biochemistry and Molecular Biology are on The Cutting Edge of Research Trend"***

yang diselenggarakan pada 12 – 14 November 2015 di Denpasar, Bali

Peserta : 10 SKP, Pembicara : 12 SKP, Moderator : 4 SKP, Panitia : 2 SKP

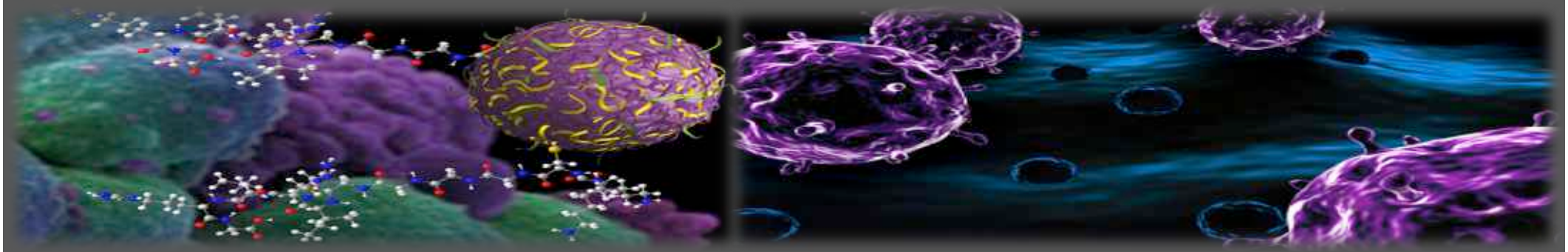
(SKP IDI No: 06/XI/2015/SKP/IDI-BALI)

Panitia Kongres XI dan Seminar Nasional XIX
Perhimpunan Biokimia dan Biologi Molekuler Indonesia
(PBBMI)
Ketua,

Dr.rer.nat.dr.Ni Nyoman Ayu Dewi, M.Si

Perhimpunan Biokimia dan Biologi Molekuler Indonesia
(PBBMI)
Ketua,

Prof. dr. Nur Asikin, PhD



# MODULASI *JUNCTION* ANTARSEL

## STRATEGI MENINGKATKAN PENGHANTARAN OBAT

Ernawati Sinaga
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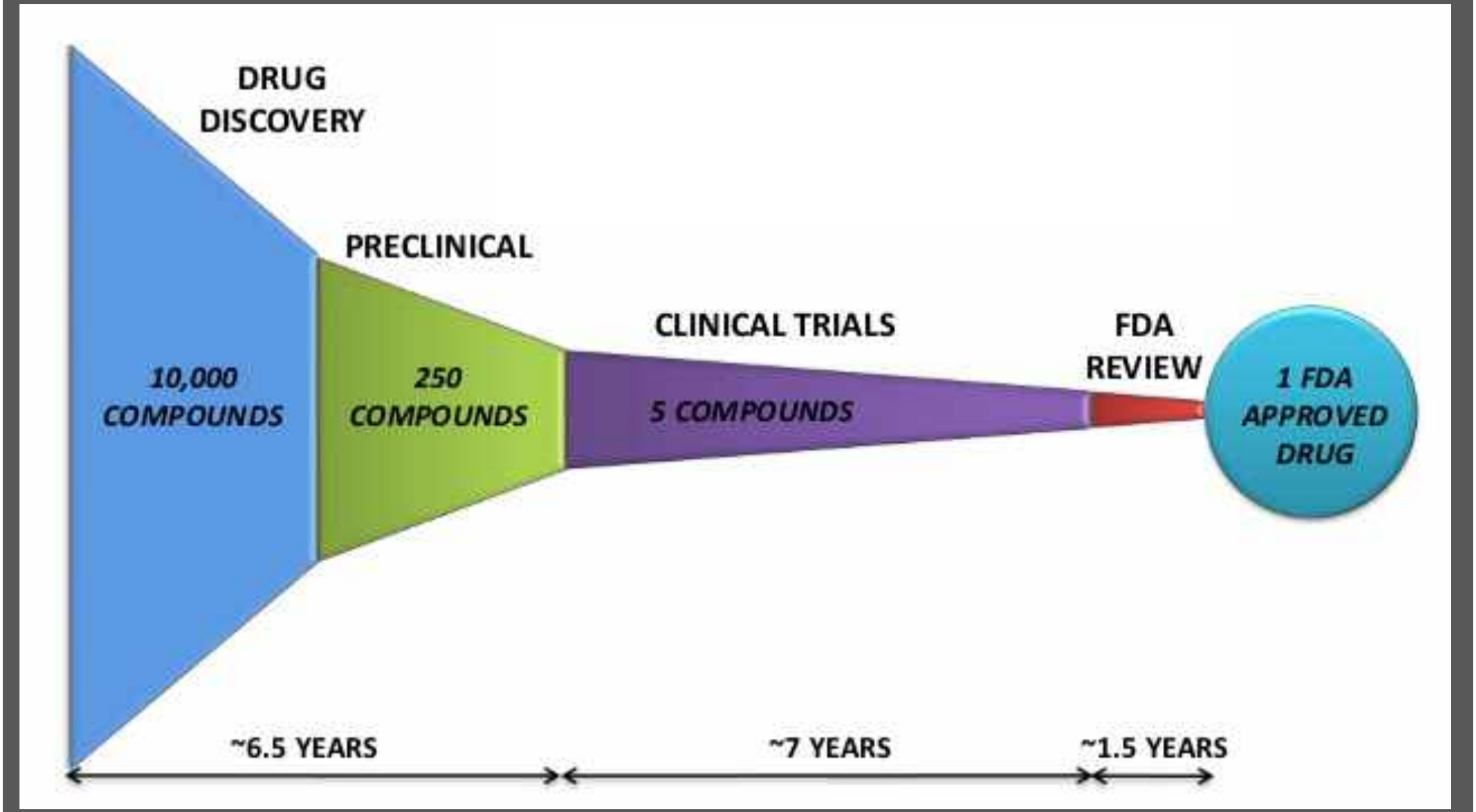
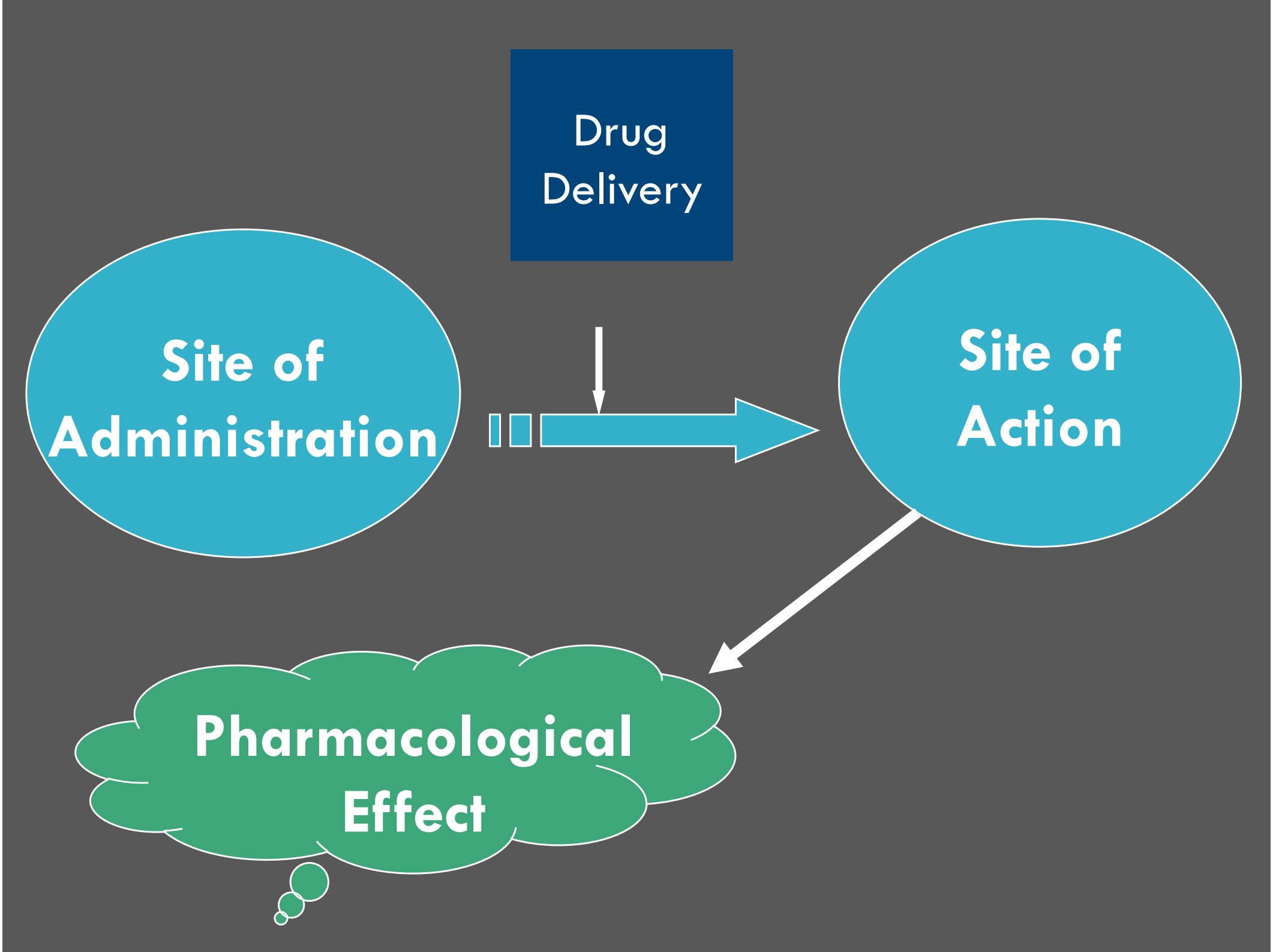
# Drug Discovery & Development

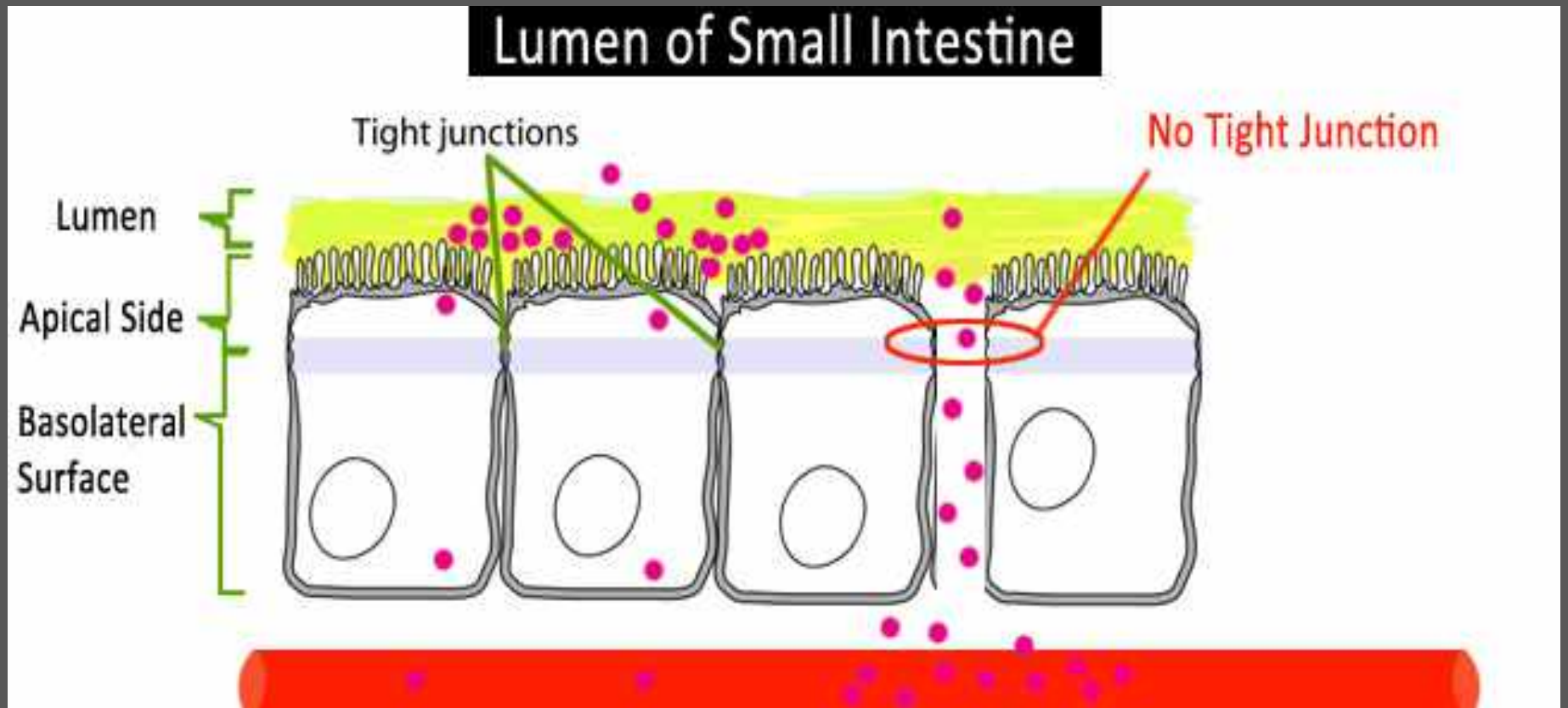


# Drug Discovery & Drug Development

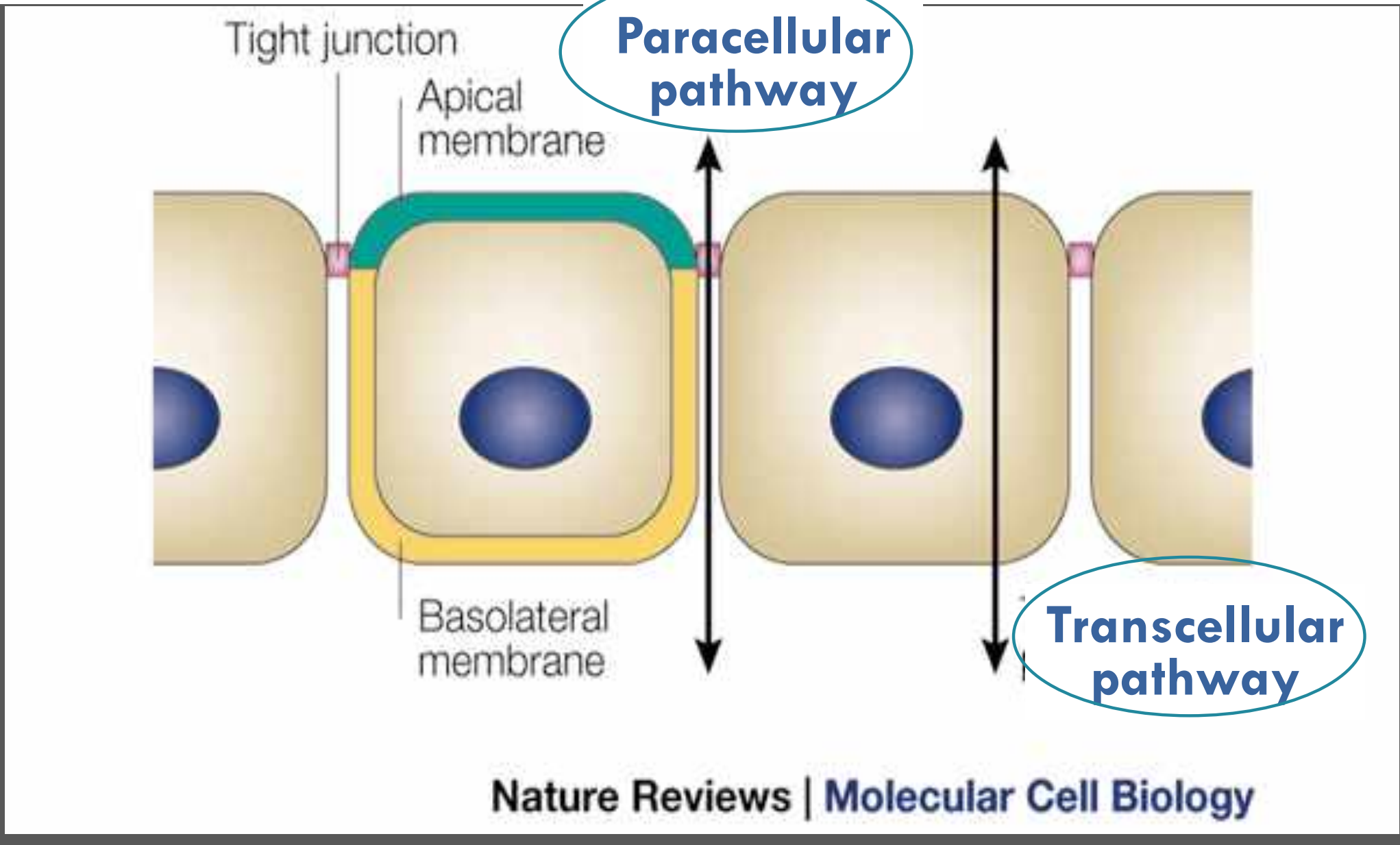

Drug Delivery
Site of Administration
Site of Action
Pharmacological Effect

# Biological Barrier

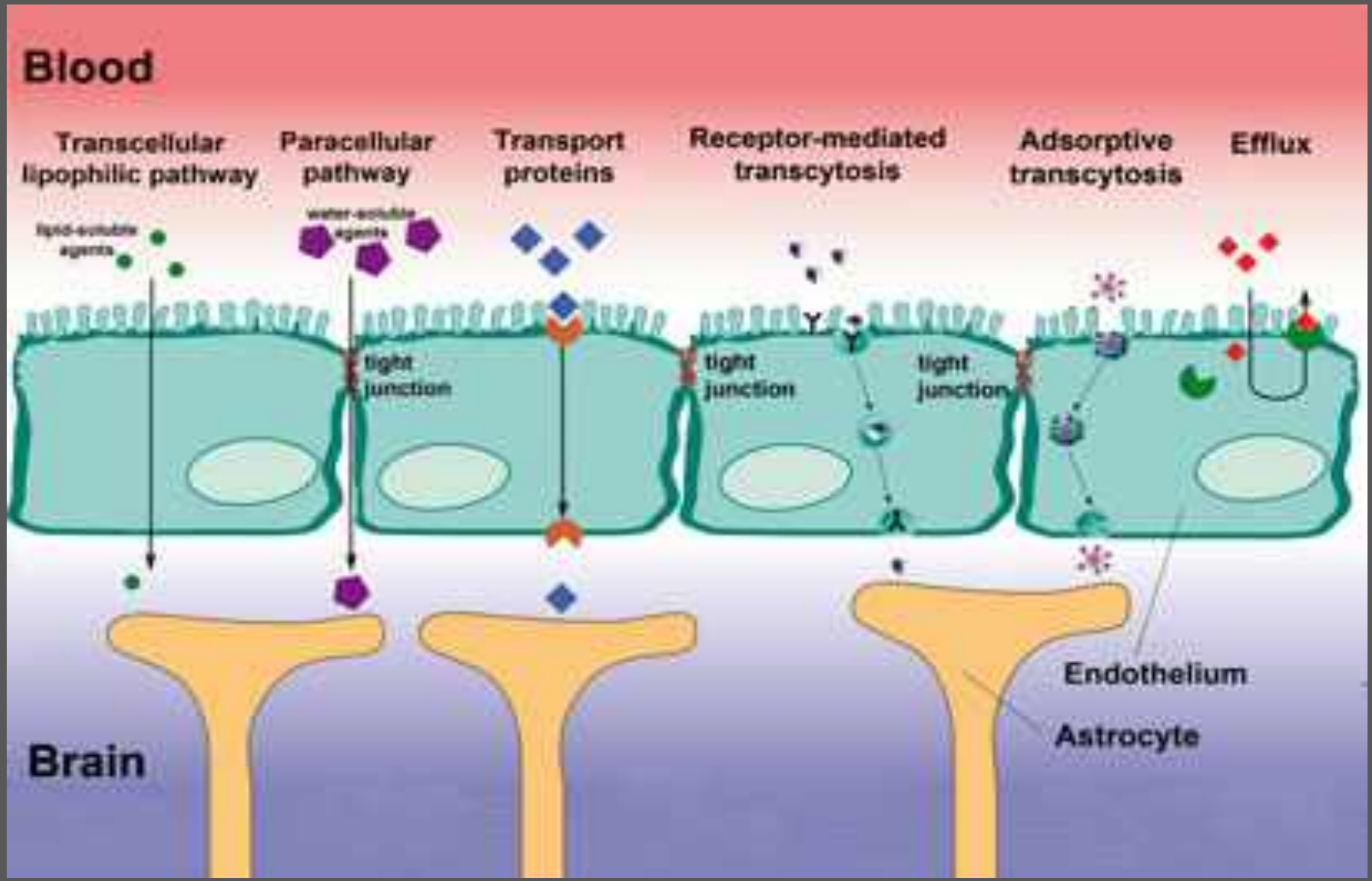
- Blood-Brain Barrier
- Blood-Kidney Barrier
- Intestinal Barrier
- Skin Barrier



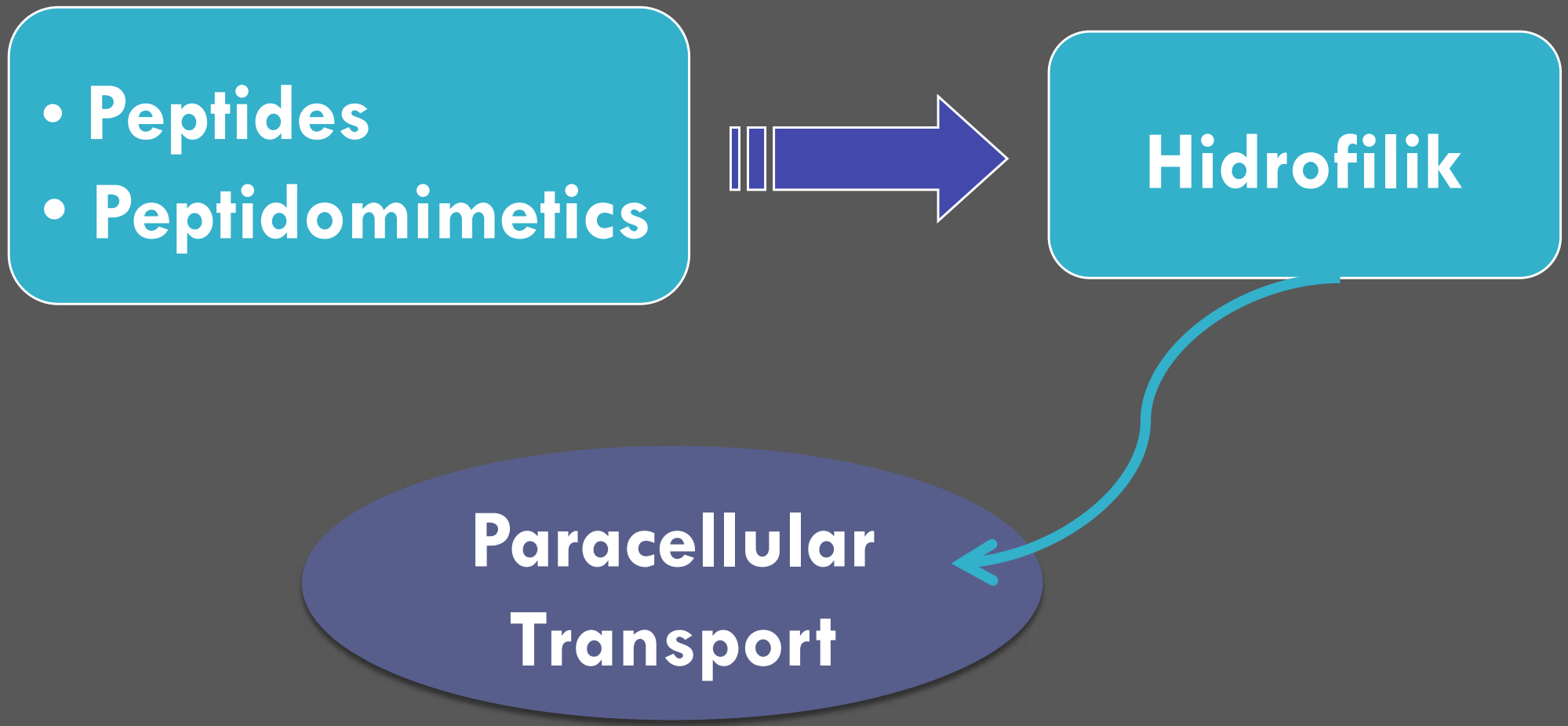
# Transport of Molecules

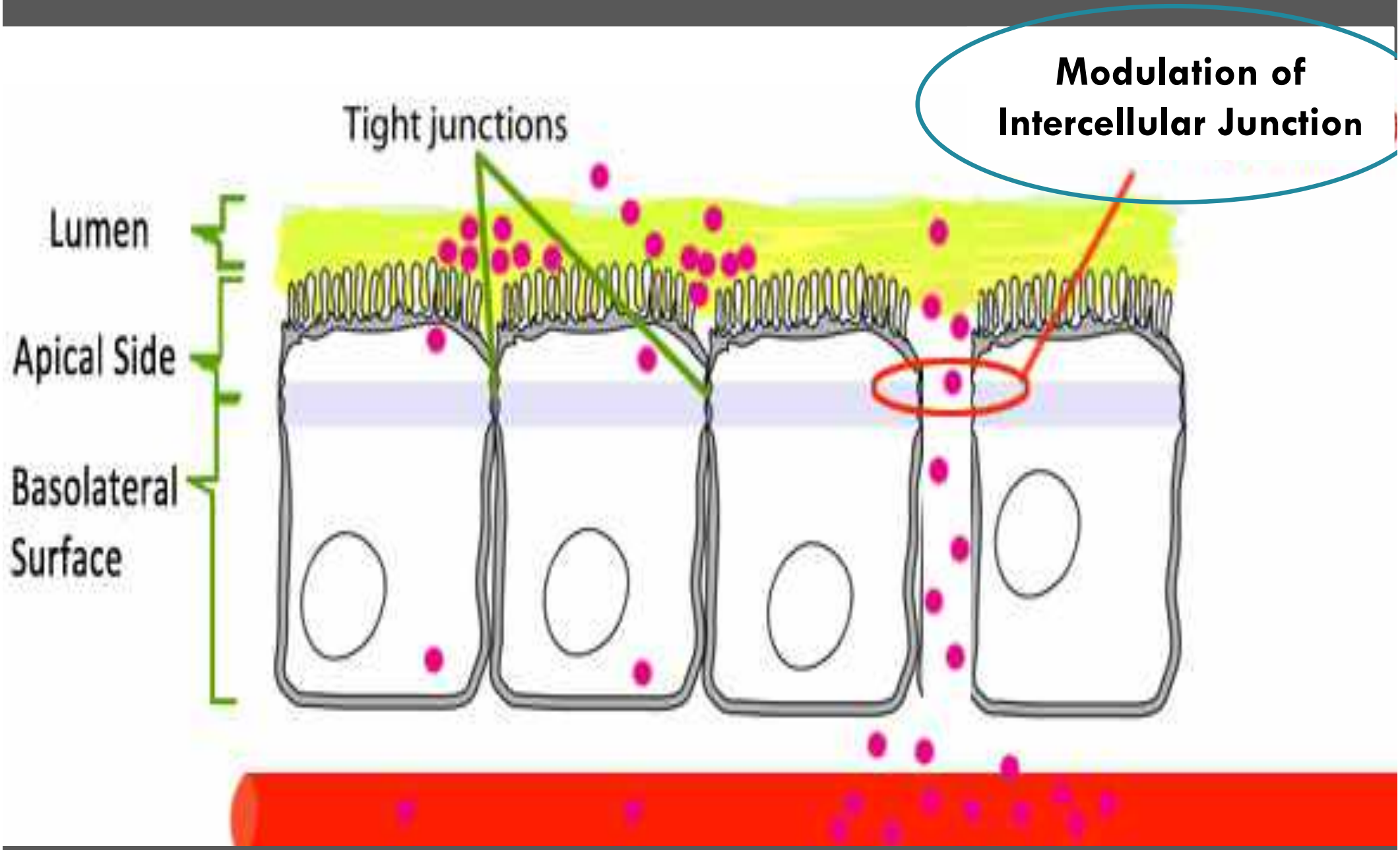


# Blood-Brain Barrier

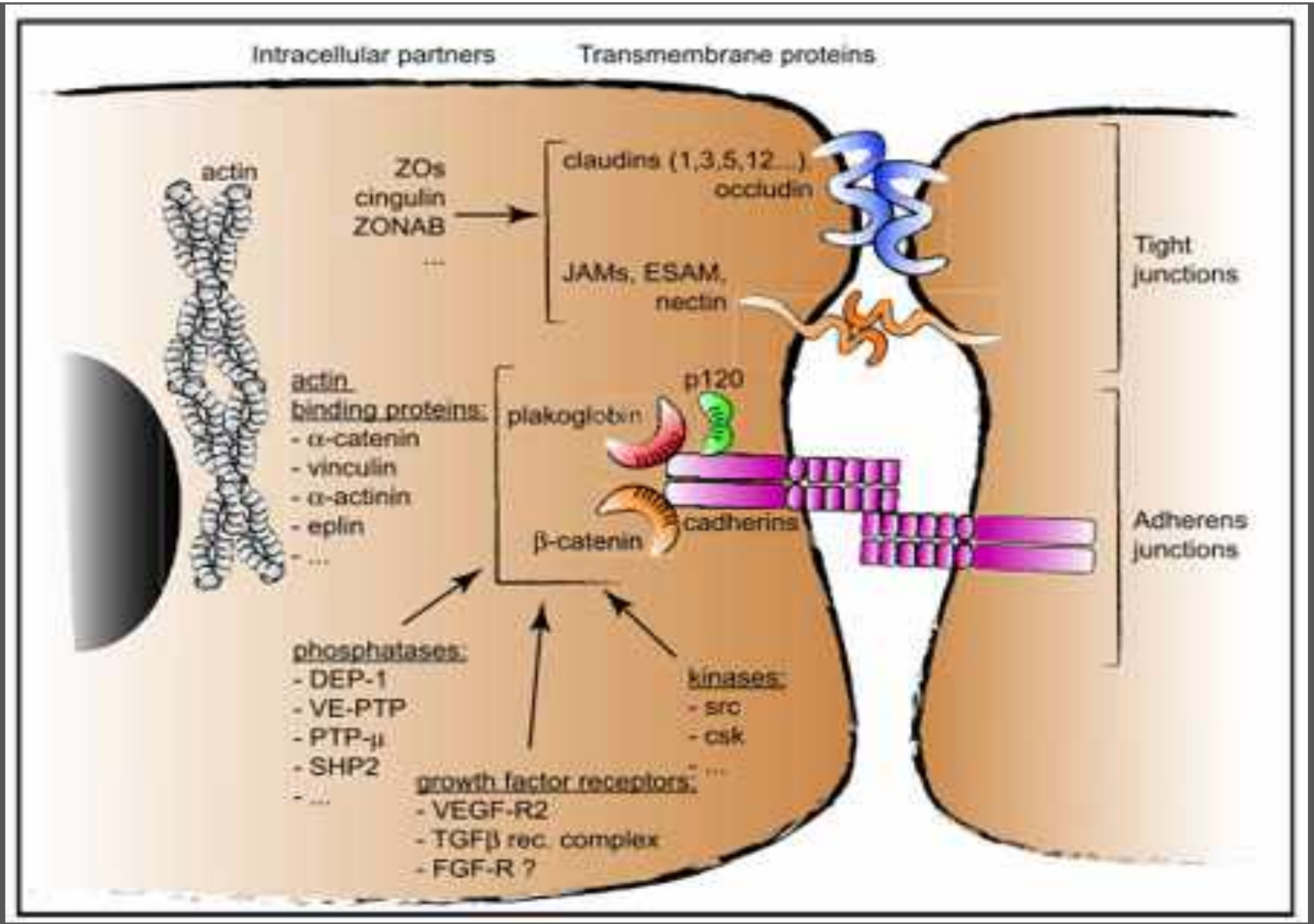


# New Drug Molecules

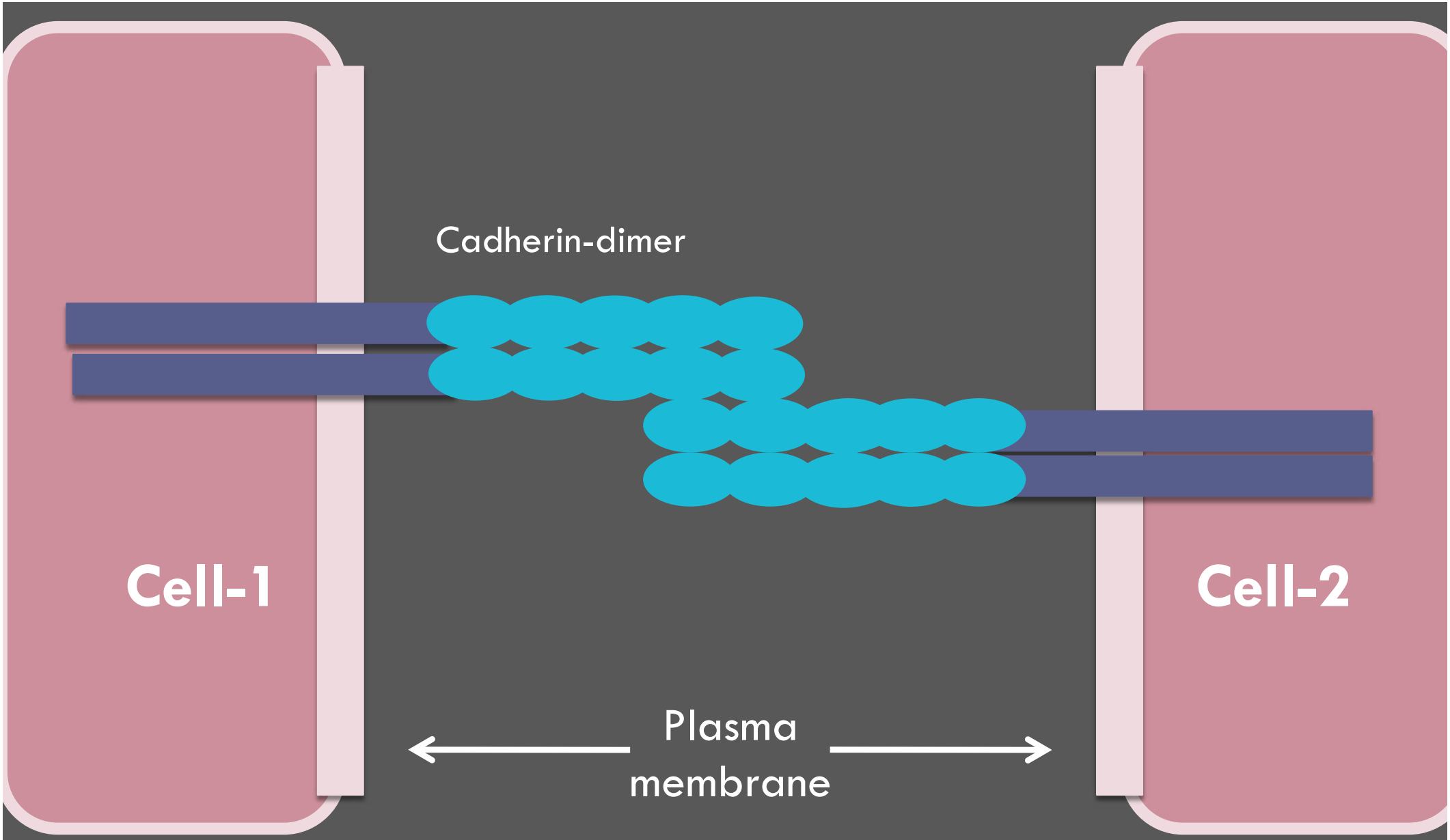



Modulation of Intercellular Junction
Tight junctions
Lumen
Apical Side
Basolateral Surface

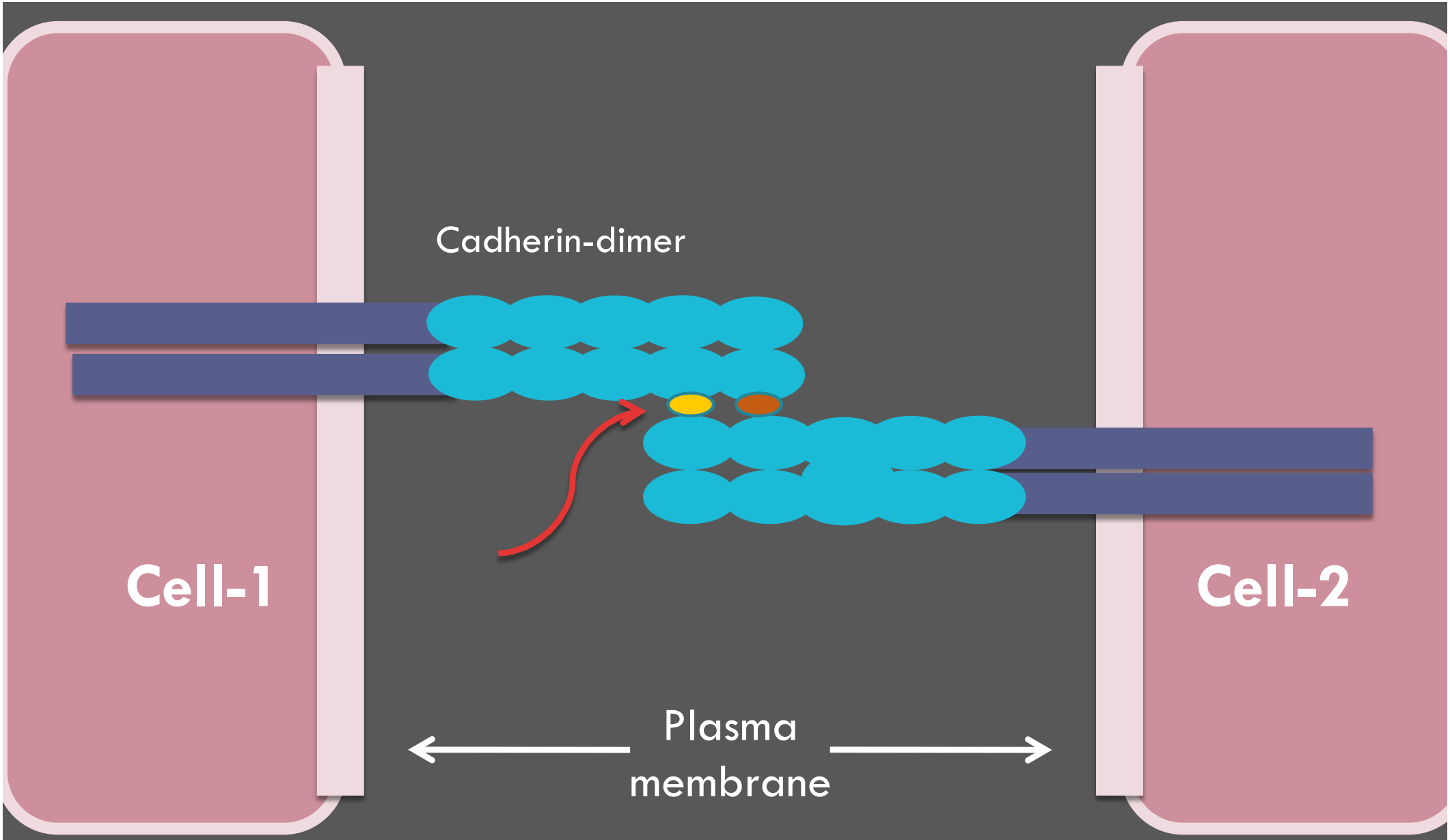
# Modulation of Intercellular Junction

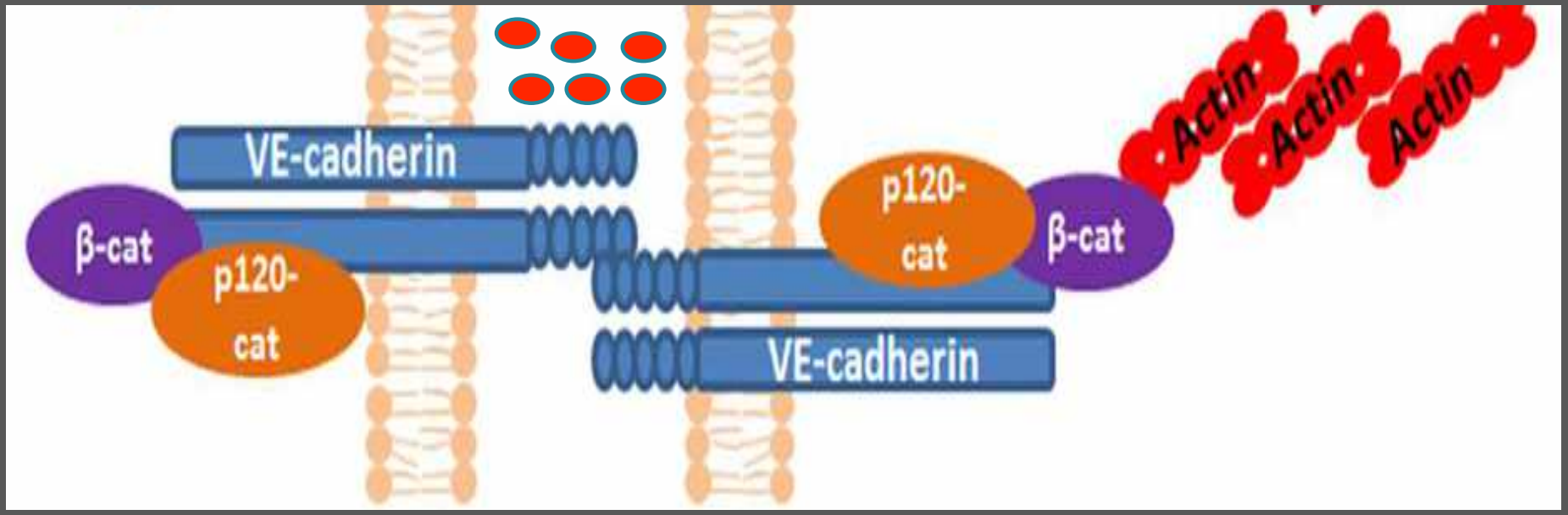


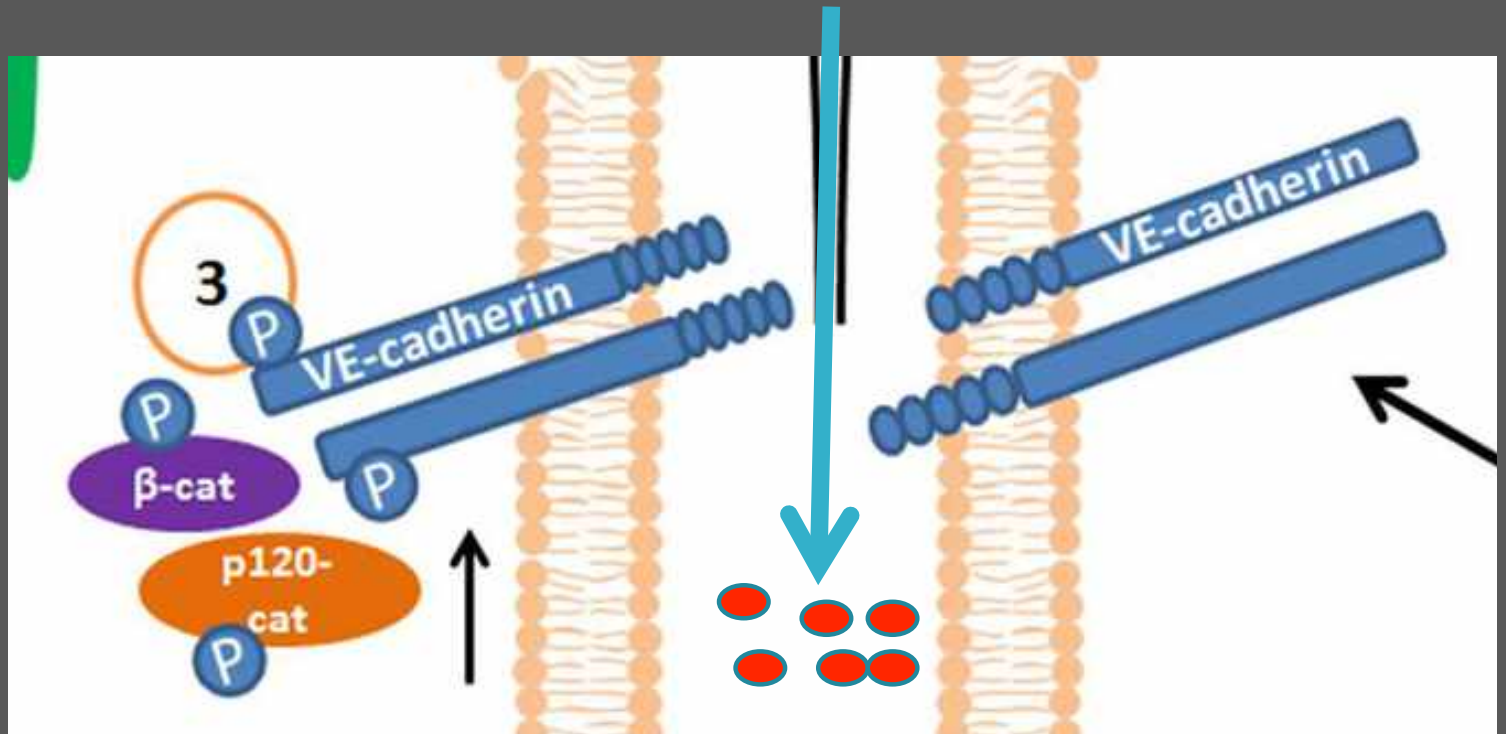
# Cadherin interaction



# Cadherin interaction



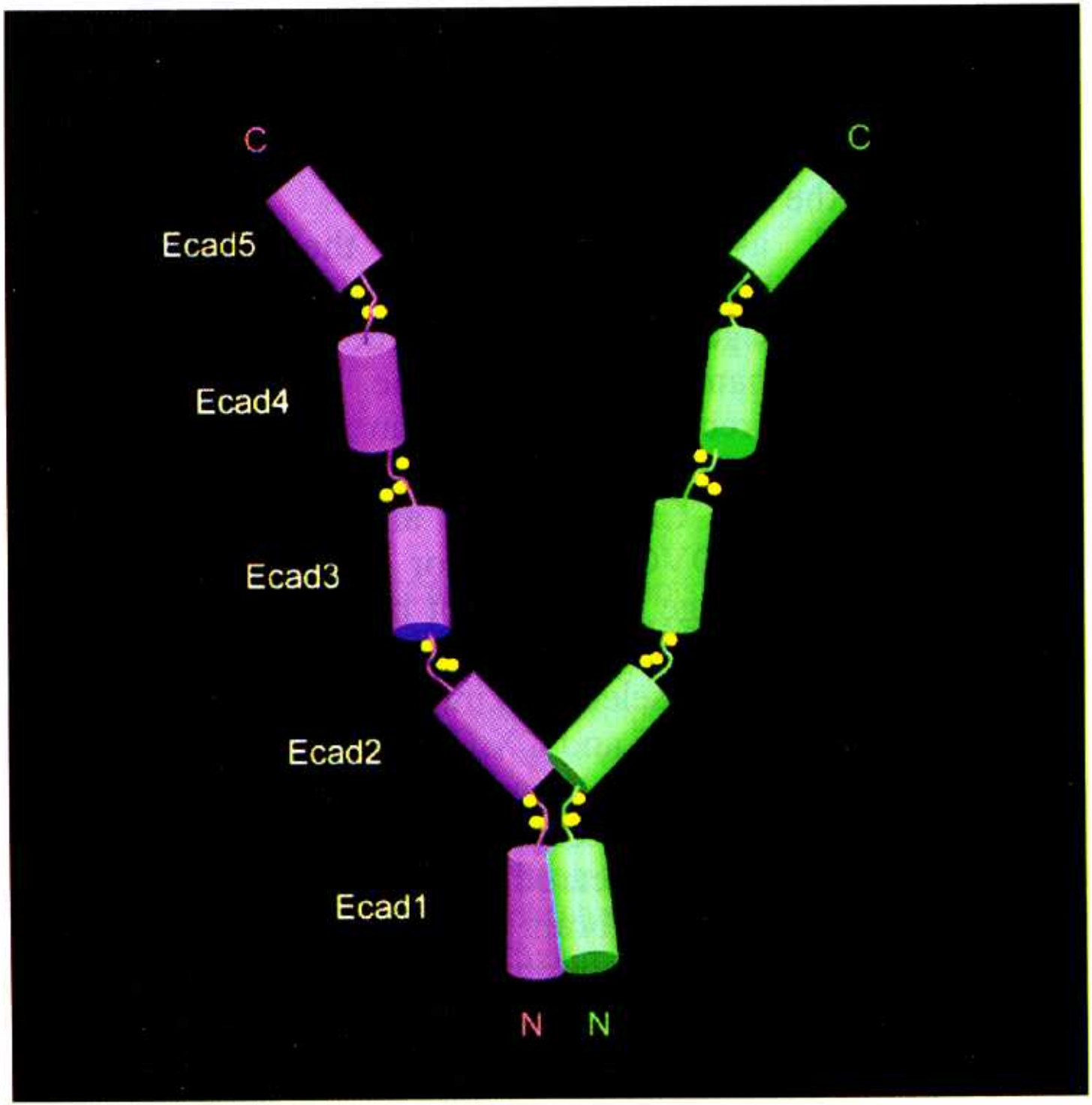

VE-cadherin
β-cat
p120-
cat
p120-
cat
β-cat
Actin
Actin
Actin
VE-cadherin


3
P
VE-cadherin
P
P
β-cat
p120-
cat
P
VE-cadherin

# Modulation of intercellular junction

- Calcium Chelating agent : EDTA, EGTA
- Surfactant : Na dodecyl sulphate
- Bile acids
- Hormone and neurotransmiter
- Cytocalasine B dan D
- Inhibitor of glycolysis and phosphorilative oxydation
- Modulation of cadherin

EC-domain (simple dimer)



# Human E-Cadherin

dwvippiscpenekgp fpknlvqiksnkdkegkvfysitg **qgadtppvgv** fiiere tgwlkvtepl dreriatyt **lfshavssng** naved pmeilitvt**dqndn**kp eftqevfkgsvmegalpgtsvmevtatdadddvntynaaiaytilsqdpelpdknmfti nrntgvisvvttgldresfptytlvvqaadlqgeglsttatavitvtdtndnppifnp ttykgqvpeneanvvittlkvtdadapntpaweavytilnddggqfvvttnpvnndgilk takgldfeakqqyilhvavtnvvpfevslttstatvtvdvldvneapifvppe krvevsedfgvgqeitsytaqepdtfmeqkityriwrdtanwleinpdtgaistraeldred fehvknstytaliiatdngspvatgtgtlllilsdvndnapipeprtiffcernpkpq viniidadlp pntspftaelthgasanwti qyndptqesiilkpkmalevgdykinlklm dnqnkdqvtt levsvcdceg aagvcrkaqp veaglqipai lgilggilallililllllf lrrravvkep llppeddtrd nvyyydeegg geedqdfdls qlhrgldarpevtrndvapt lmsvprylpr panpdeignf idenlkaadt dptappydsllvfdyegsgseaaslsslns sesdkdqdyd ylnewgnrfk kladmyggge dd

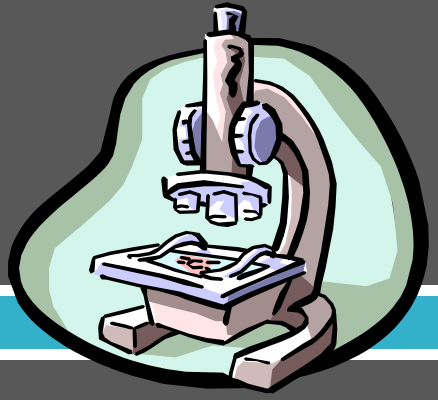
| Nama | Simbol 3 H | Simbol 1 H |
|---|---|---|
| **Glisin** | **Gli** | **G** |
| **Alanin** | **Ala** | **A** |
| **Prolin** | **Pro** | **P** |
| **Valin** | **Val** | **V** |
| **Leusin** | **Leu** | **L** |
| **Isoleusin** | **Ile** | **I** |
| **Fenilalanin** | **Phe** | **F** |
| **Metionin** | **Met** | **M** |
| **Serin** | **Ser** | **S** |
| **Treonin** | **Tre** | **T** |

| Nama | Simbol 3 H | Simbol 1 H |
|---|---|---|
| **Tirosin** | **Tyr** | **Y** |
| **Asparagin** | **Asn** | **N** |
| **Glutamin** | **Gln** | **Q** |
| **Histidin** | **His** | **H** |
| **Triptofan** | **Trp** | **W** |
| **Sistein** | **Cys** | **C** |
| **Asam Aspartat** | **Asp** | **D** |
| **Asam Glutamat** | **Glu** | **E** |
| **Lisin** | **Lys** | **K** |
| **Arginin** | **Arg** | **R** |

# Peptida Kadherin

1. Ac-SHAVSS-NH2 (HAV-6)
2. Ac-LFSHAVSSNG-NH2 (HAV-10)
3. Ac-ADTPPV-NH2 (ADT-6)
4. Ac-QGADTPPVGV-NH2 (ADT-10)
5. Ac- SSVAHS-NH2 (VAH-6)
6. Ac-ADKPPV-NH2 (ADK-6)
7. ADK6-aminocaproic-HAV6
8. HAV6-aminocaproic-ADK6

# Parameter

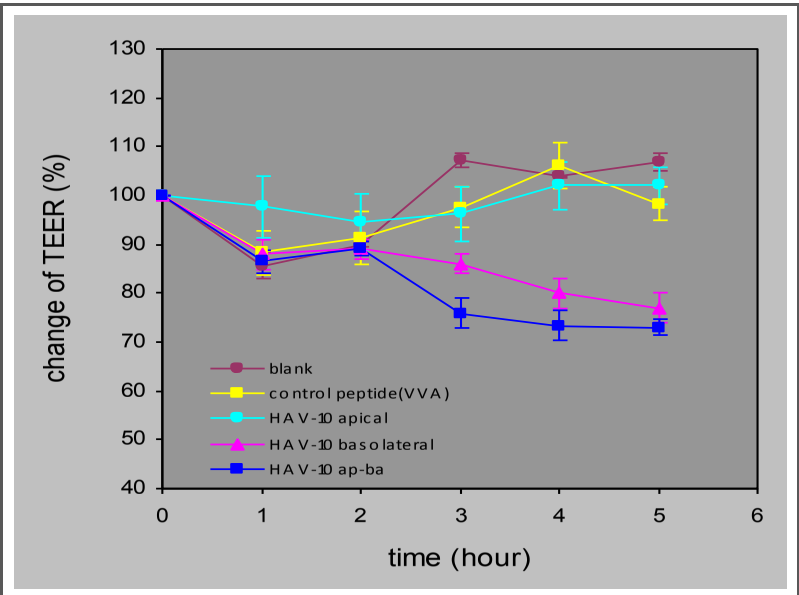


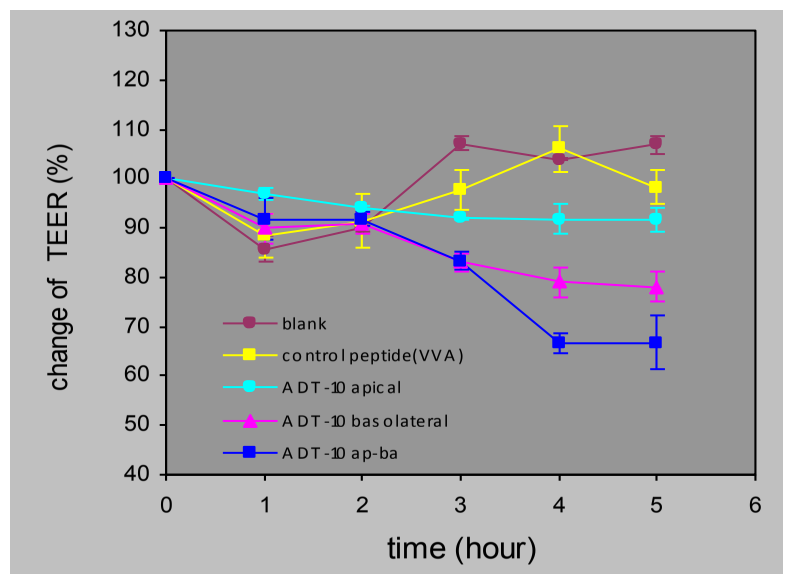
- TEER (Transepithelial Electrical Resistance)
- Cell adhesion (cell reaggregation)
- Paracellular transport ($^{14}$C-Mannitol)

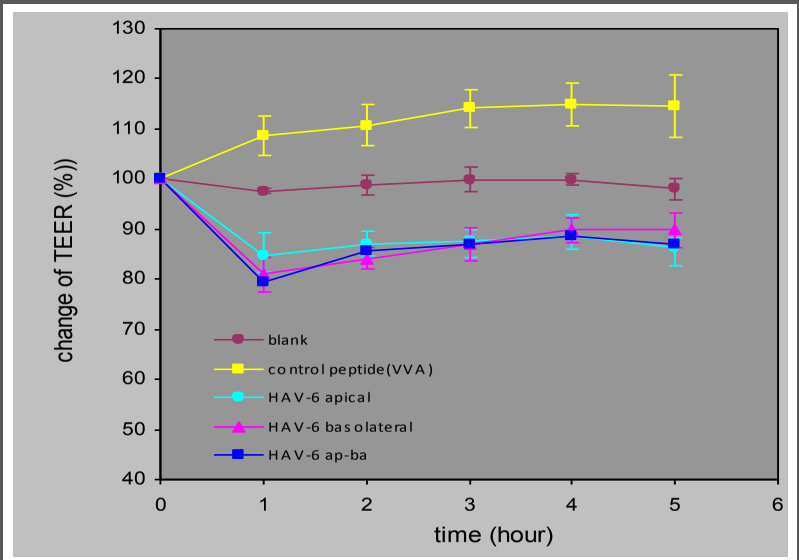
# Tahap Pekerjaan

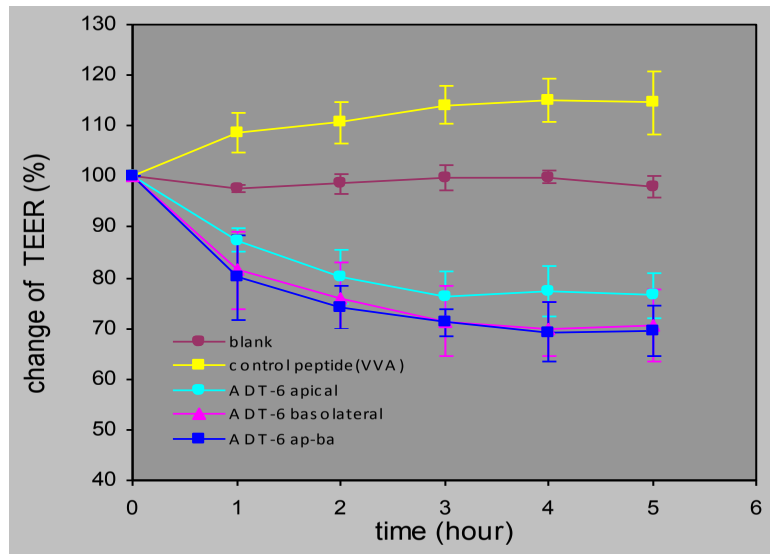
- Sintesis & pemurnian peptida (HAV-10, HAV-6, ADT-10, ADT-6, dll)
- Kultur sel (MDCK dan CACO-2)
- Analisis efek peptida terhadap nilai TEER
- Analisis efek peptida terhadap daya reagregasi sel
- Analisis efek peptida dalam meningkatkan transport paraseluler

# Efek peptida terhadap nilai TEER lapis tunggal sel-sel MDCK

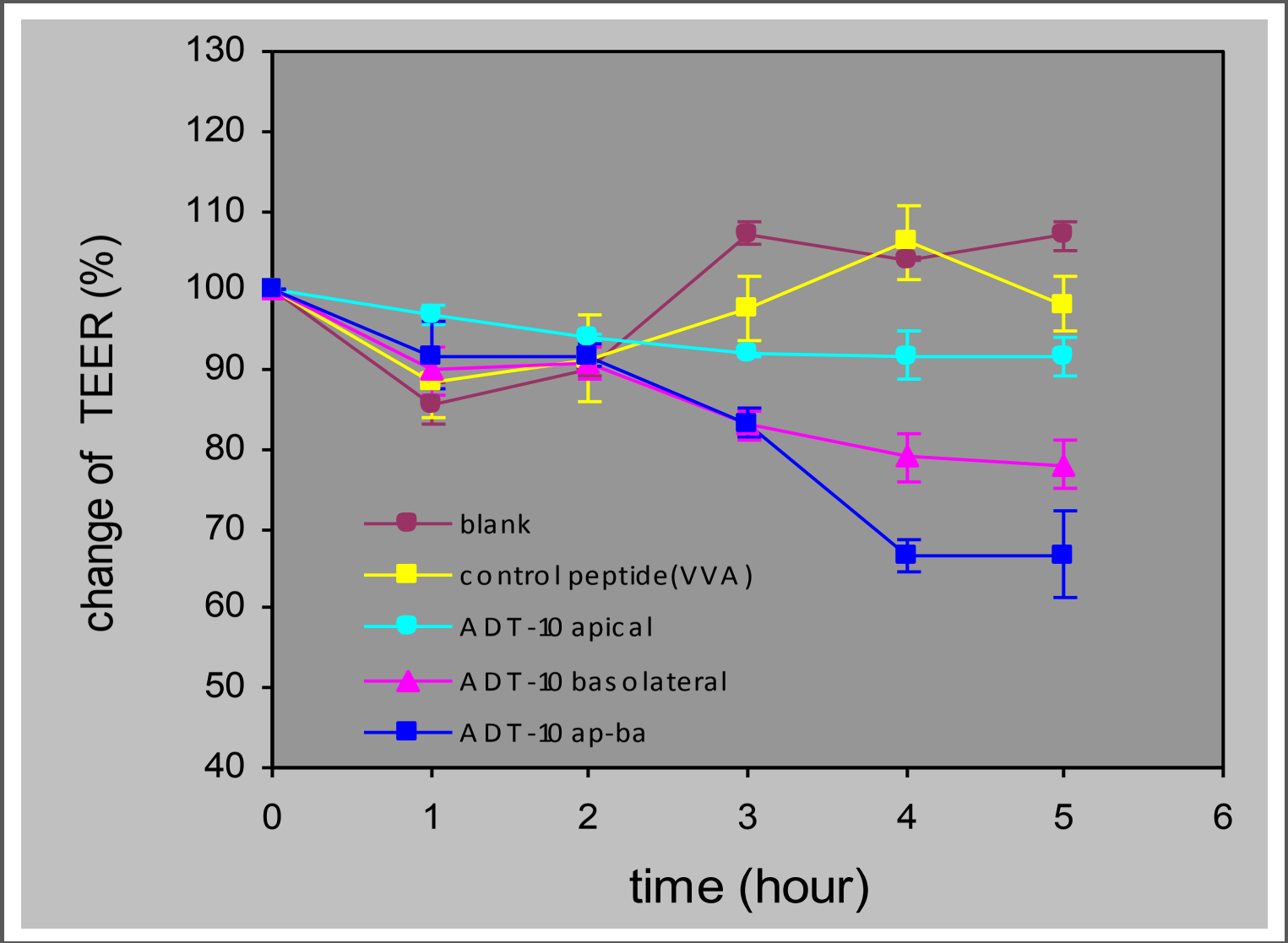


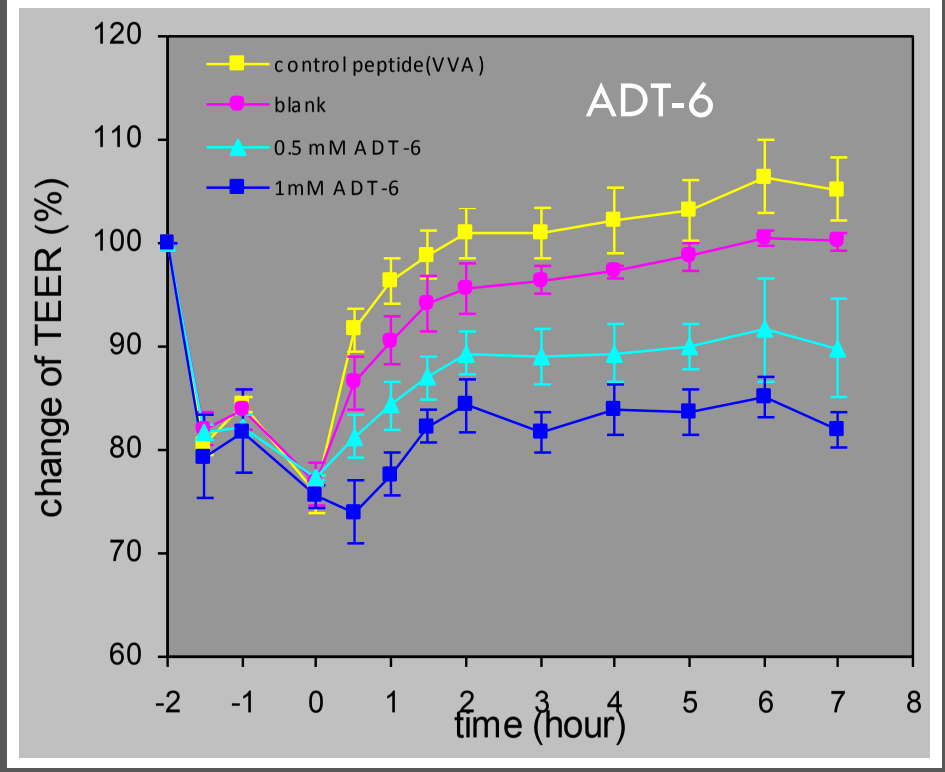



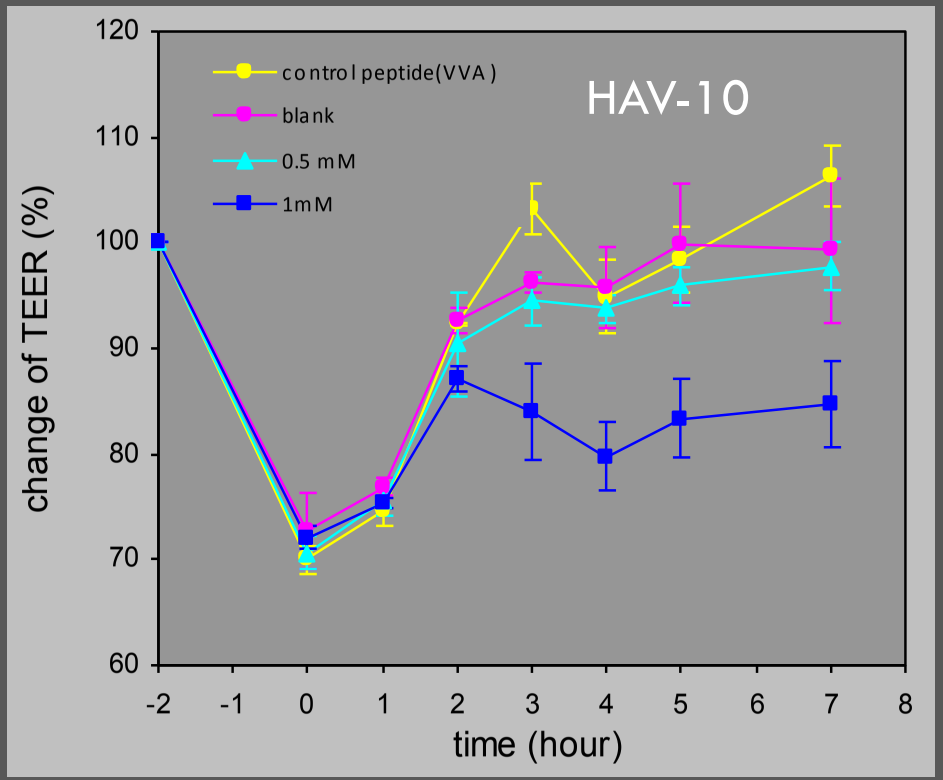



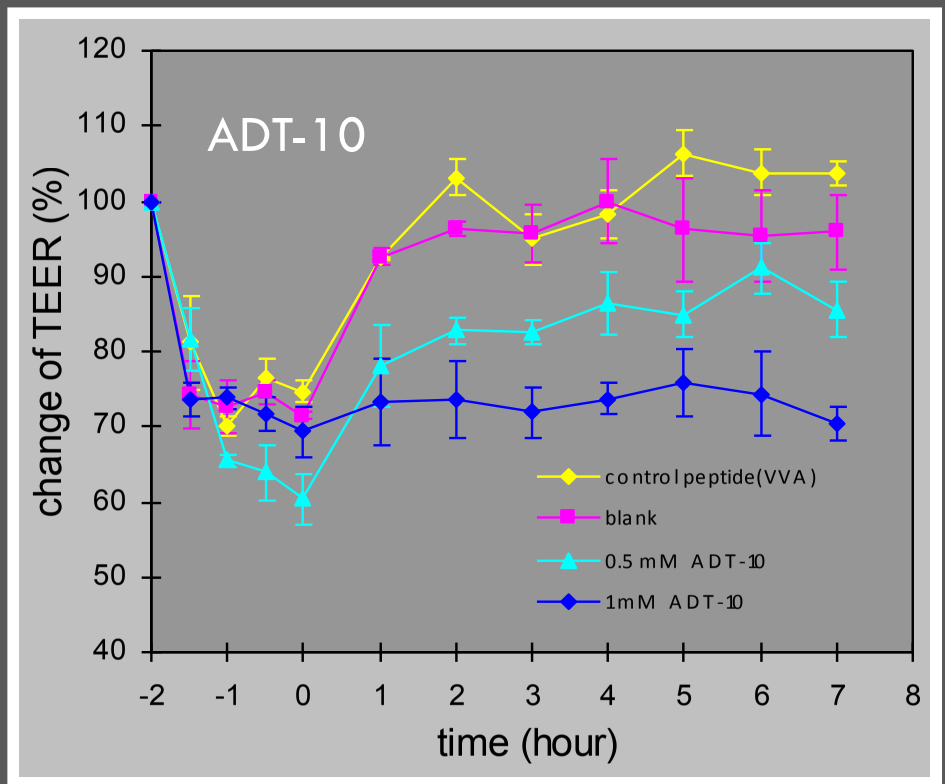
# Efek Peptida ADT-10 terhadap nilai TEER lapis tunggal sel-sel MDCK



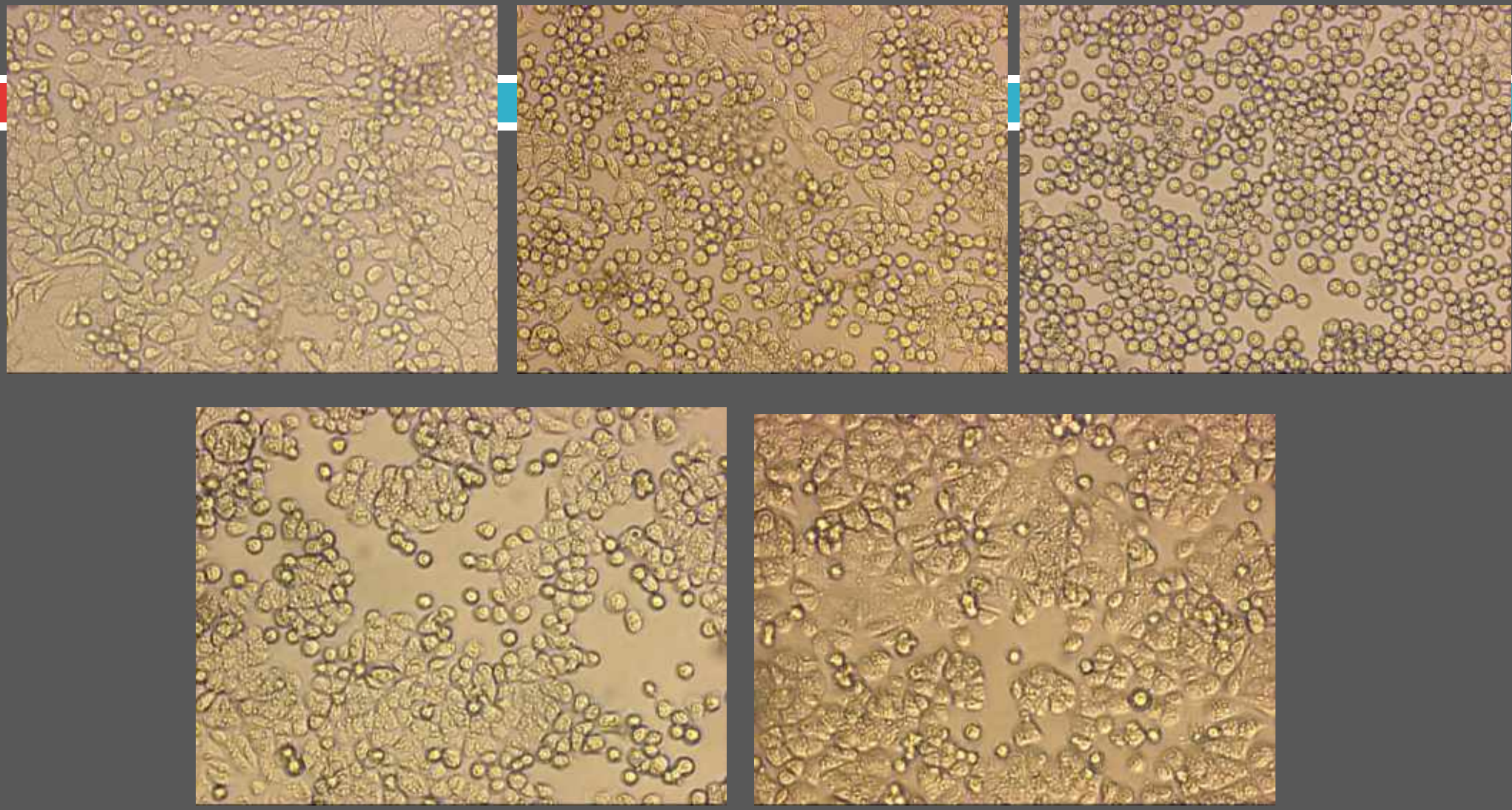
# Efek peptides terhadap nilai TEER lapis tunggal sel-sel CACO-2



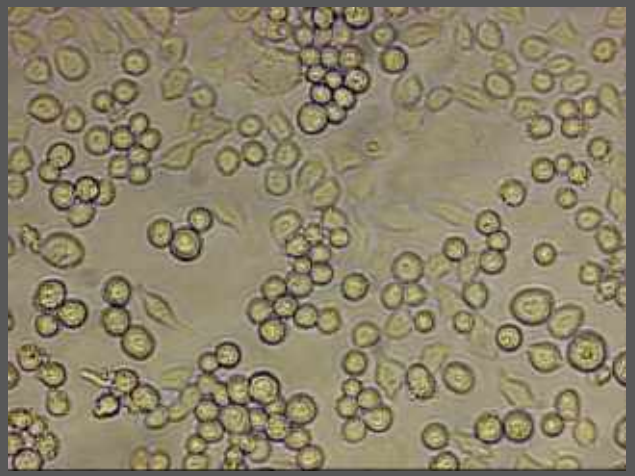
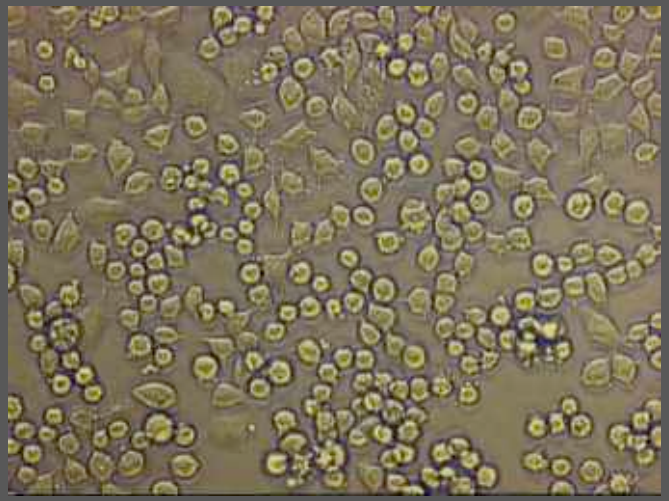
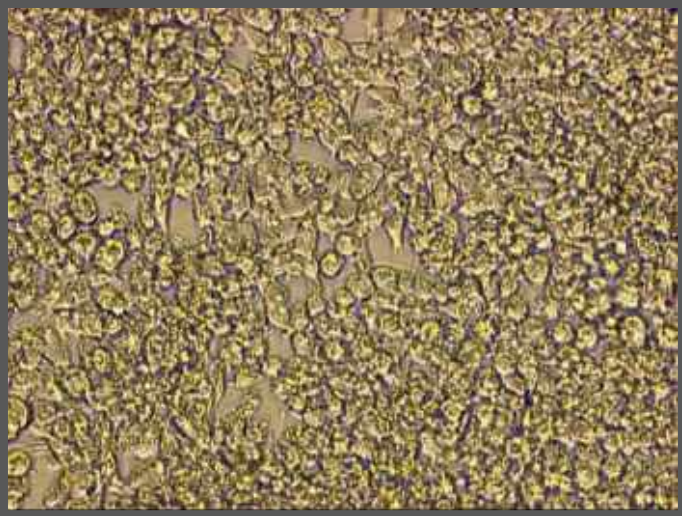
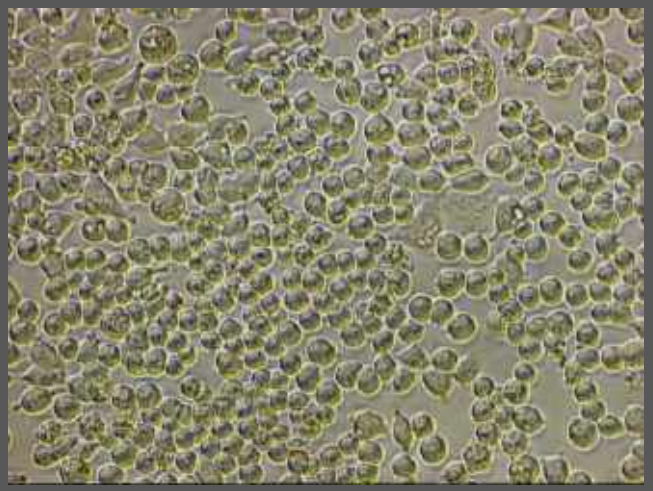
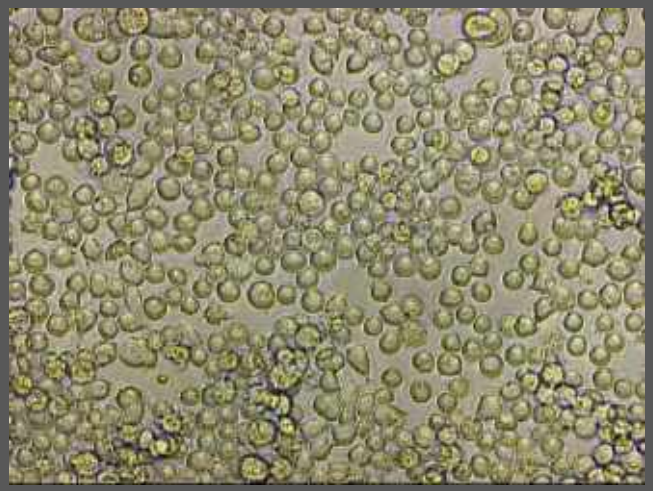
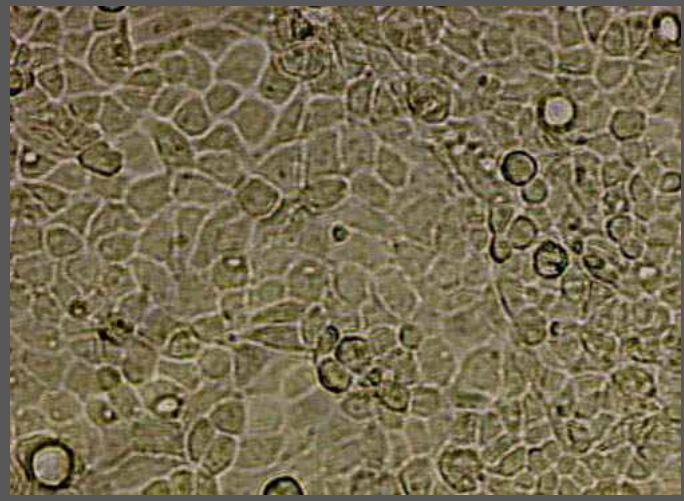



# •MDCK Cell Monolayer

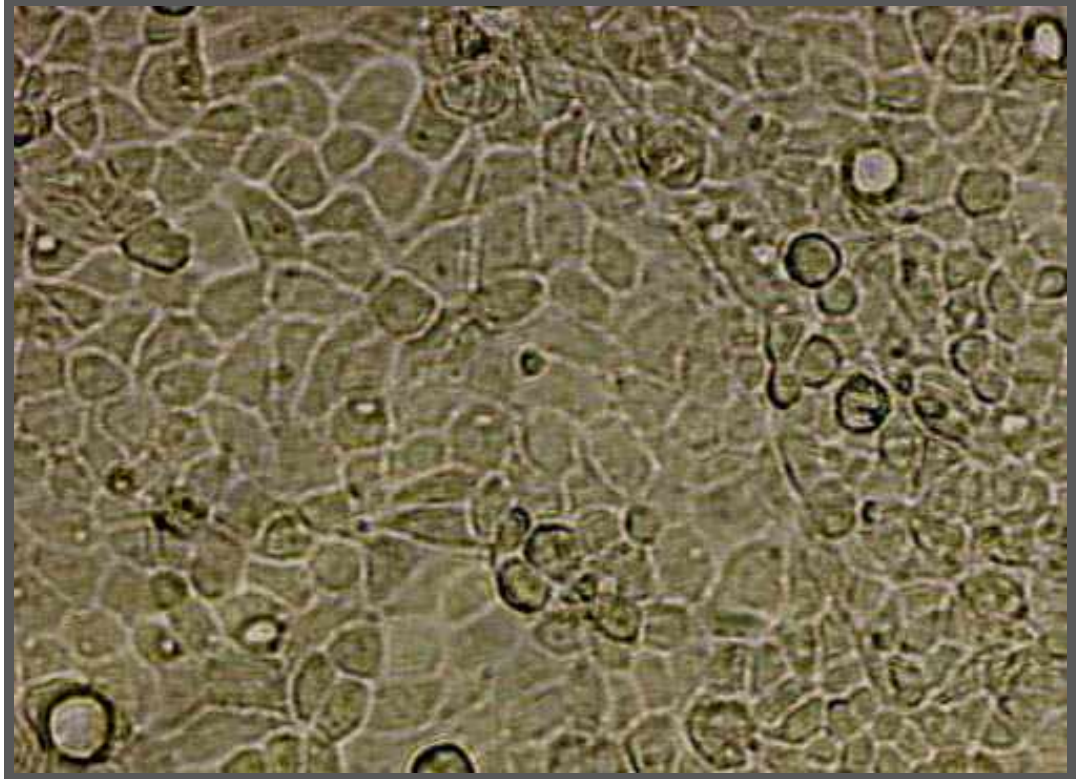


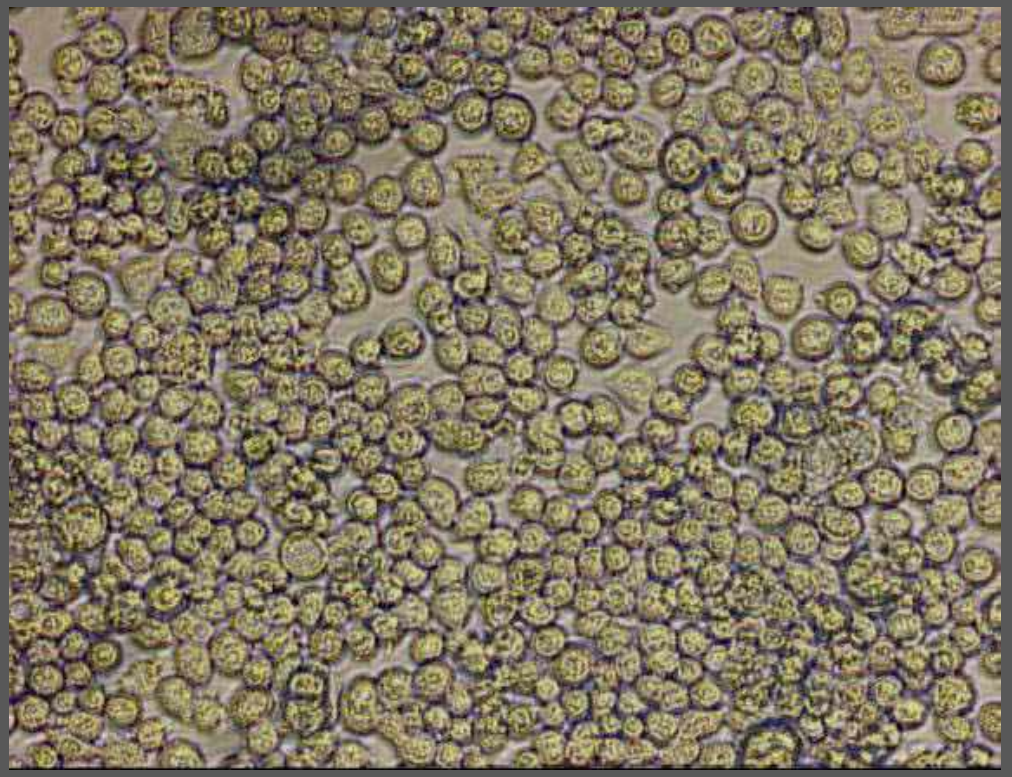
- deaggregation in Ca2+ free medium at 60, 75 and 150 min of incubation
- Reaggregation in  2 mM Ca2+ / 0,74 mM Mg2+ media after 30 and 60 min

# CACO-2 cells deaggregation & reaggregation

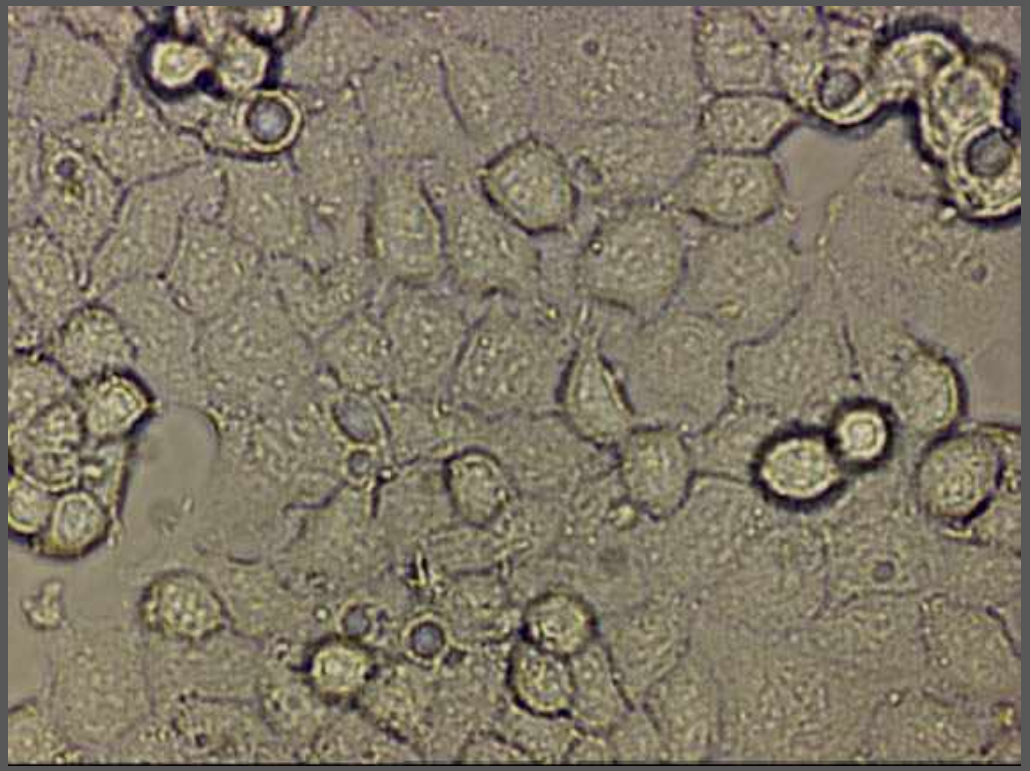
# Efek peptida ADT-10 terhadap daya reagregasi sel CACO-2

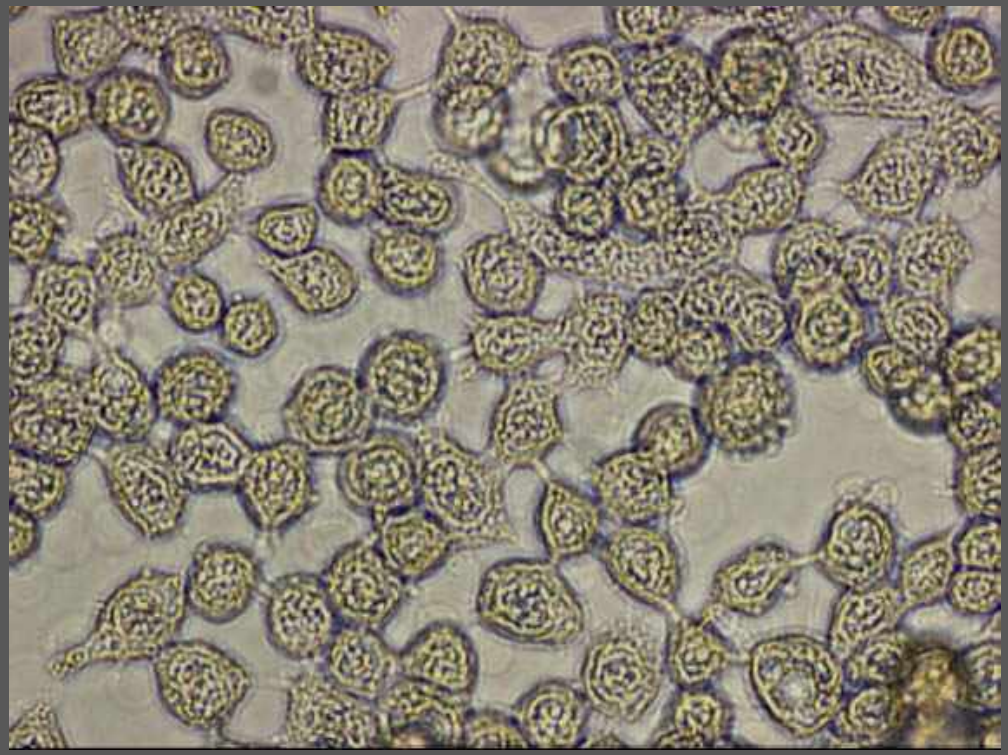




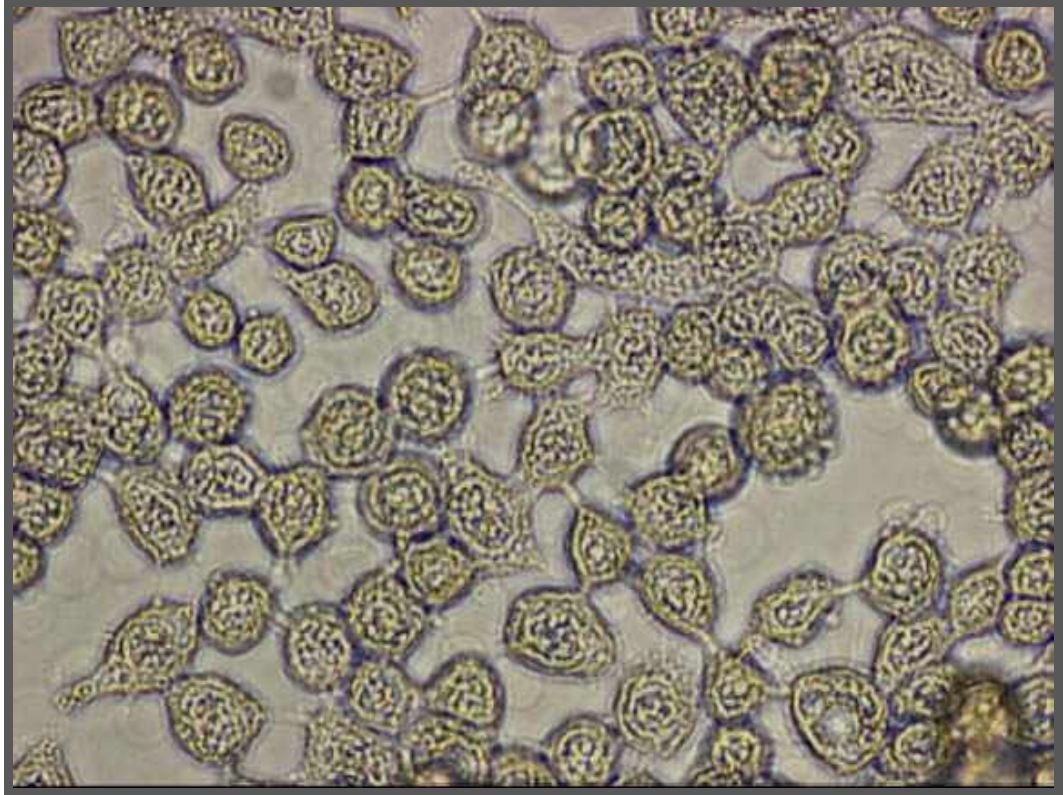
Reagregasi sel CACO-2 dalam media yang mengandung (ka) atau tanpa peptida ADT-10 (ki) setelah 1 jam pemulihan (Direkam dengan perbesaran 100x)

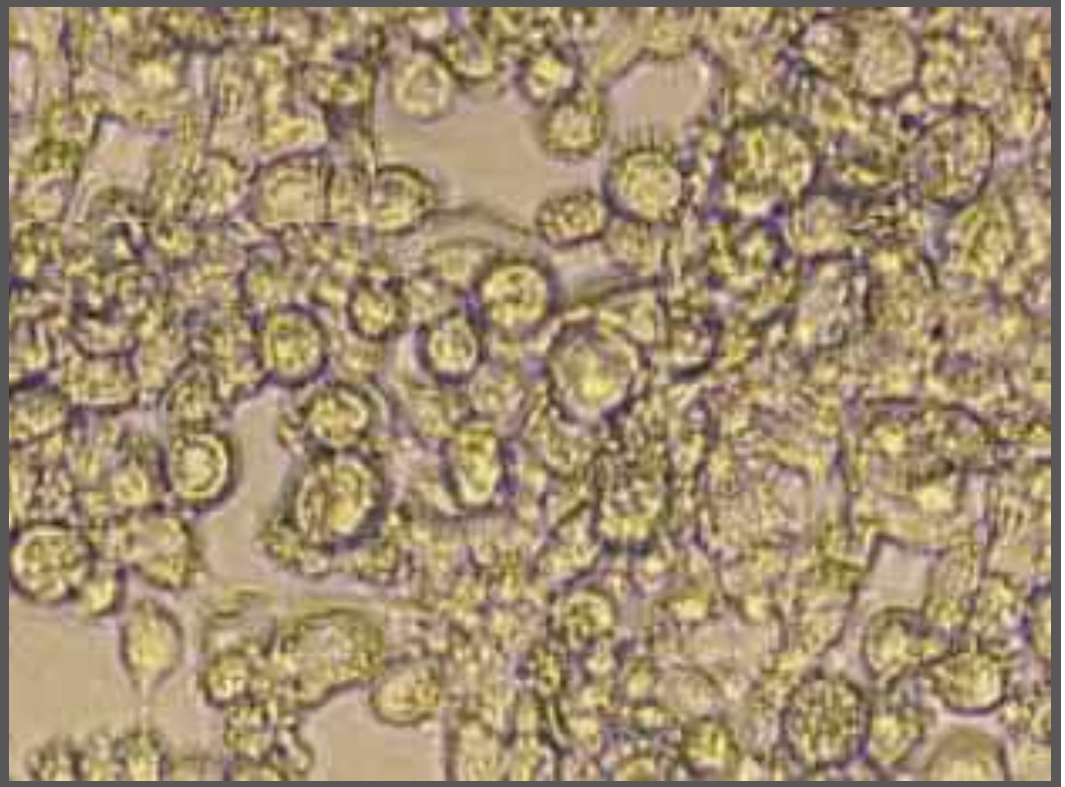
# Efek peptida HAV-10 terhadap daya reagregasi sel CACO-2



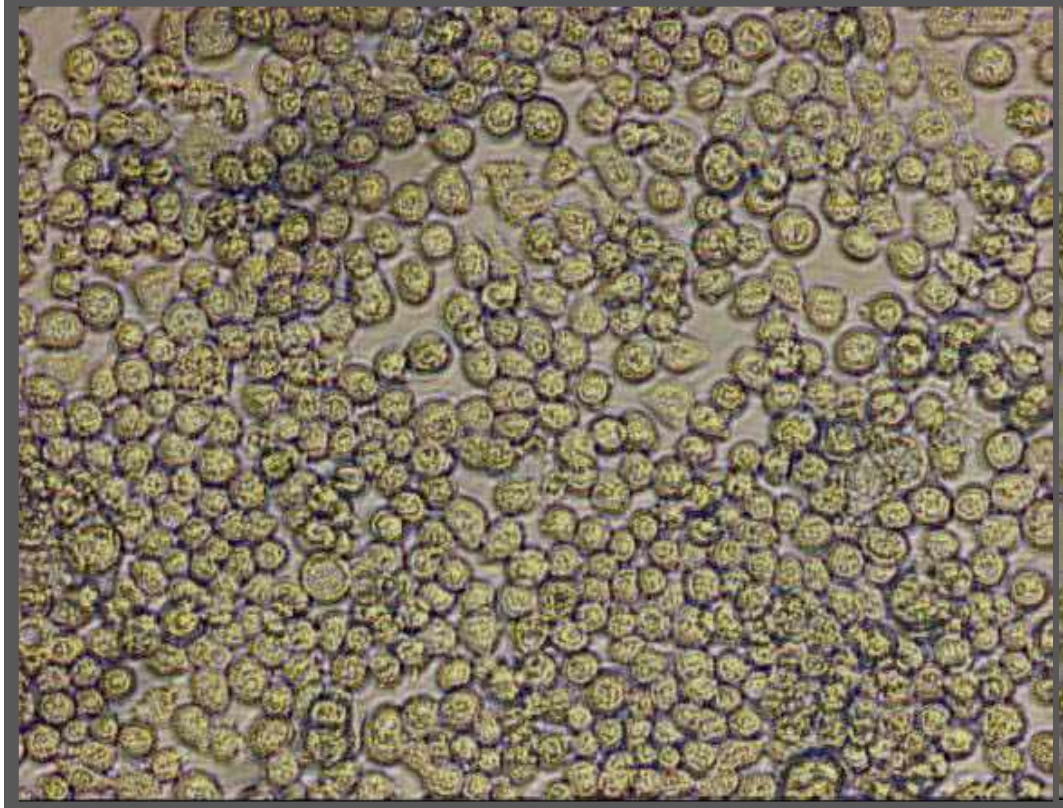


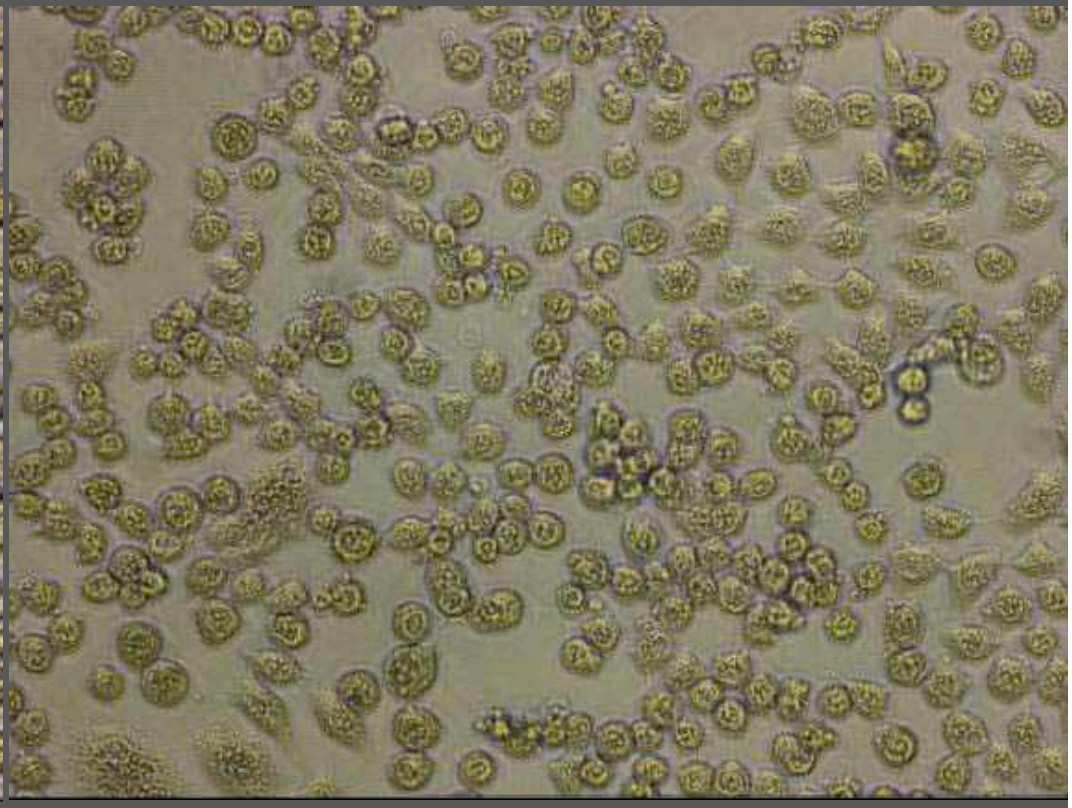
Reagregasi sel CACO-2 dalam media yang mengandung (ka) atau tanpa peptida HAV-10 (ki) setelah 1 jam pemulihan (Direkam dengan perbesaran 400x)





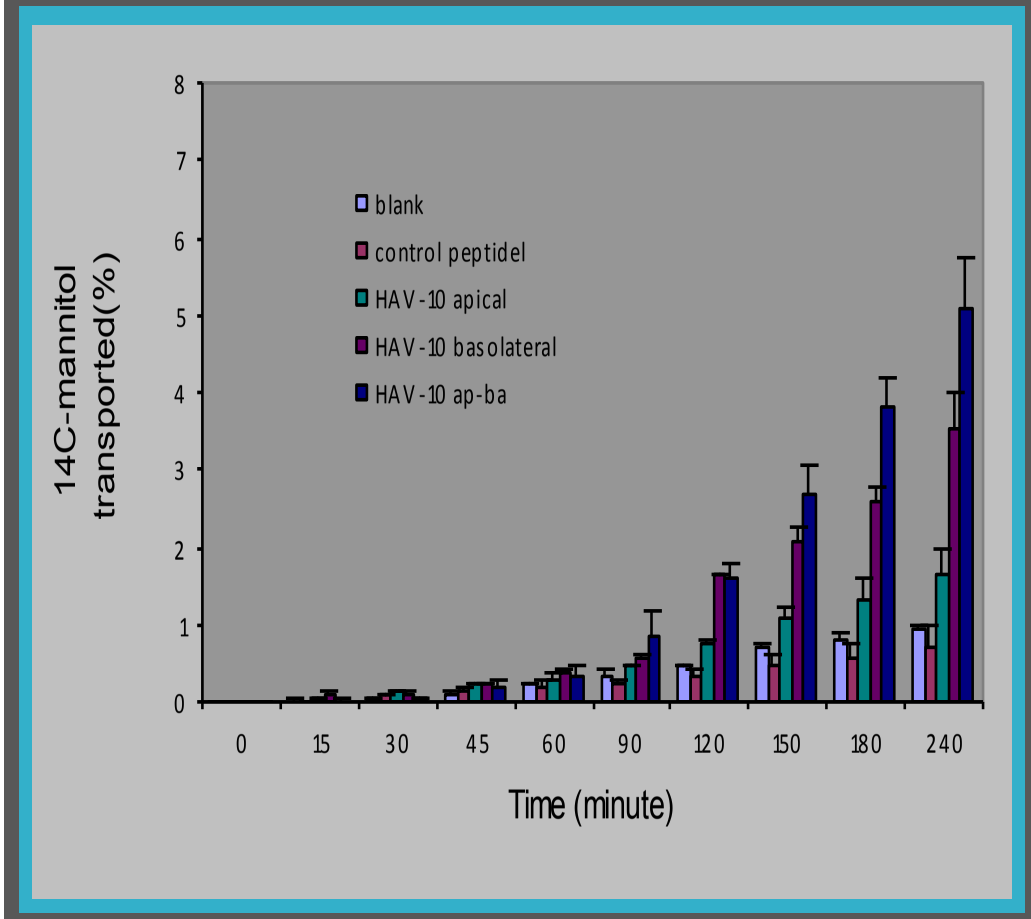
Setelah 5 jam reagregasi mulai berlangsung walaupun ada peptida HAV-10 (perbesaran 400x)

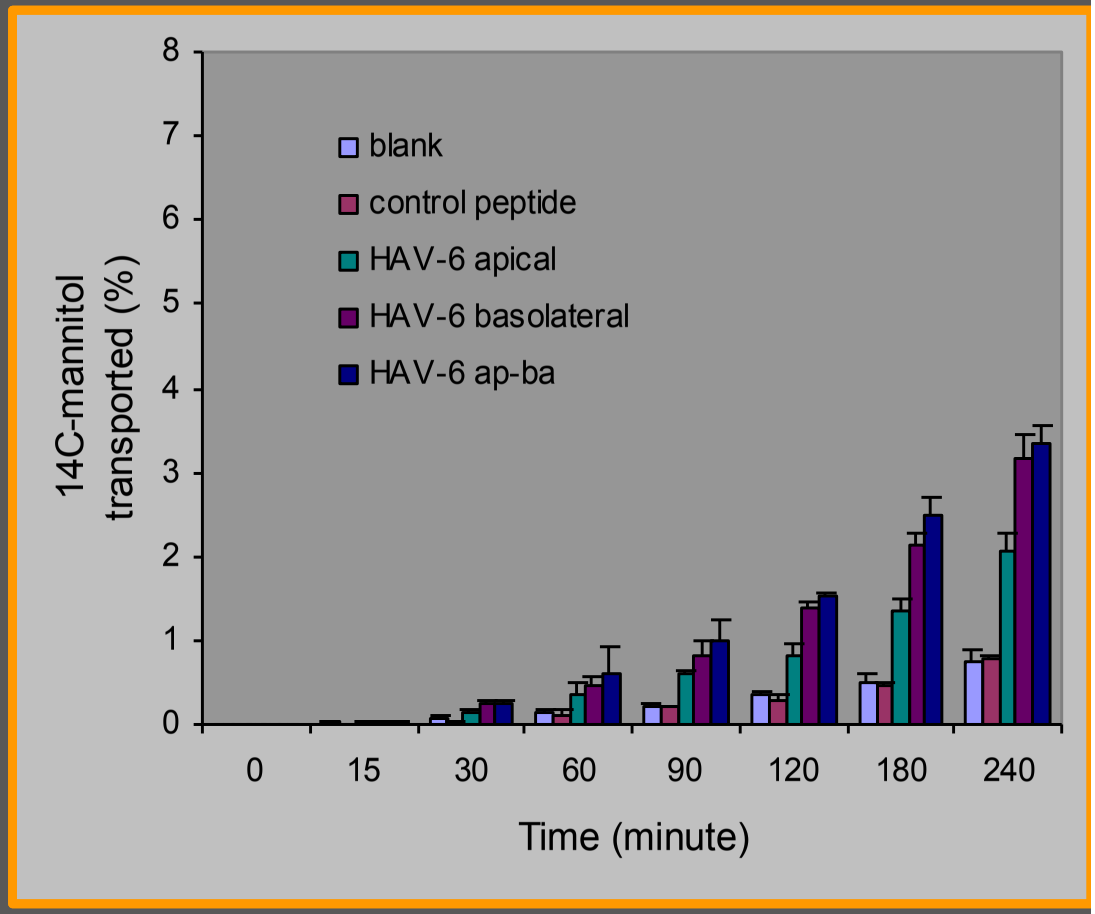




Setelah 5 jam reagregasi mulai berlangsung walaupun ada peptida ADT-10 (perbesaran 100x)

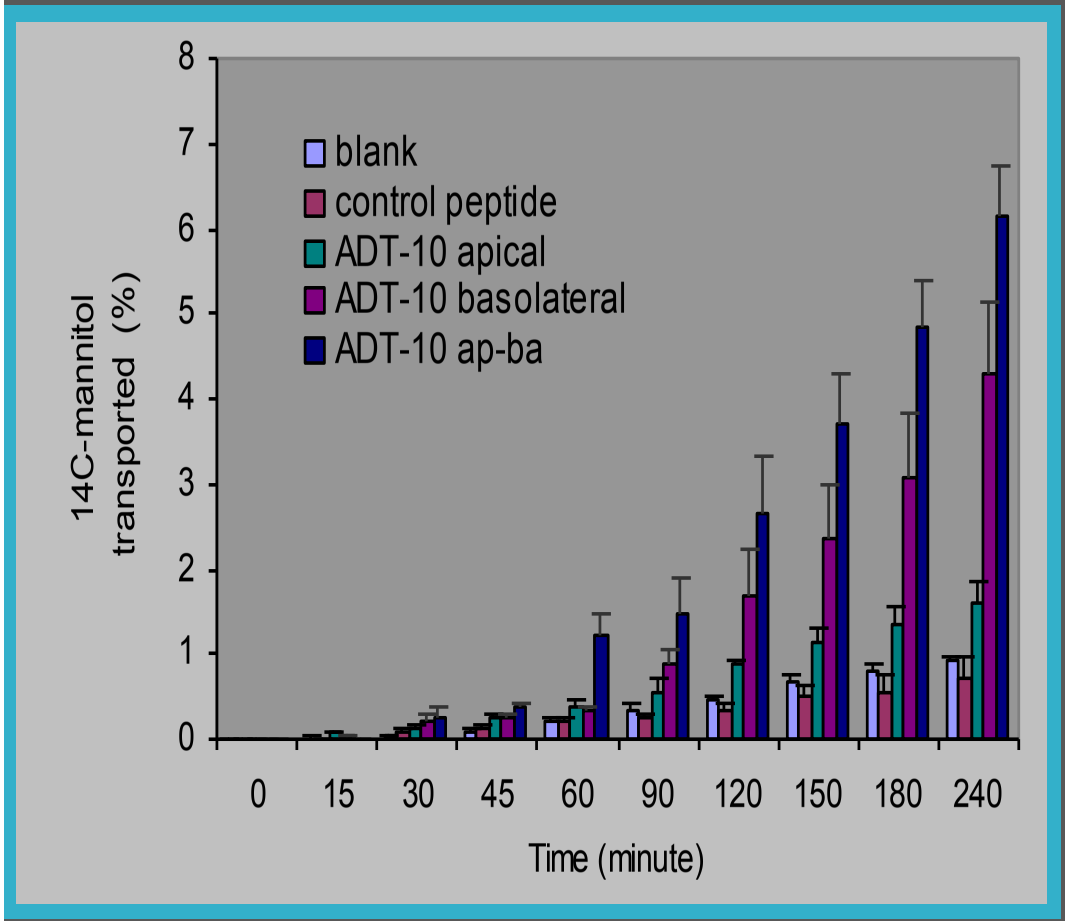
# Aktivitas peptida HAV dalam meningkatkan transpor paraseluler

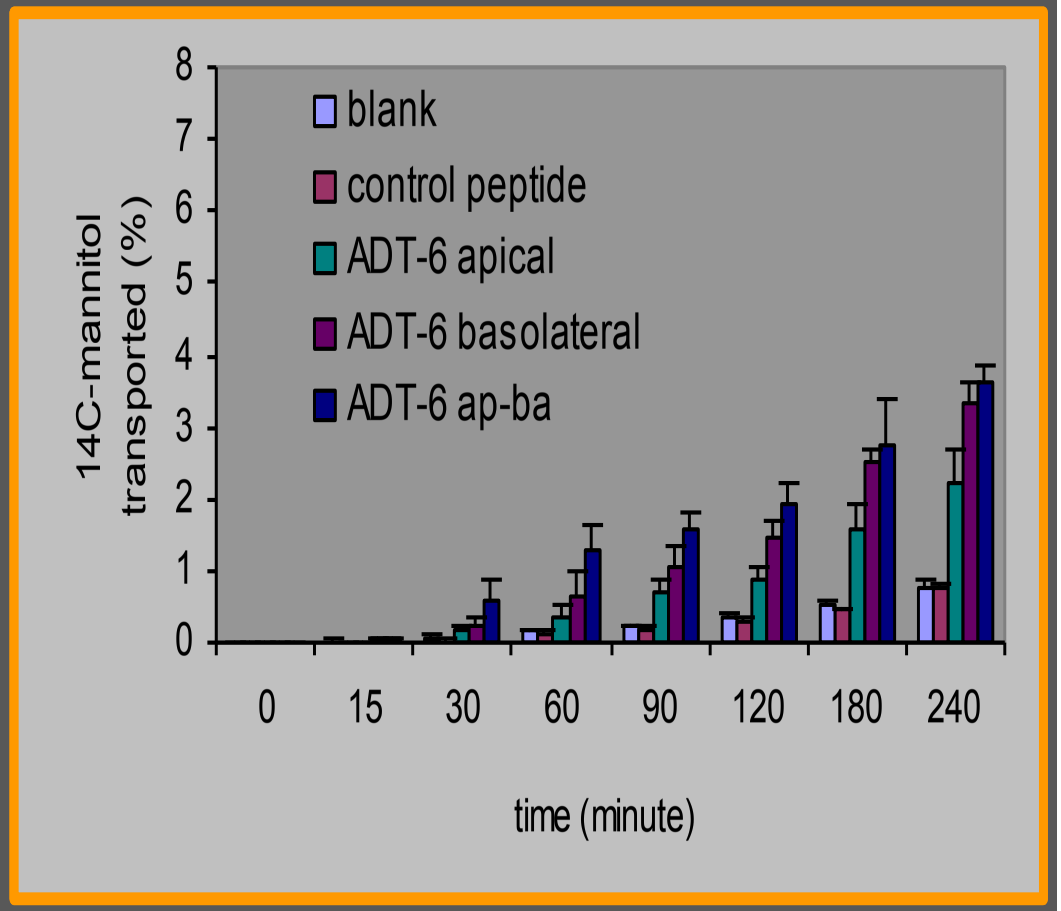



Transpor paraseluler $^{14}$C-manitol melintasi lapis tunggal sel MDCK

# Aktivitas peptida ADT dalam meningkatkan transpor paraseluler



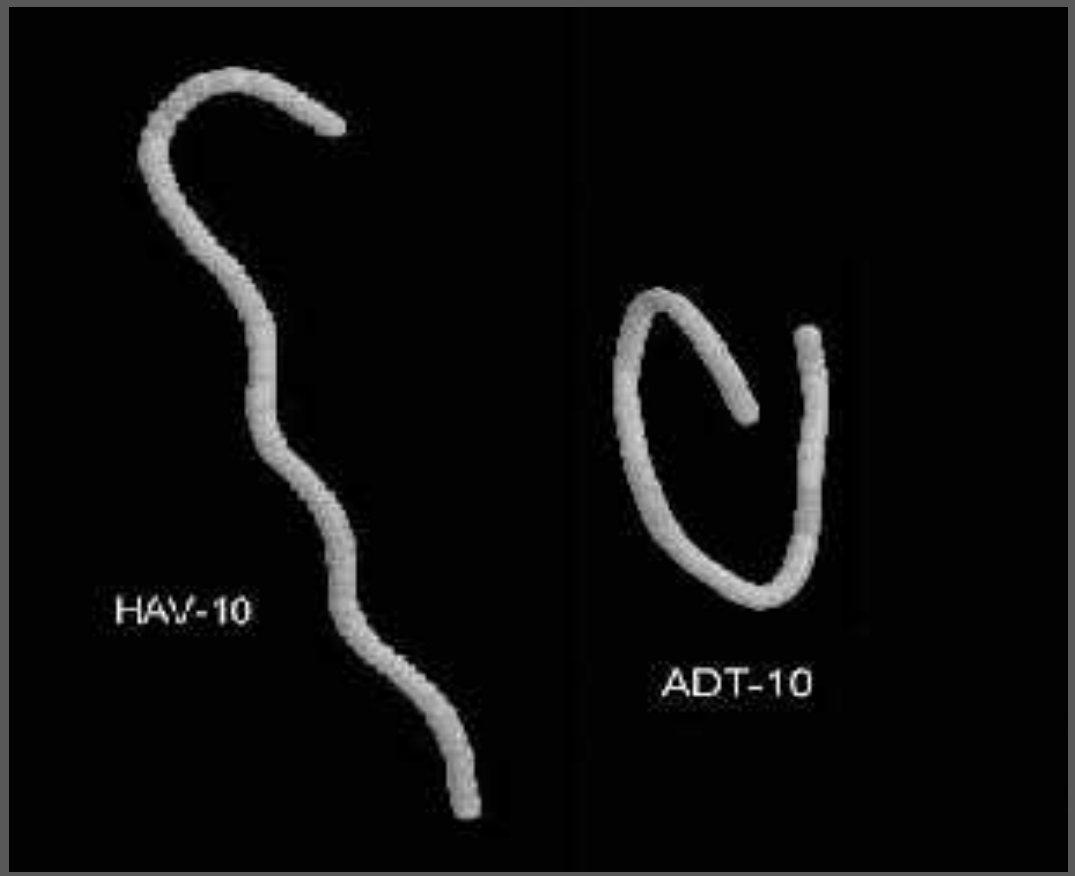




Transpor paraseluler $^{14}$C-manitol melintasi lapis tunggal sel MDCK

# Aktivitas Peptida Kadherin dalam meningkatkan transpor paraseluler

| Peptida | Koefisien Permeabilitas (cm/det. X 1000000) | | |
|---|---|---|---|
| | apikal | basolateral | ap-ba |
| Kontrol | 0.35 ± 0.04 | 0.35 ± 0.04 | 0.35 ± 0.04 |
| Blanko | 0.37 ± 0.05 | 0.37 ± 0.05 | 0.37 ± 0.05 |
| HAV-10 | 0.61 ± 0.12 | 1.31 ± 0.13 | 1.88 ± 0.23 |
| HAV-6 | 1.01 ± 0.11 | 1.59 ± 0.07 | 1.71 ± 0.12 |
| ADT-10 | 0.60 ± 0.08 | 1.57 ± 0.34 | 2.31 ± 0.24 |
| ADT-6 | 1.12 ± 0.26 | 1.70 ± 0.14 | 1.80 ± 0.17 |

# Peptides Conformation



# Ringkasan Hasil

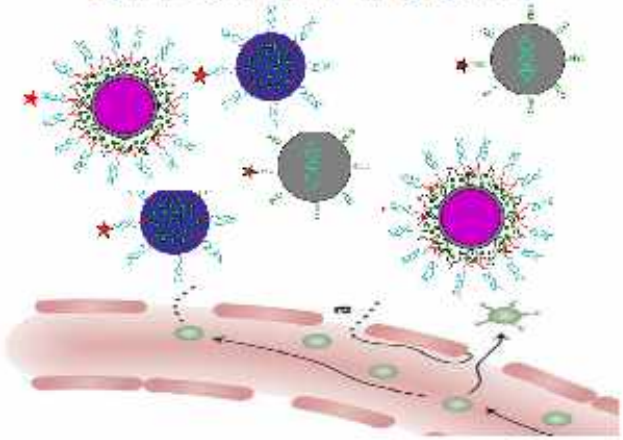
- Keempat peptida (HAV-10, HAV-6, ADT-10, dan ADT-6) memiliki aktivitas yang cukup tinggi dalam memodulasi *junction* antar sel:
  - Dapat menurunkan nilai TEER sel MDCK sampai 75% dari nilai TEER semula dalam waktu 3 jam
  - Dapat menghambat reagregasi sel-sel CACO-2 secara sementara (reversibel)dalam waktu maksimal 5 jam
  - Dapat meningkatkan permeasi/transport paraseluler sampai

# Kesimpulan

- Beberapa peptida sintetik, yaitu LFSHAVSSNG, QGADTPPVGV, SHAVSS dan ADTPPV, telah terbukti dapat memodulasi junction antar sel dan meningkatkan permeasi paraseluler.
- Hasil penelitian ini membuka jalan untuk mengembangkan suatu metode baru dalam penghantaran obat, yaitu dengan memodulasi junction antar sel menggunakan peptida sintetik.
- Untuk itu diperlukan penelitian lebih lanjut dan mendalam agar metode/teknik penghantaran obat ini dapat digunakan secara klinis.

# THANK YOU

# Strategi Meningkatkan Penghantaran Obat Di Dalam Tubuh



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