

**TRANSPORT  
TRANSMEMBRANAR ACTIV**

# CARACTERISTICI

- ❑ PROCES ENDERGONIC ( $\Delta G > 0$ )
- ❑ ÎN SENSUL CREȘTERII GRADIENTULUI
- ❑ REALIZAT DE PROTEINE MEMBRANARE
- ❑ VIZIUNEA CLASICĂ:  
SITUS DE LEGARE MOBIL
- ❑ VIZIUNEA ACTUALĂ:  
MODIFICĂRI CONFORMAȚIONALE

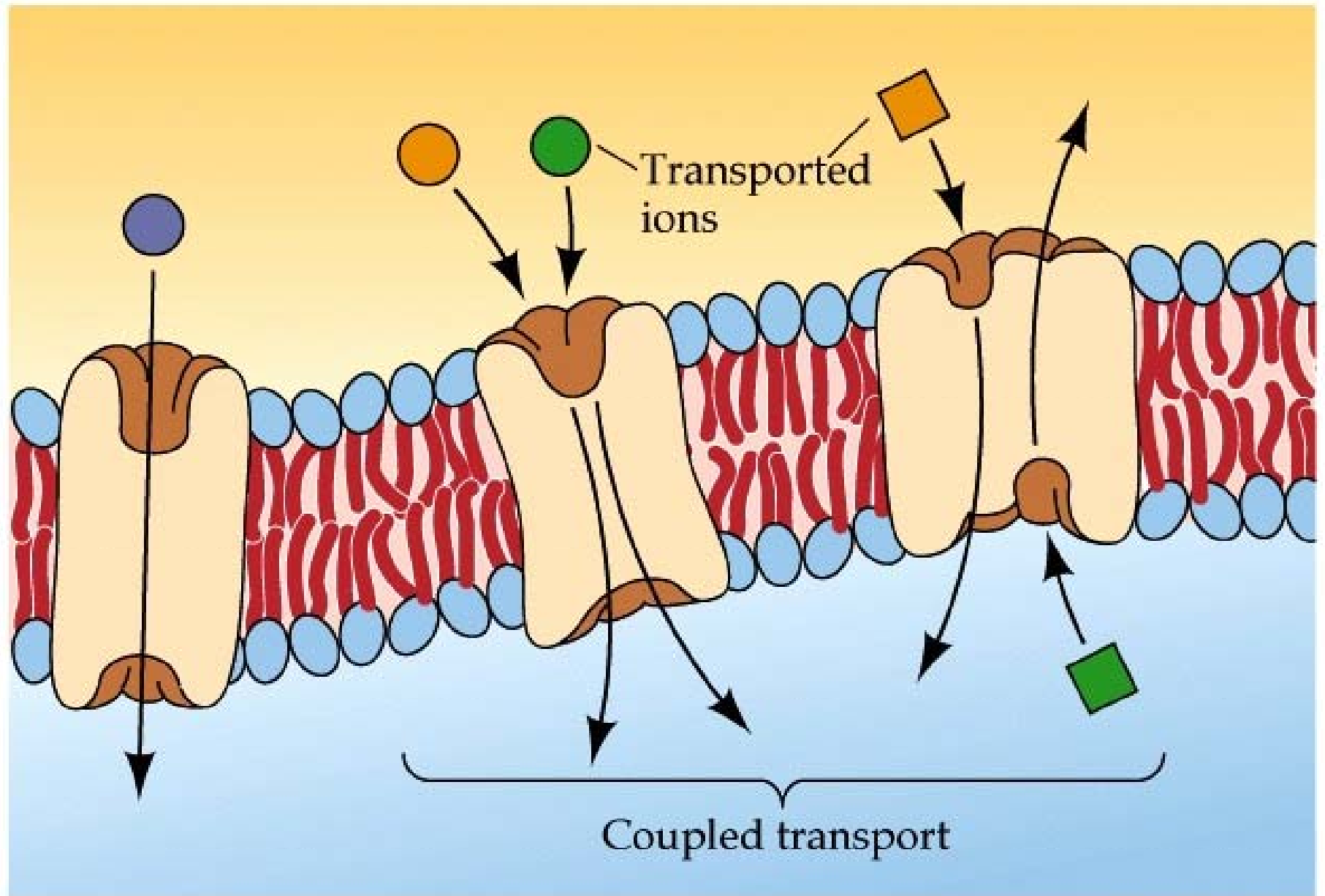
# CLASIFICARE DUPĂ SURSA DE ENERGIE

– **PRIMAR:** ENERGIA PROVINE DE LA O REACȚIE EXERGONICĂ SAU DE LA LUMINĂ

– **SECUNDAR:** ENERGIA PROVINE DE LA TRANSPORTUL PASIV AL UNEI ALTE SPECII CHIMICE

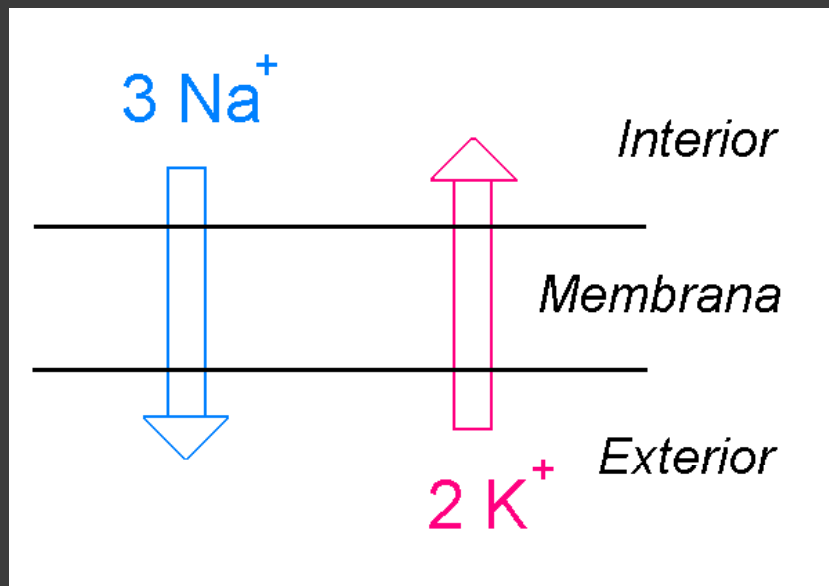
➤ **SIMPORT (COTRANSPORT)**

➤ **ANTIORT (CONTRATRANSPORT)**



IONI TRANSPORTAȚI	LOCALIZARE
TIP $E_1E_2$	
$H^+$ $H^+ / K^+$ $Na^+ / K^+$ $Ca^{2+}$ $Ca^{2+}$	Membrana plasmatică (plante, animale) Mucoasa gastrică (animale) Membrana plasmatică (animale) Membrana sarcoplasmatică (animale) Membrana plasmatică (animale)
TIP $F_0F_1$	
$H^+$ $H^+$ $H^+$	Membrana unor celule bacteriene Membrana internă a mitocondriilor (animale) Membrana cloroplastelor (plante)
TIP VACUOLAR	
$H^+$ $H^+$ $H^+$	Membrana lizozomală (animale) Granule secretoare (eucariote) Granule de stocare (eucariote)

## POMPA DE $\text{Na}^+/\text{K}^+$



*10 December 1997 will always stand as one of the brightest days in the annals of the University of Aarhus – the day in which the first researcher from this University was awarded a Nobel Prize.*



Skou, J. C. The influence of some cations on an adenosine triphosphatase from peripheral nerves. *Biochim. Biophys. Acta* 1000, 439–446 (1957).

# ROLURI ALE POMPEI DE $\text{Na}^+/\text{K}^+$

DĂ NAȘTERE ȘI MENTȚINE GRADIENTȚI DE POTENȚIAL ELECTROCHIMIC AI IONILOR  $\text{Na}^+$  ȘI  $\text{K}^+$  ÎN CELULE ANIMALE

Excitabilitate  
electrică

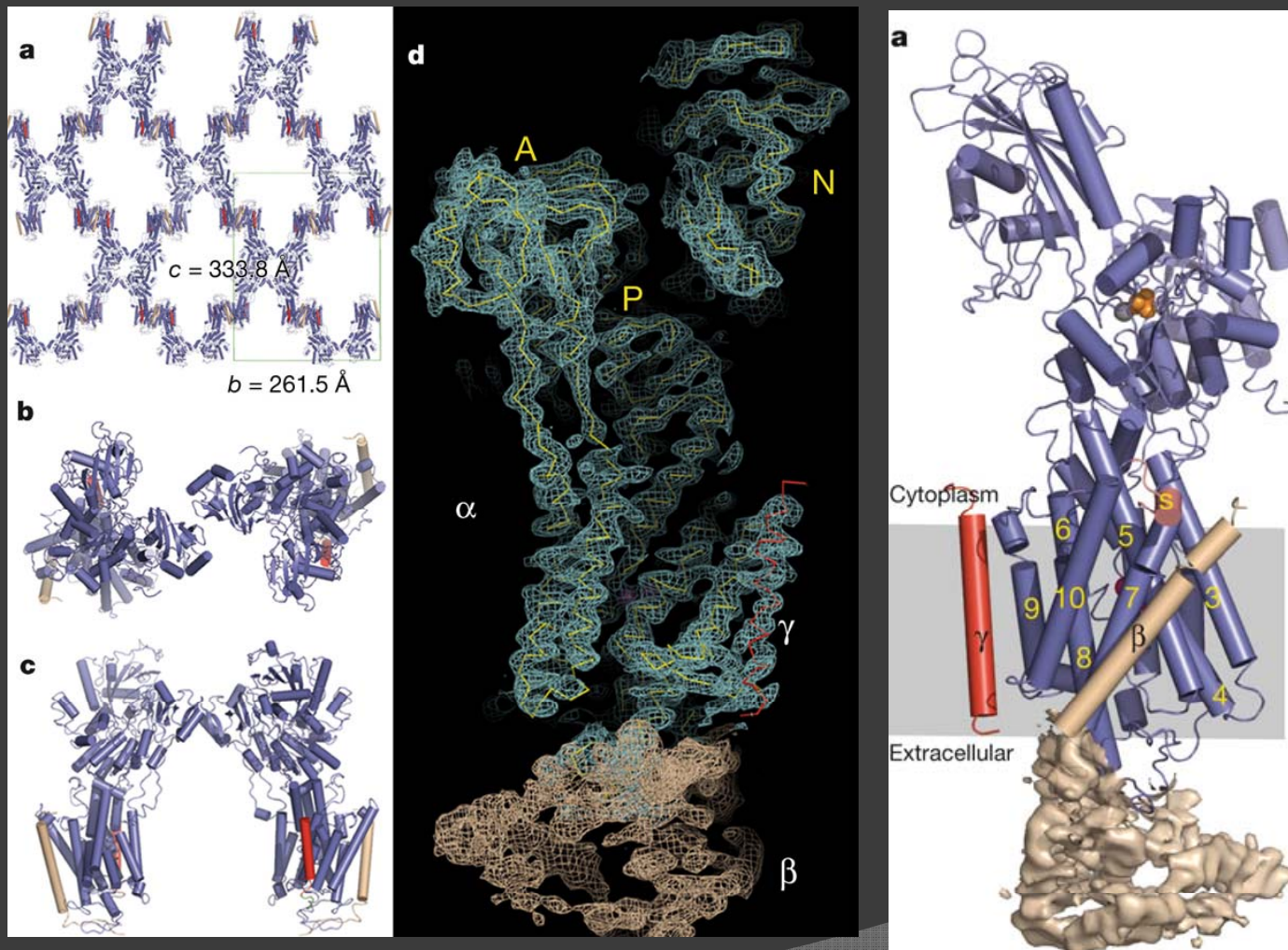
Reglează pH-ul  
intracelular

Transport activ secundar  
(uptake of ions, amino acids,  
nutrients, neurotransmitters )

Reglează volumul  
celular. (Apa  
urmează sărurile.)



# STRUCTURA MOLECULARĂ A POMPEI DE $\text{Na}^+/\text{K}^+$



P. Morth *et al.* *Nature* **450**, 1043-1049 (2007); doi:10.1038/nature06419

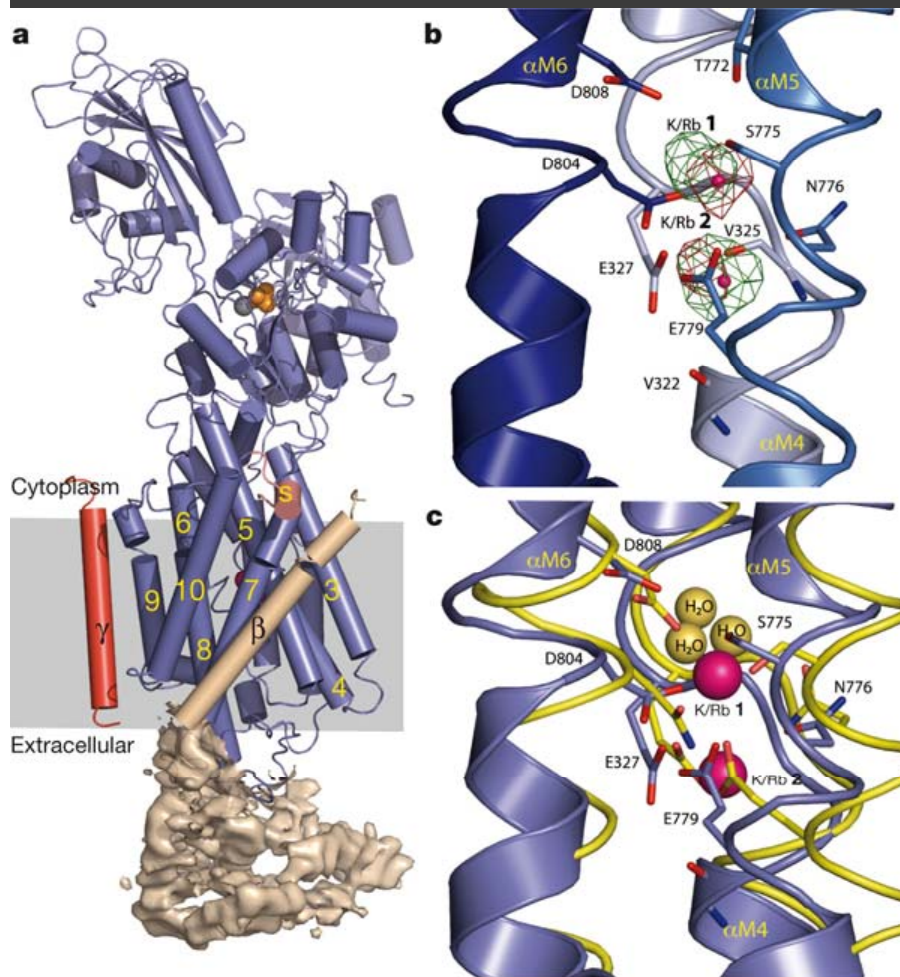
nature



# STRUCTURA MOLECULARĂ A POMPEI DE $\text{Na}^+/\text{K}^+$

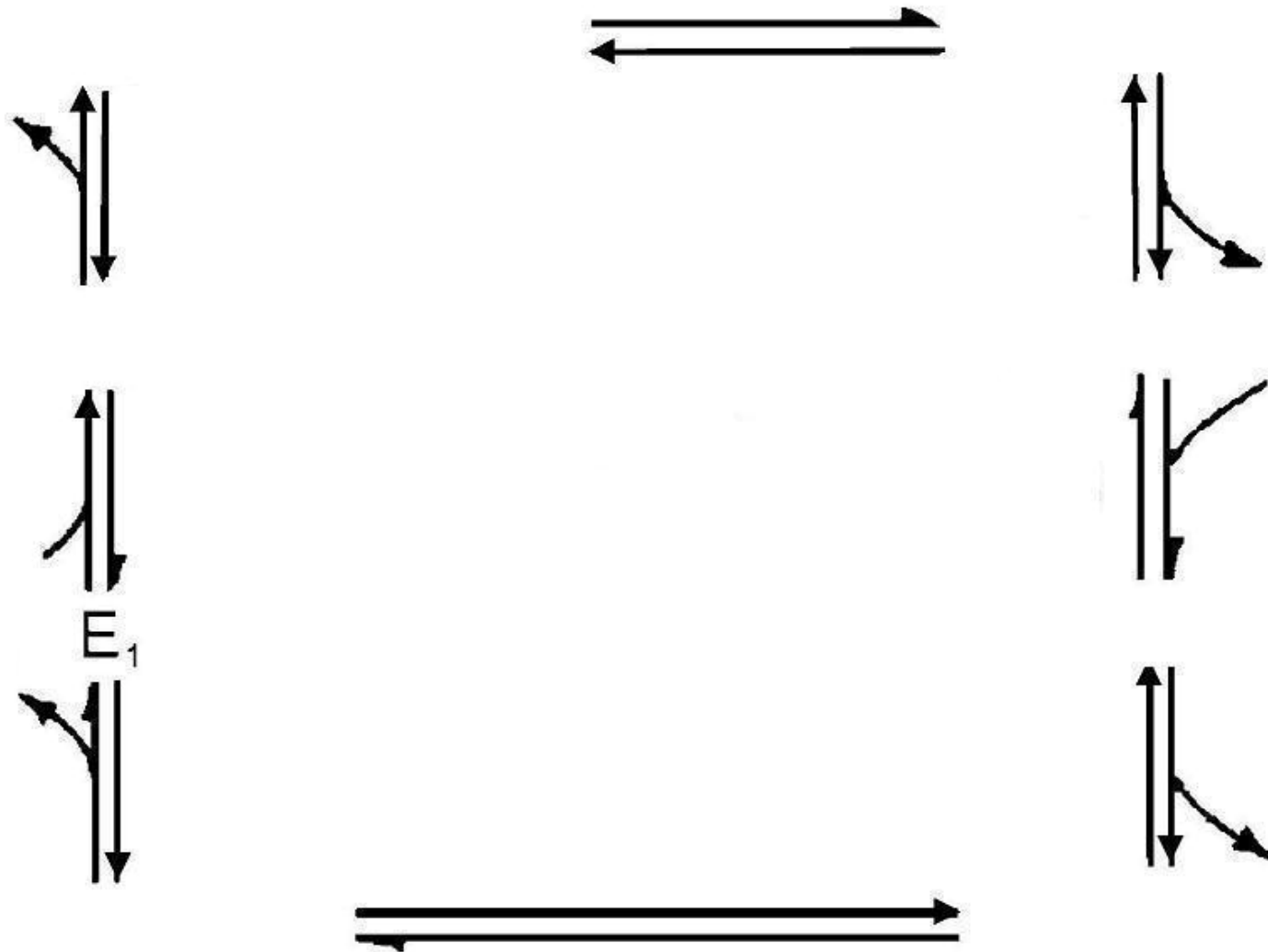
M=245-290 kDa

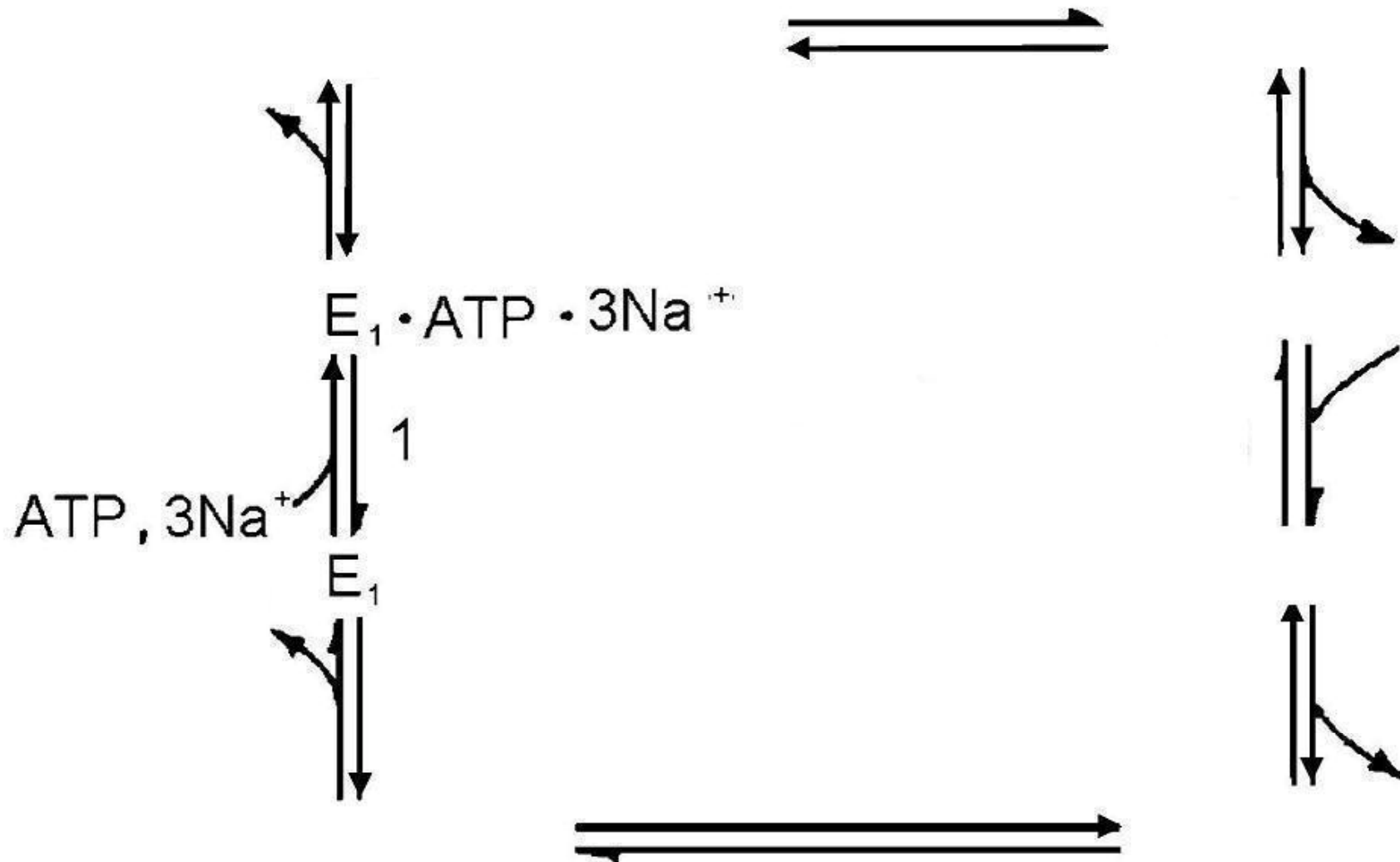
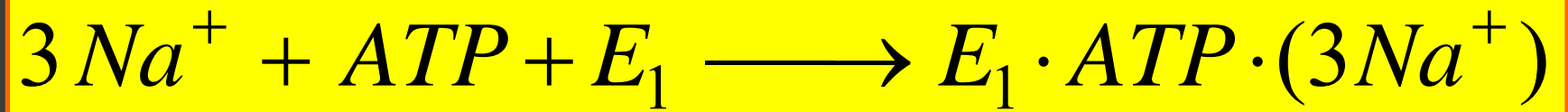
➤ STRUCTURĂ MOLECULARĂ: heterodimer ( $\alpha$   $\beta$ )

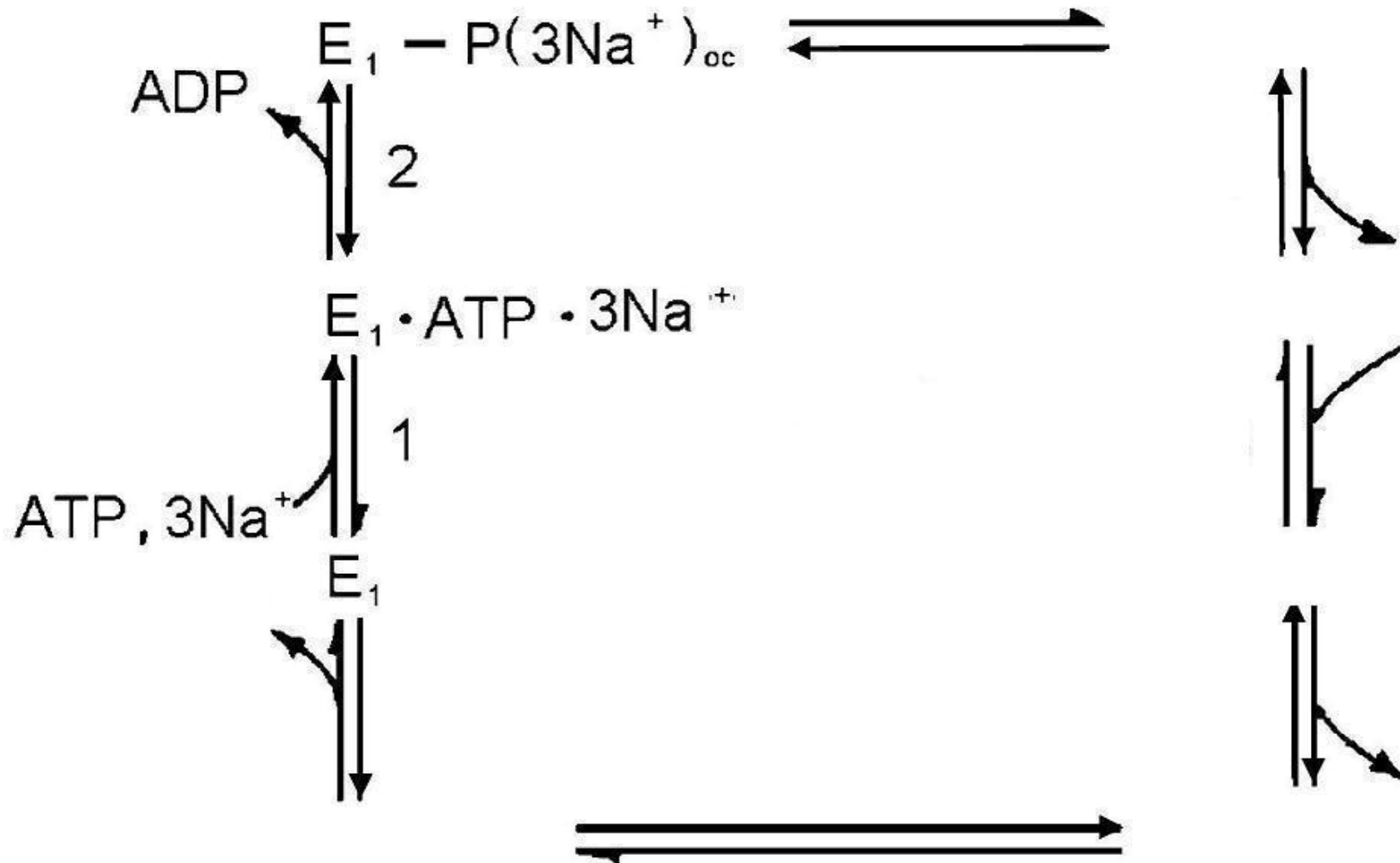
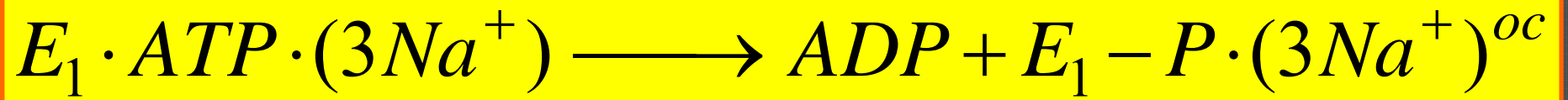


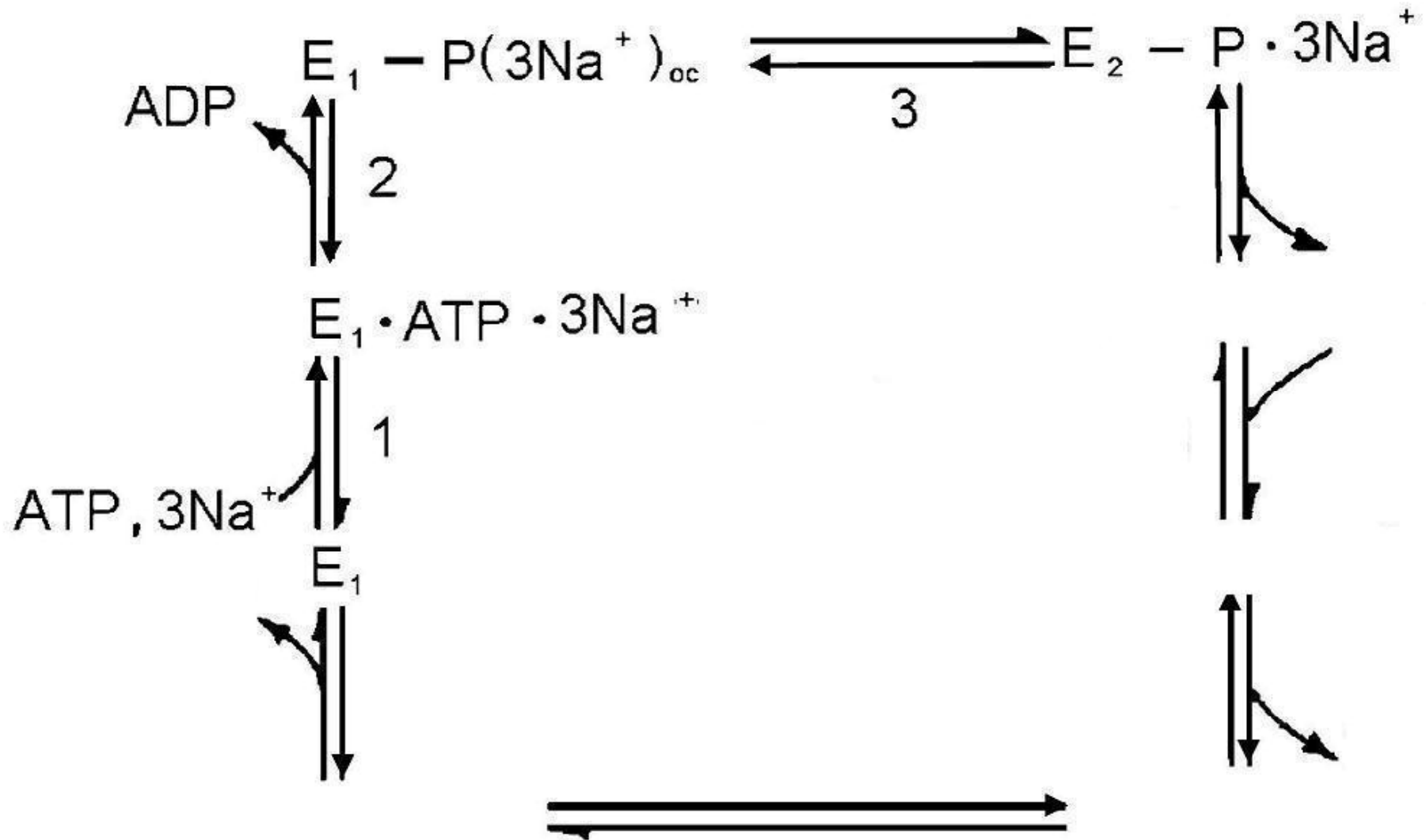
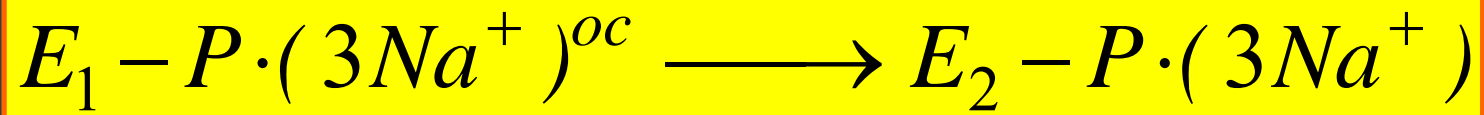
- $\alpha$ : 1020 amino acizi  
10 elici  $\alpha$  + bucle  
transport; activitate enzimatică
- $\beta$ : 1 elice  $\alpha$  + 1 domeniu extracelular  
legare de  $\text{K}^+$  ; stabilizare
- FXYP ( $\gamma$  în rinichi): 1 elice  $\alpha$   
➤ Subunitatea  $\alpha$  - analogă  
 $\text{Ca}^{2+}$ -ATP-azei din reticulul  
sarcoplasmatic (SERCA)

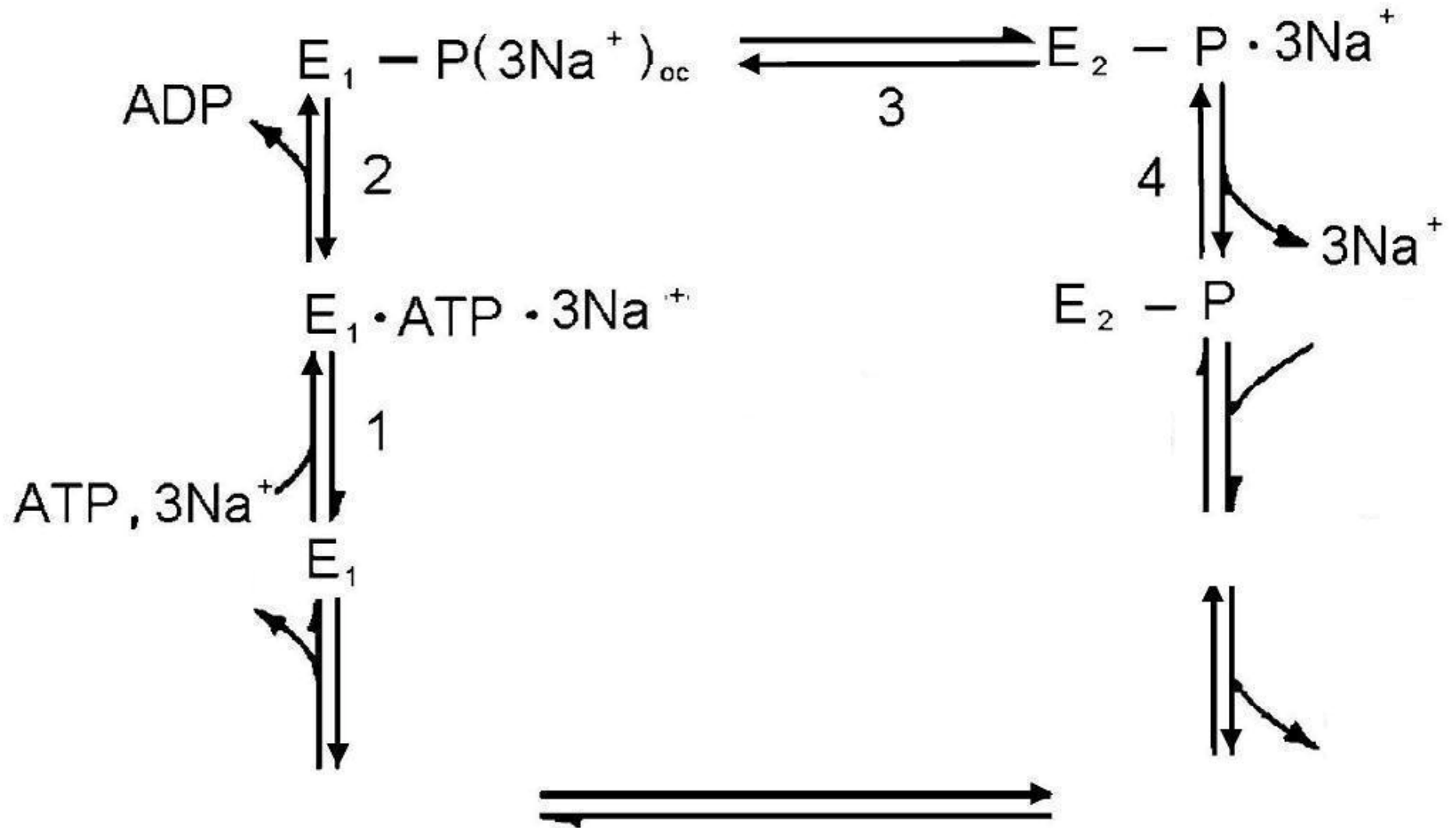
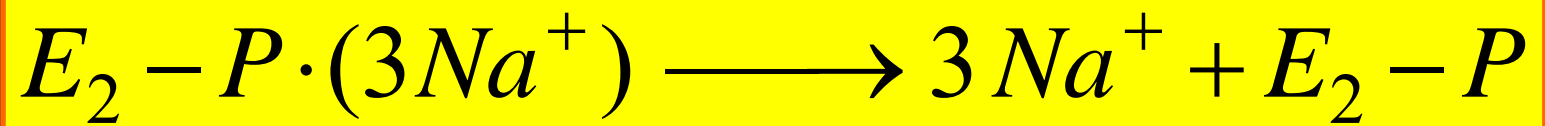
# FUNCȚIONAREA $\text{Na}^+/\text{K}^+$ – ATPazei (SCHEMA POST-ALBERS)



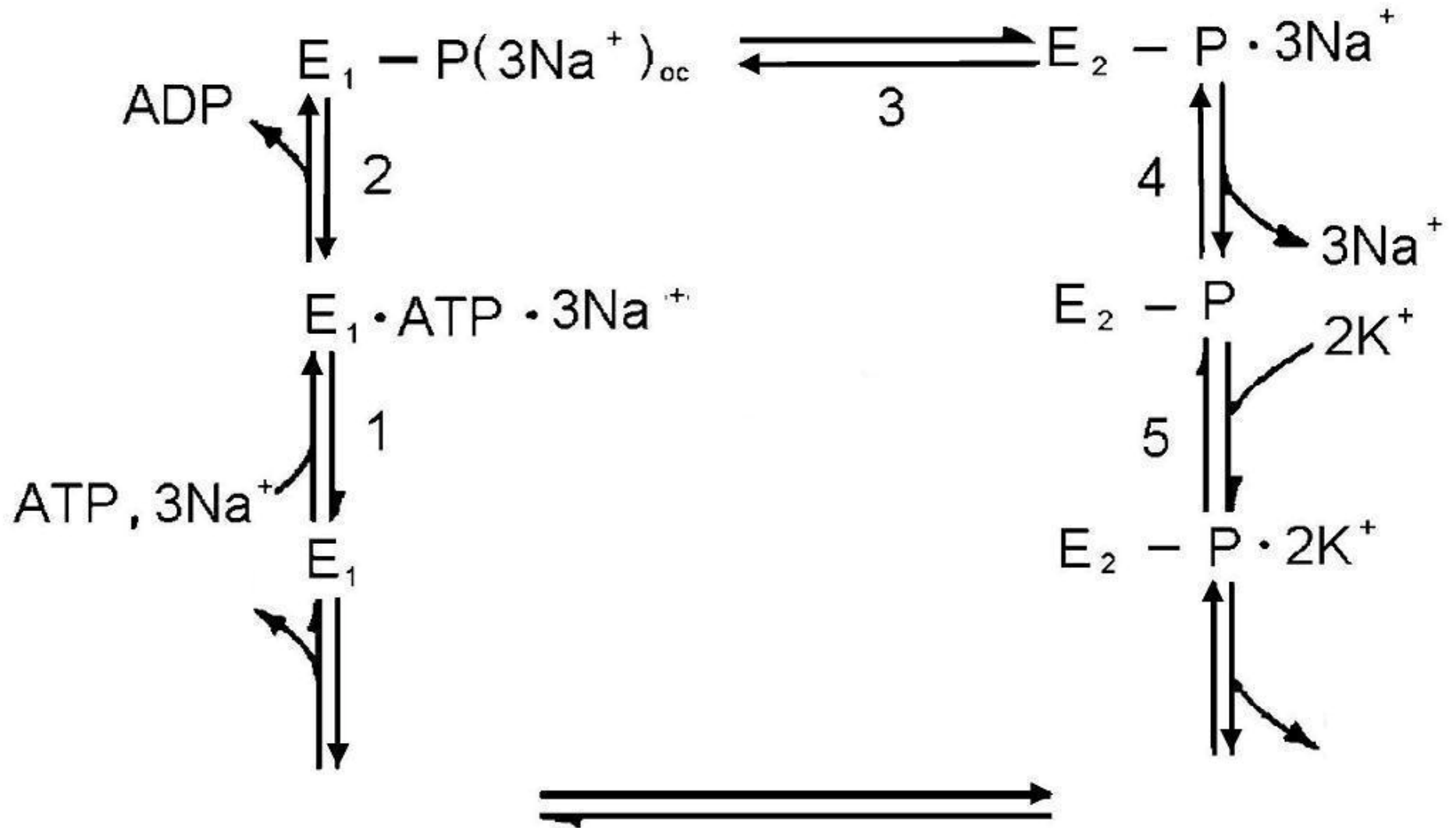
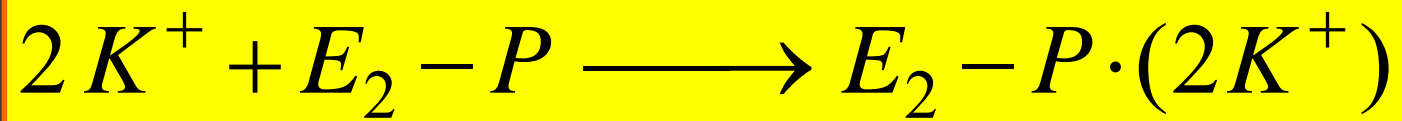


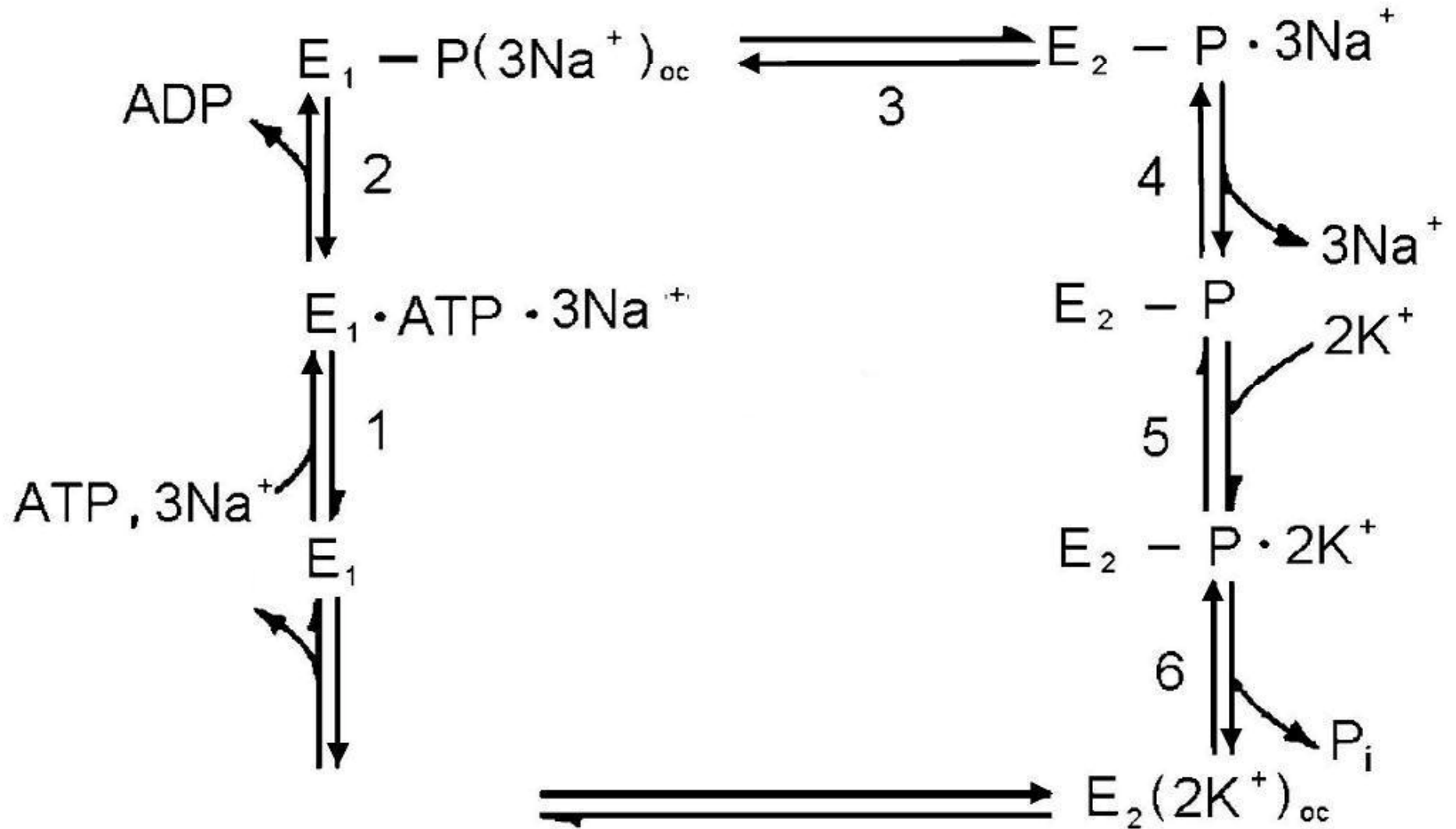
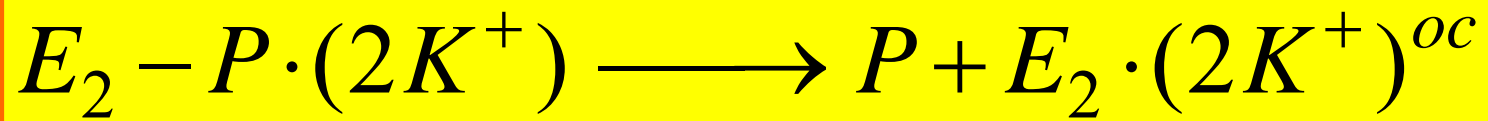


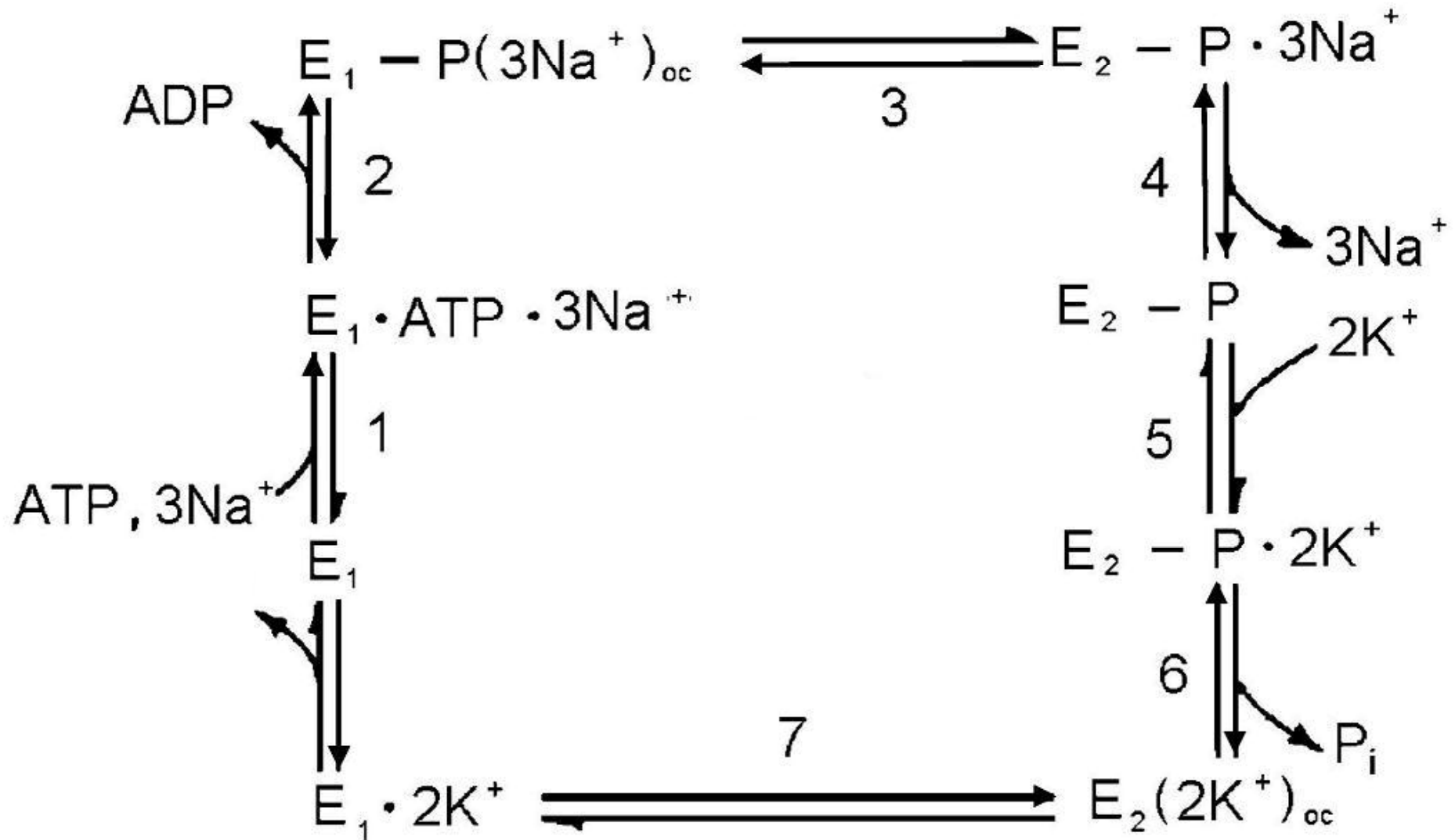
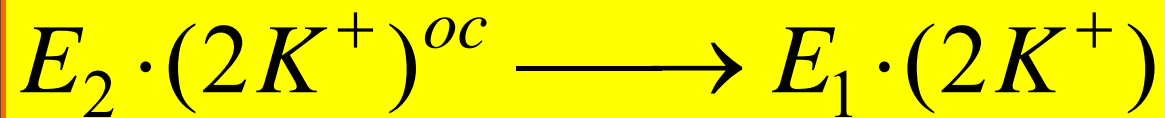


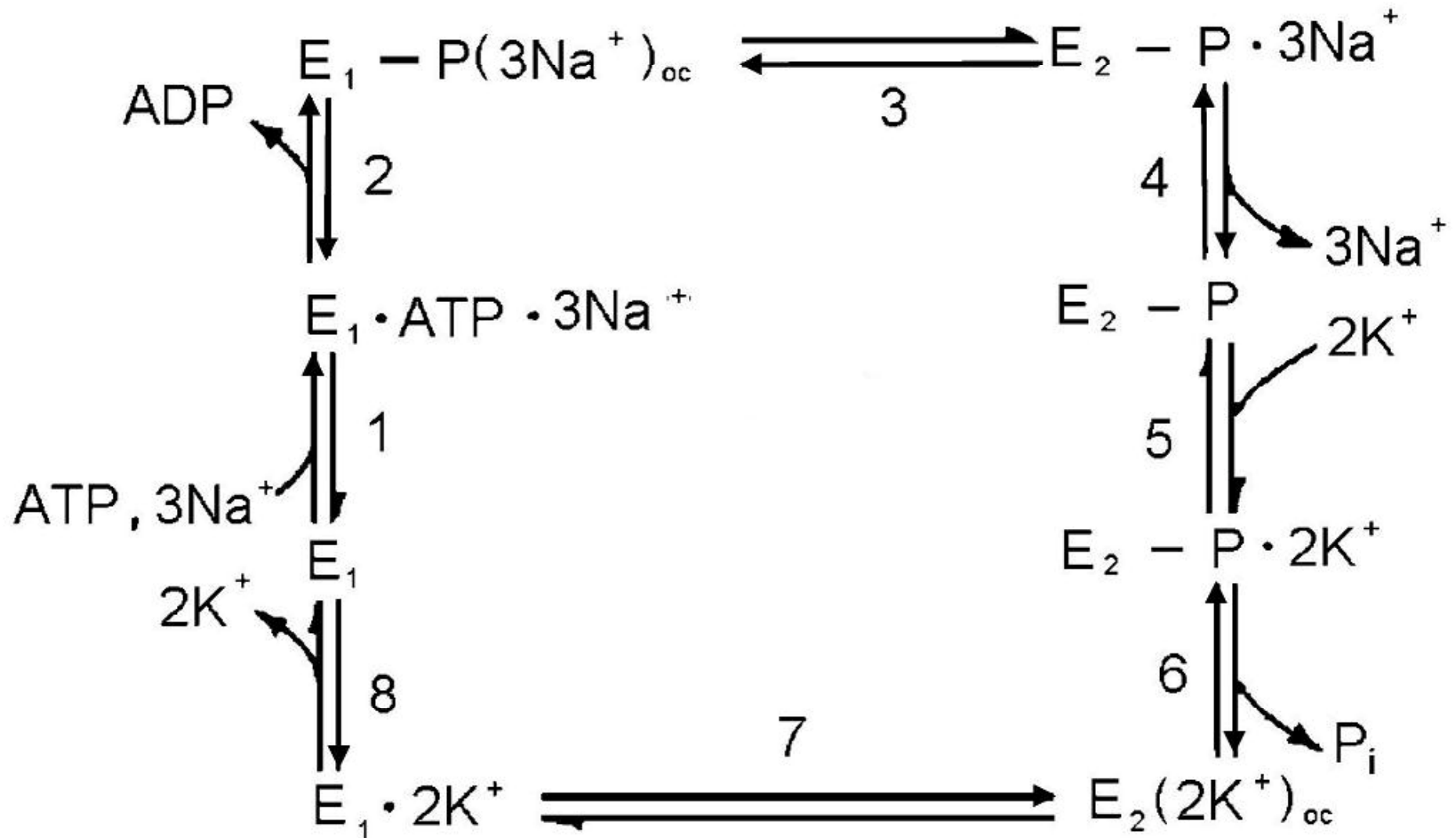
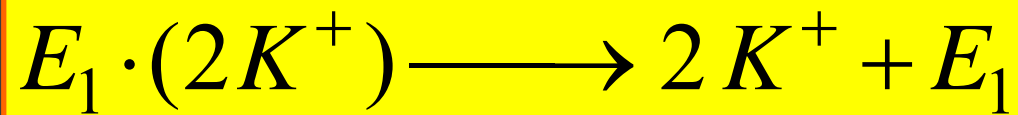




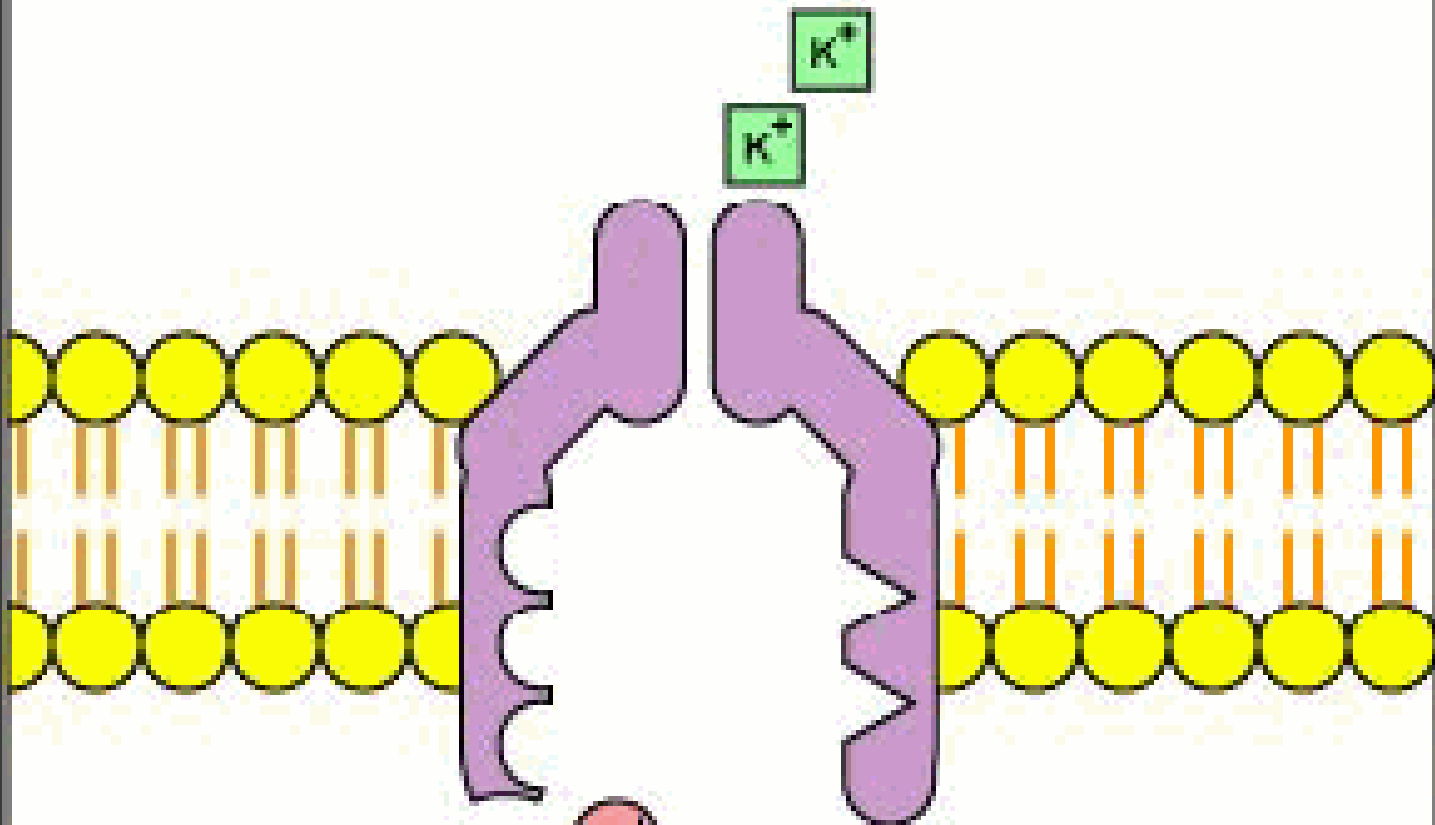








Outside



Inside

## TRANSPORTUL ACTIV SECUNDAR

➤ ENERGIA NECESARĂ REZULTĂ PRIN CUPLAREA CU UN PROCES DE TRANSPORT PASIV (EXERGONIC)

➤ EXEMPLE:

simport  $\text{Na}^+$ /glucoză (membrana citoplasmatică)

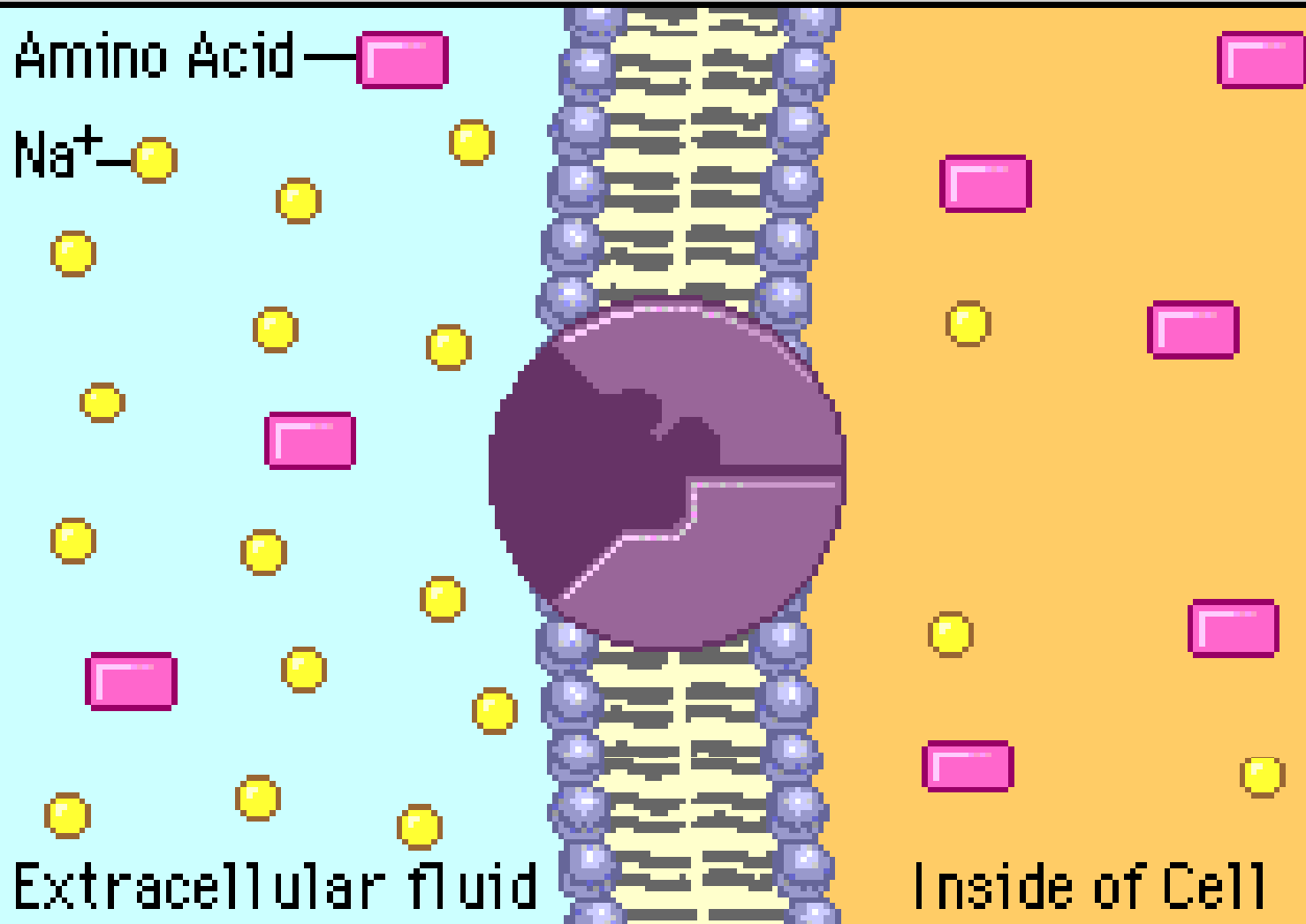
simport  $\text{Na}^+$ /colină (membrana presinaptică)

antiport  $\text{Na}^+$ / $\text{Ca}^{2+}$  (membrana celulelor excitabile)

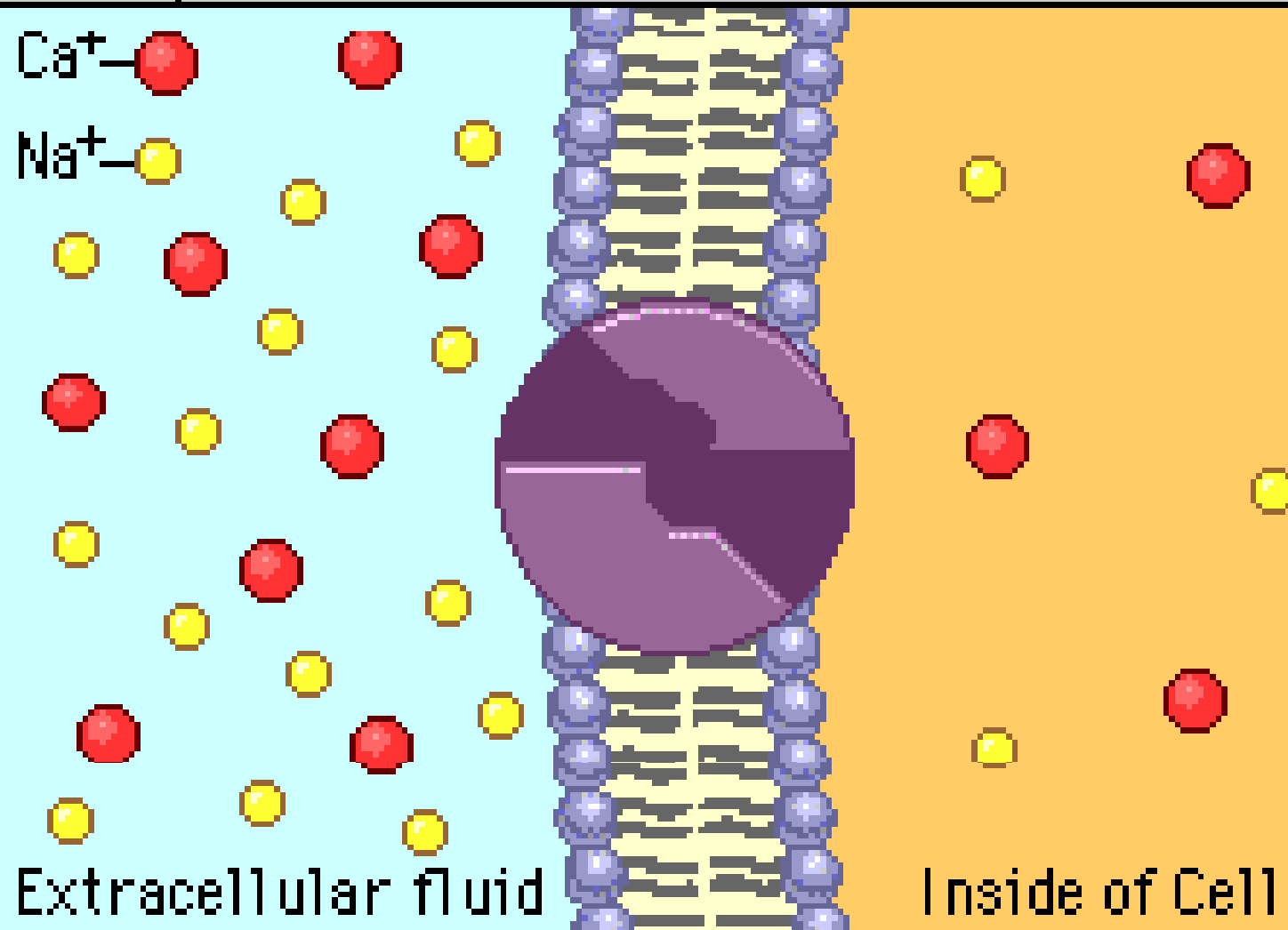
simport  $\text{Na}^+$ /aminoacizi (membrana citoplasmatică)

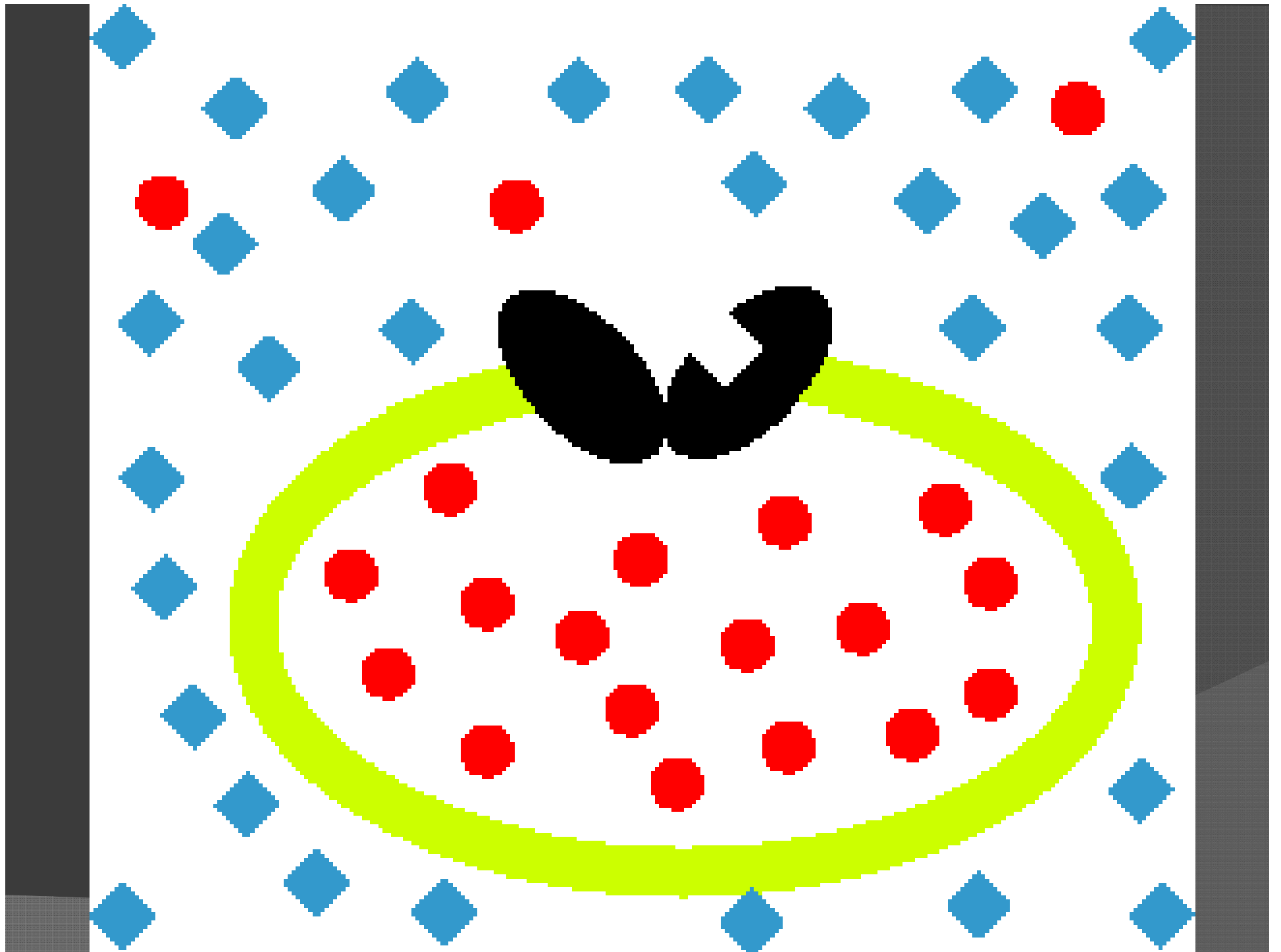


Step 1. Two substrates bind to the extracellular side of the transporter. One substrate ( $\text{Na}^+$  ion) is traveling “downhill”, and will energize transport of the other substrate (amino acid).



Step 1. Two substrates bind to the opposite sides of the transporter. One substrate ( $\text{Na}^+$  ion) is traveling “downhill”, and will energize transport of the other substrate ( $\text{Ca}^{2+}$  ion).





# GLICOZIDE CARDIACE (OUABAINA)

➤ AGENȚI FARMACOLOGICI UTILIZAȚI ÎN INSUFICIENȚA CARDIACĂ

➤ INHIBĂ ACTIVITATEA POMPEI DE  $\text{Na}^+/\text{K}^+$



Scade ritmul expulzării ionilor de  $\text{Ca}^{2+}$  prin antiport  $\text{Na}^+/\text{Ca}^{2+}$ .

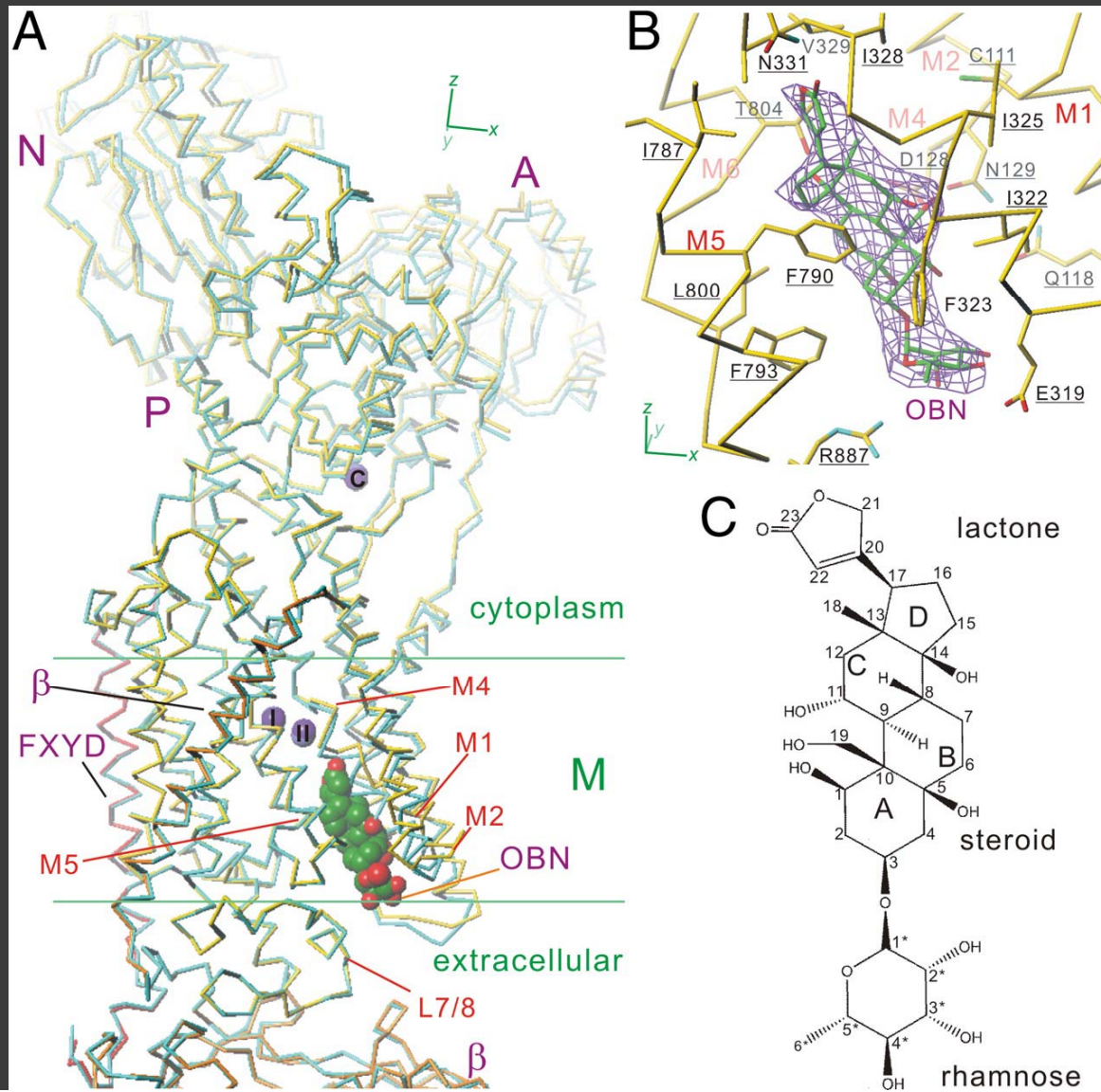


Crește concentrația intracelulară de  $\text{Ca}^{2+}$ .



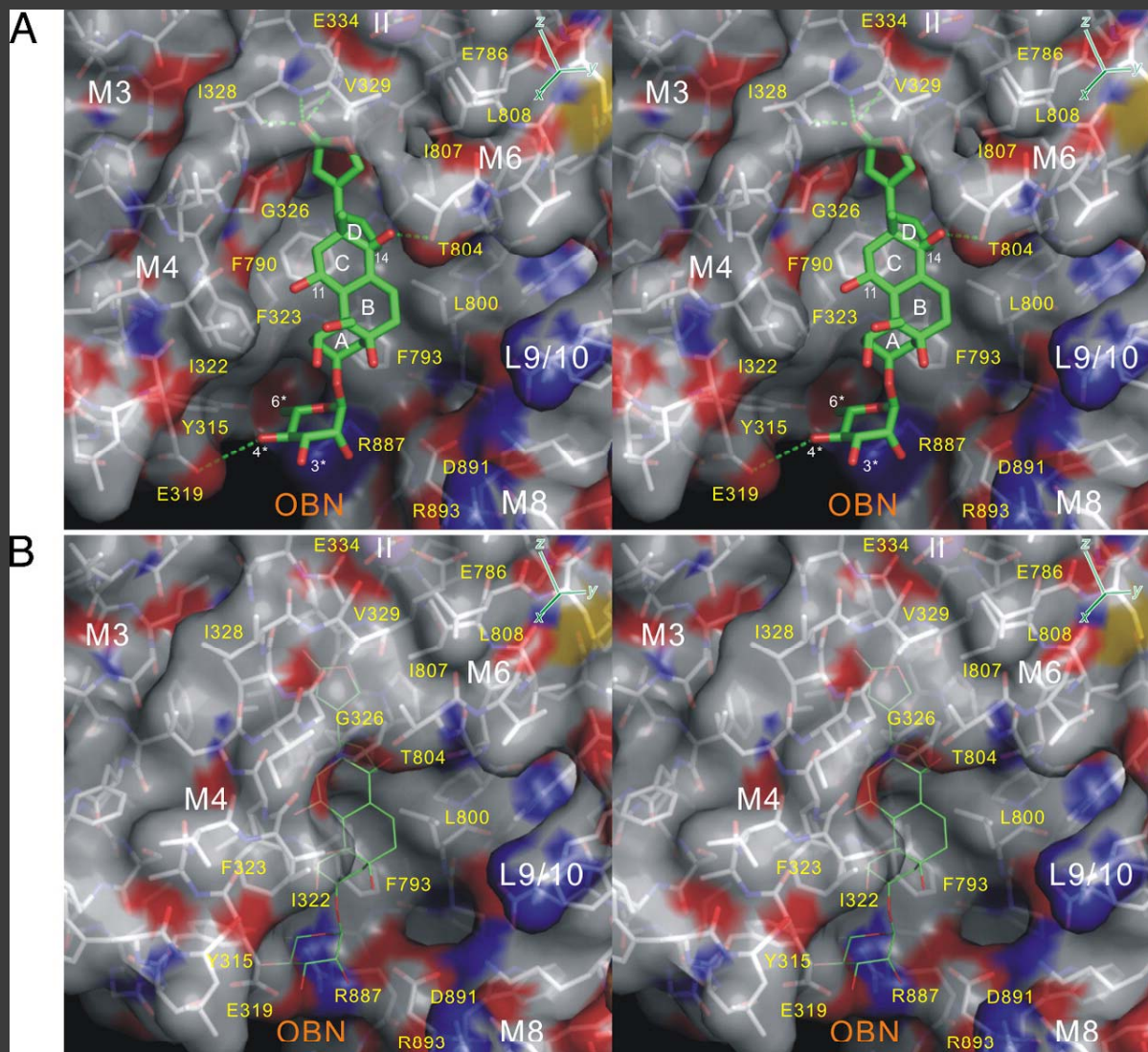
Mușchiul cardiac se contractă mai puternic.

# LEGAREA OUABAINEI LA POMPA DE $\text{Na}^+/\text{K}^+$



Ogawa H et al. PNAS 2009;106:13742-13747

# MODIFICĂRI STRUCTURALE ÎN REGIUNEA ELICILOR TRANSMEMBRANARE M4–M6 CA URMARE A LEGĂRII OUABAINEI



Ogawa H et al. PNAS 2009;106:13742-13747



# STUDII EXPERIMENTALE

- **USSING - ZERAHN** : transport prin membrane epiteliale
- **VOLTAGE CLAMP**: măsurarea curenților transmembranari la potențial fixat
- **PATCH CLAMP** : voltage clamp pe un petec de membrană; permite măsurarea unor curenți monocanal.
- **INHIBITORI**: TTX, TEA, ouabaina.
- **IZOTOPI RADIOACTIVI**: măsurarea fluxurilor