

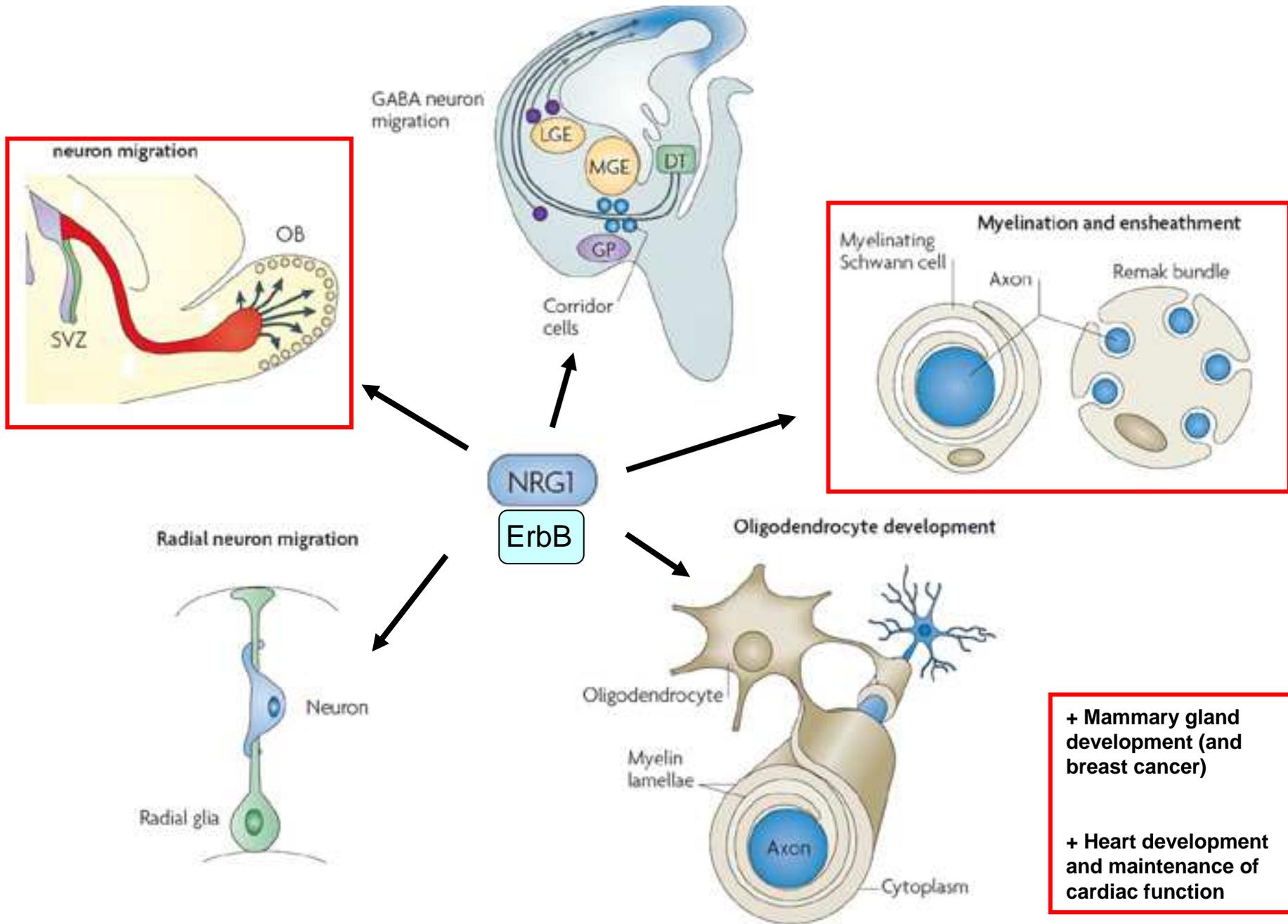
## Qual è il ruolo dello *splicing* alternativo nella comunicazione cellula-cellula?

- esempi di recettori e ligandi le cui capacità e modalità di segnalazione cambiano profondamente in seguito a *splicing* alternativo

## Qual è il ruolo dello *splicing* alternativo nella comunicazione mediata dalla Neuregulina 1?

- descrizione della Neuregulina 1, la proteina oggetto delle esercitazioni di Biotecnologie Cellulari

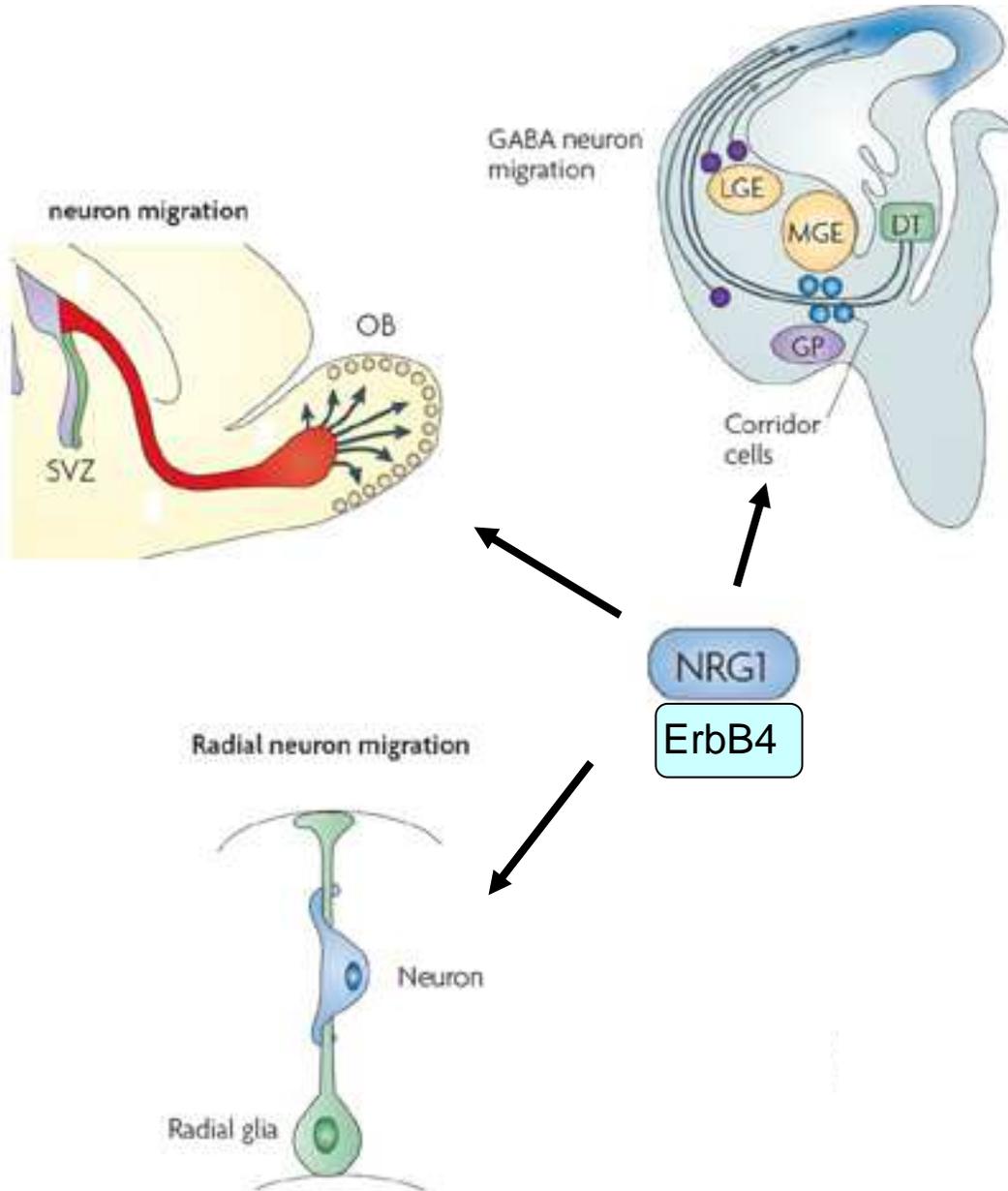
# Neuron migration and axon guidance



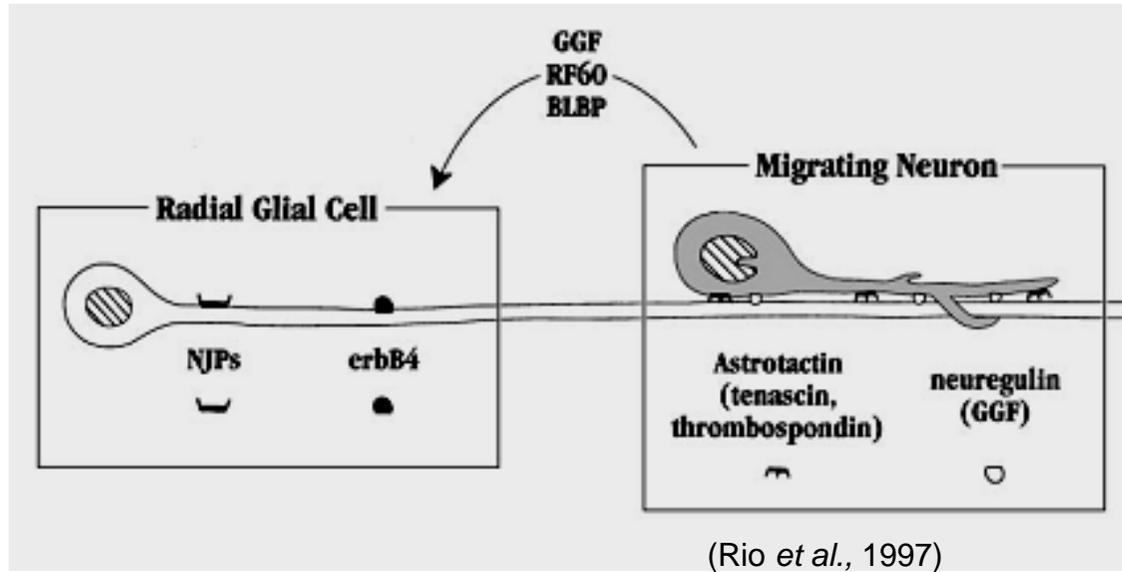
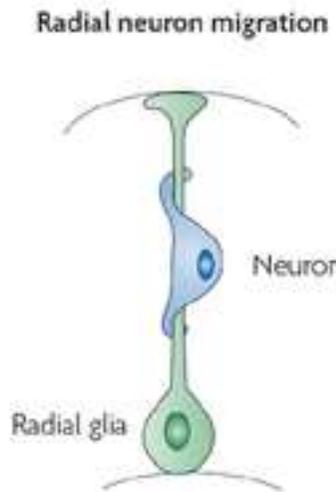
**+ Mammary gland development (and breast cancer)**

**+ Heart development and maintenance of cardiac function**

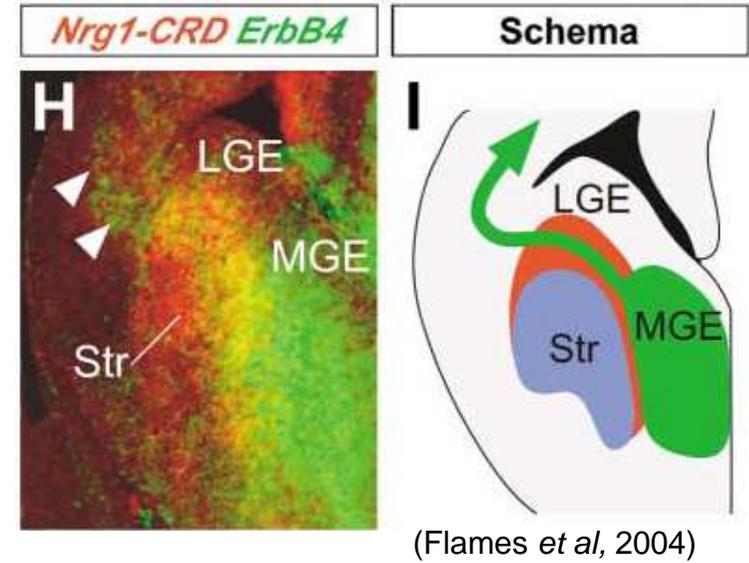
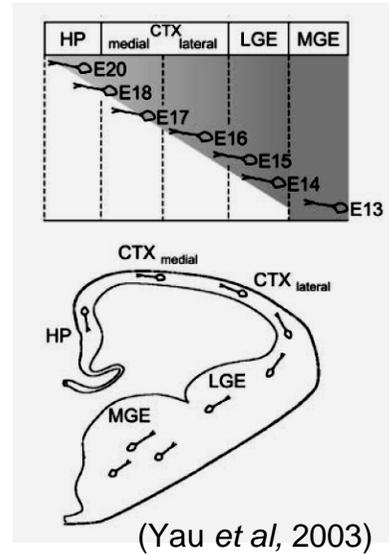
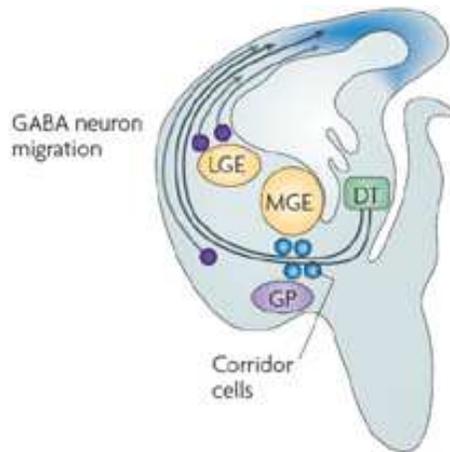
# Neuron migration and axon guidance



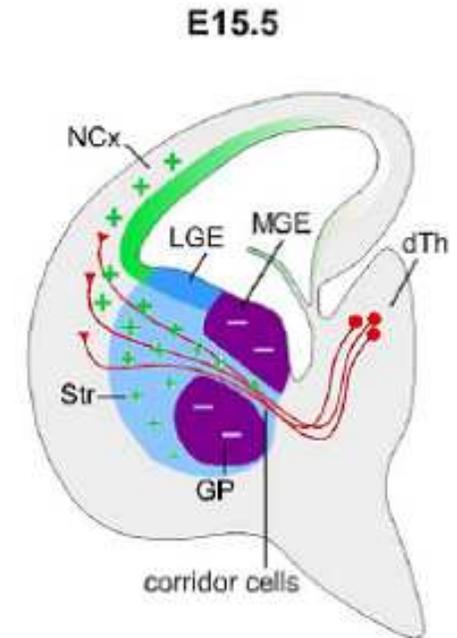
# Cerebellum



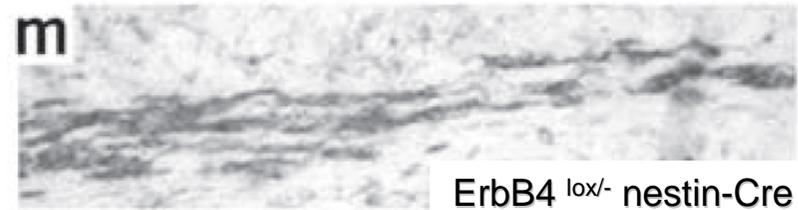
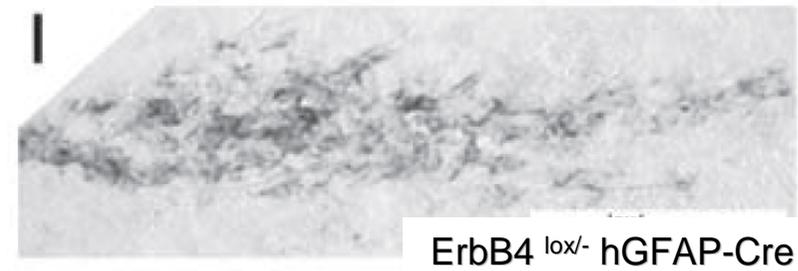
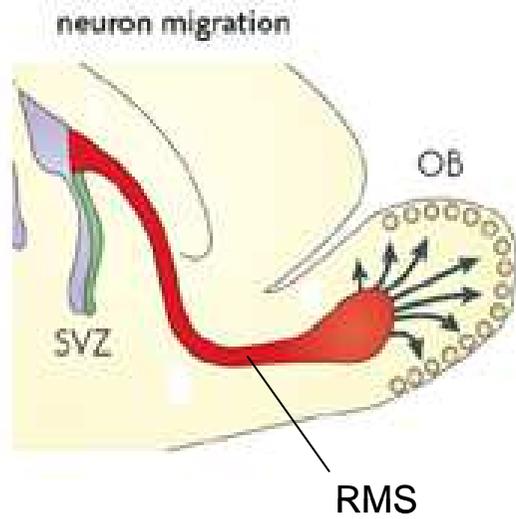
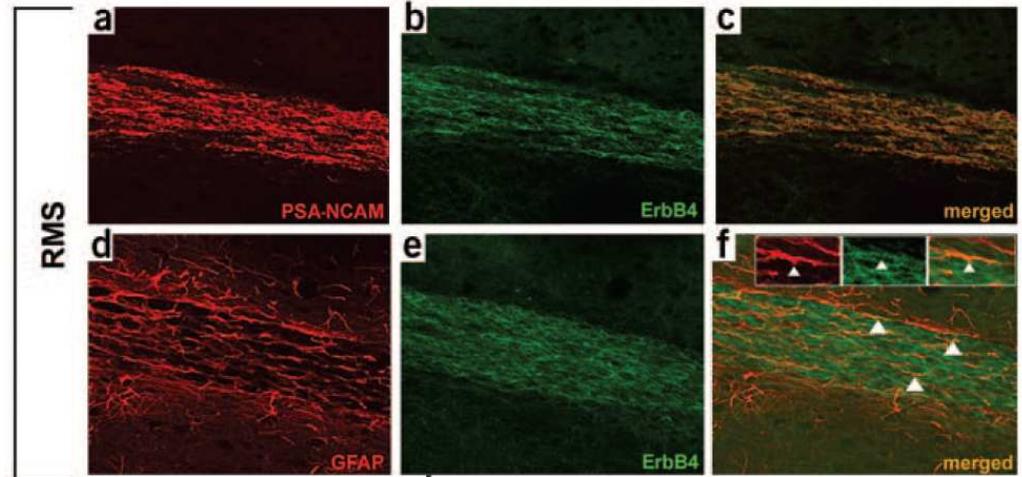
## Cortical interneuron migration



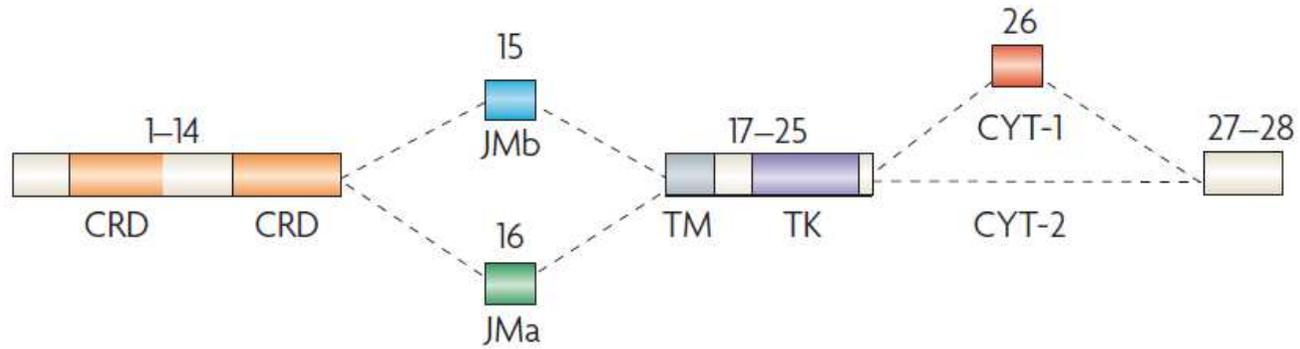
## Thalamocortical axon pathfinding



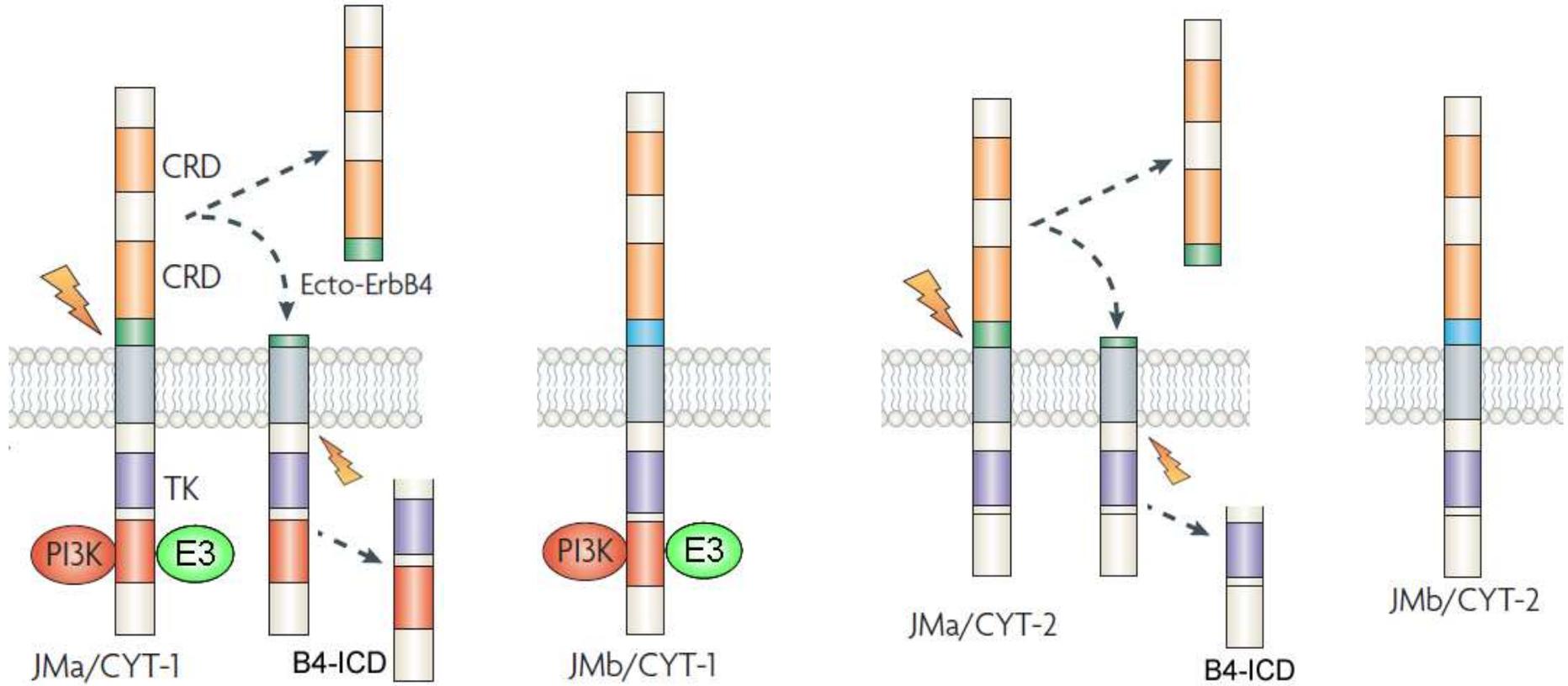
(Lopez-Bendito *et al*, 2006)



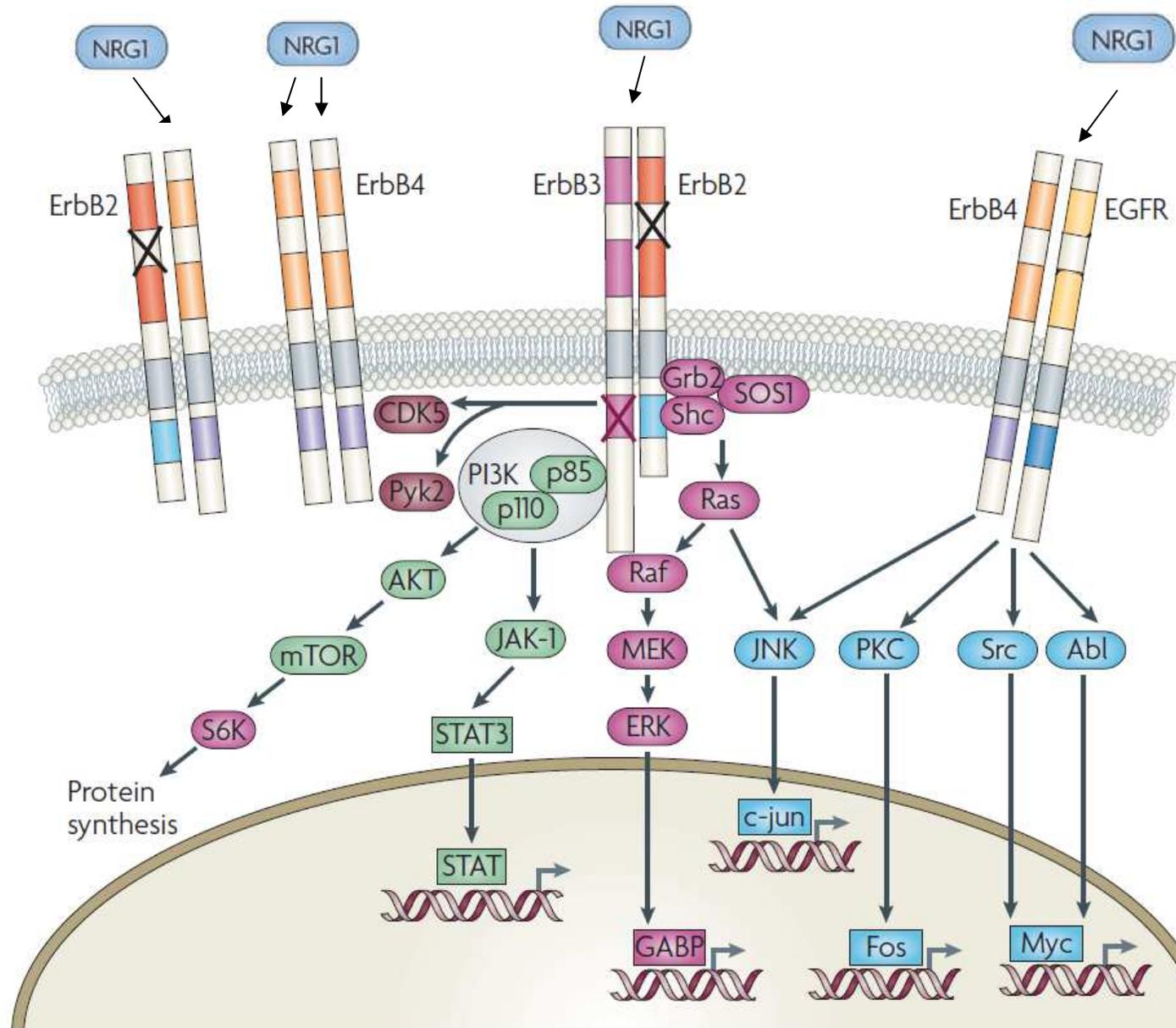
(Anton *et al.*, 2004)



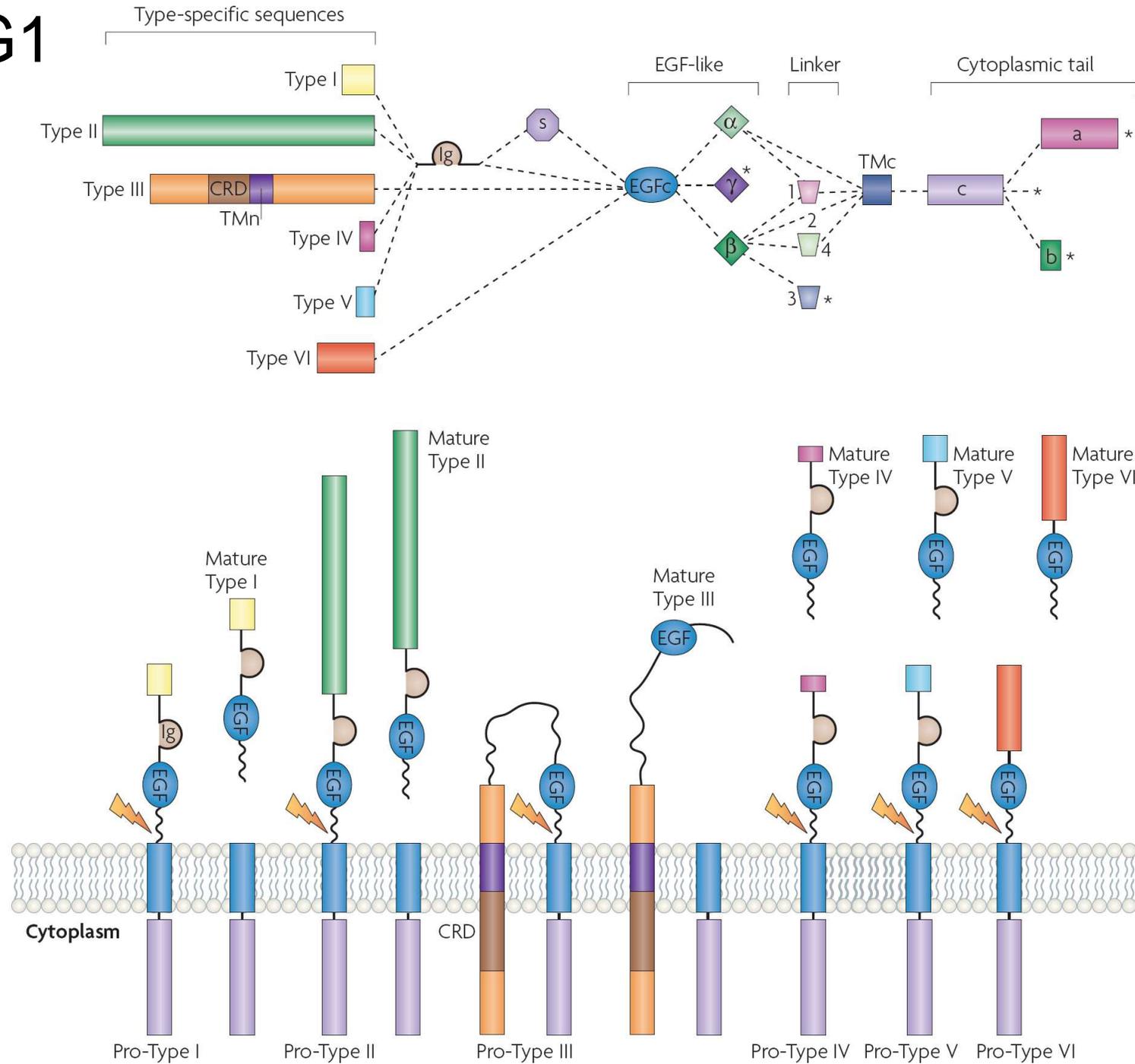
## ErbB4 isoforms

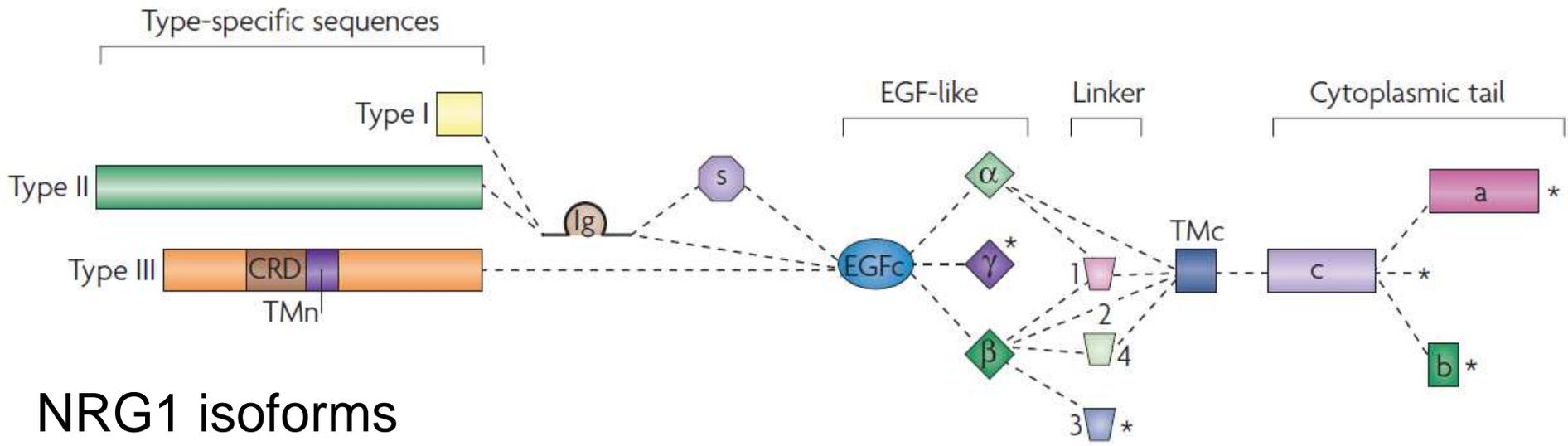


# ErbB receptor family

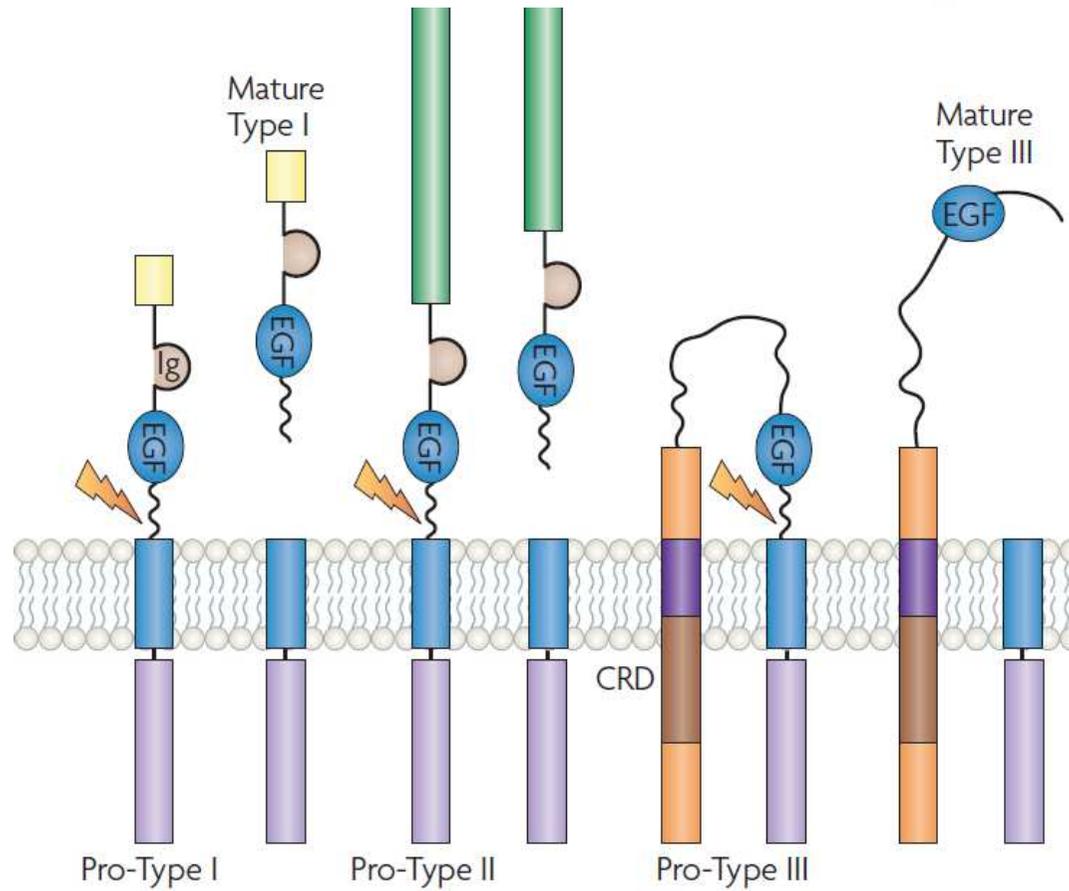


# NRG1

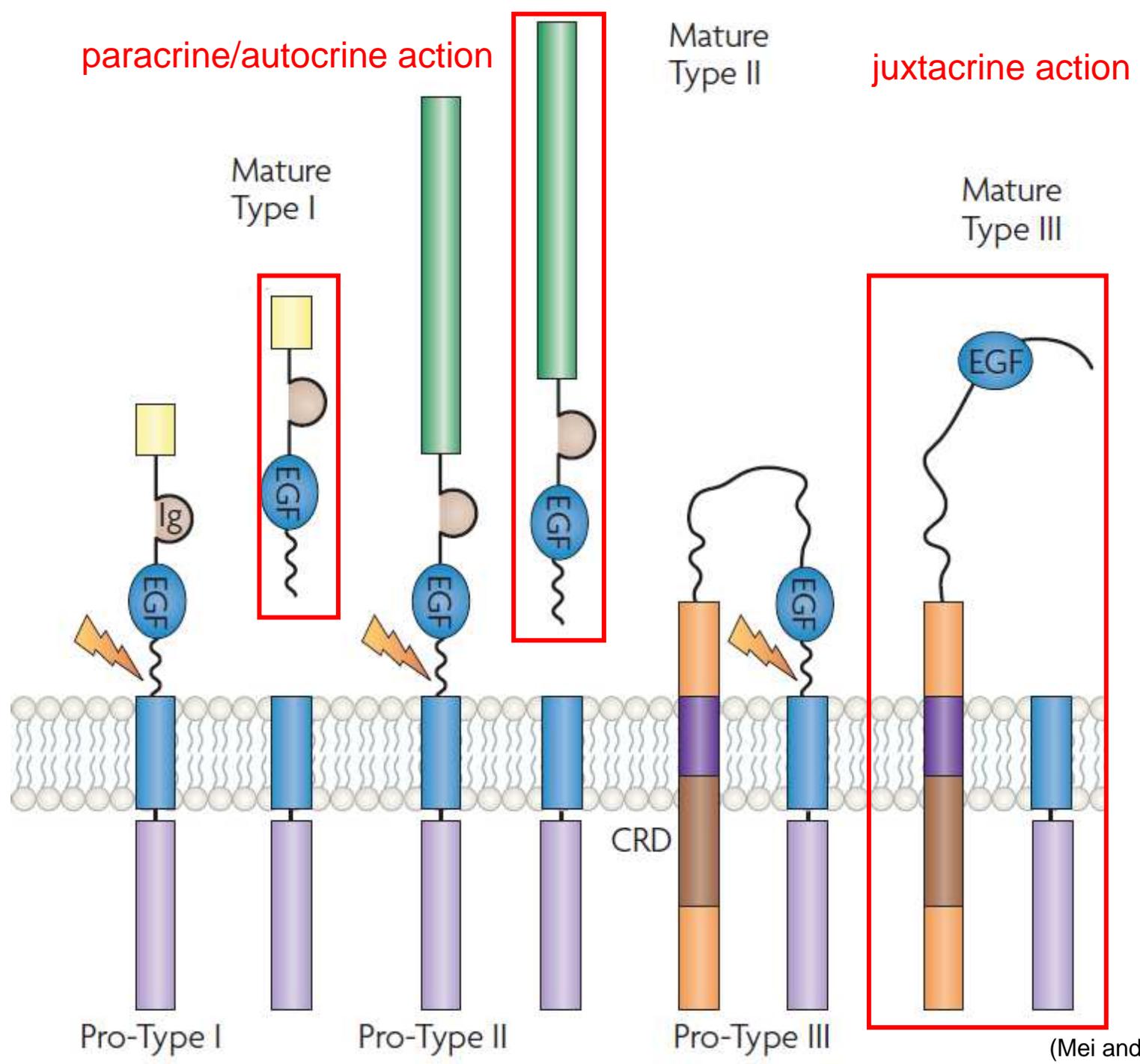




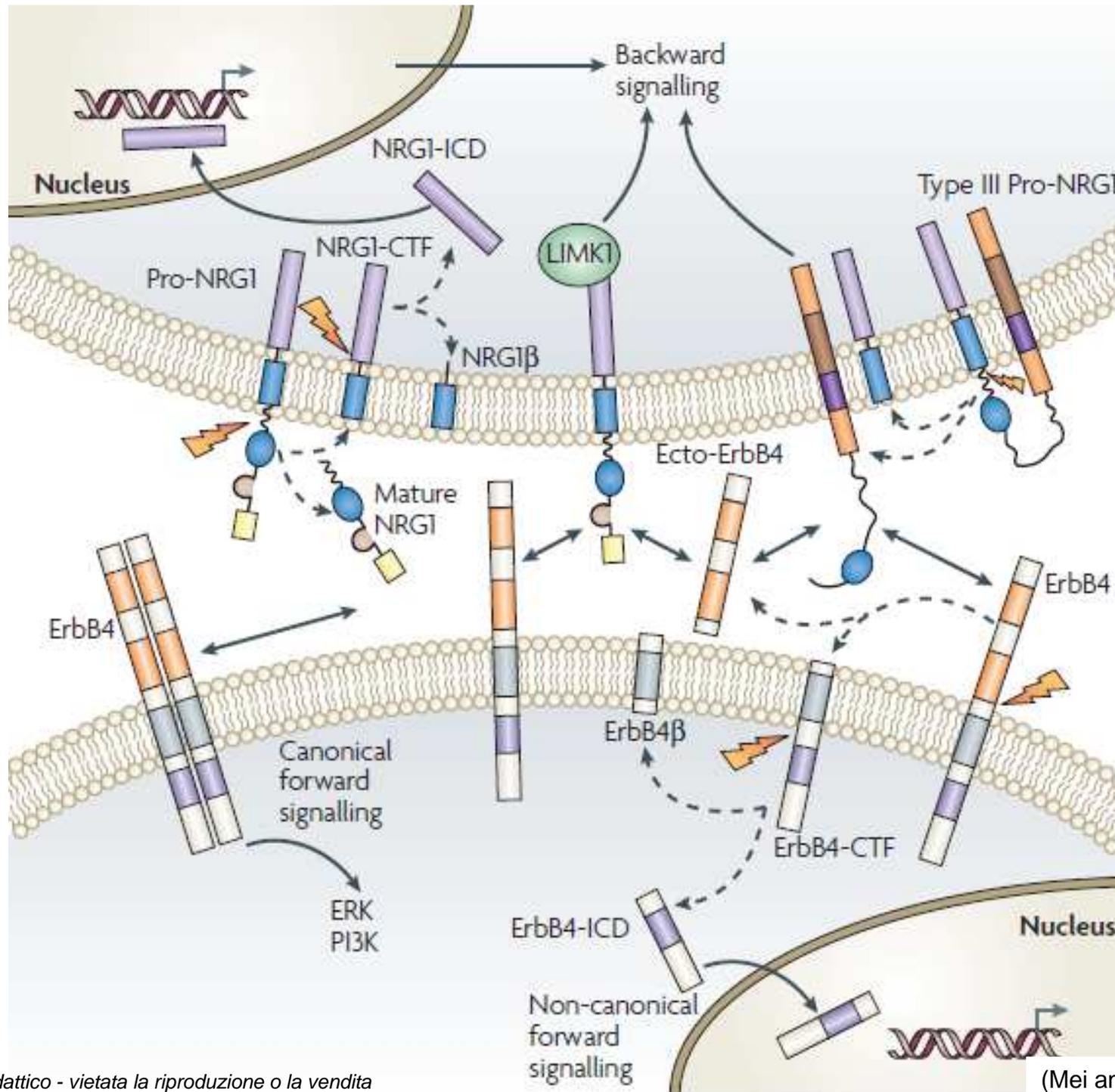
# NRG1 isoforms



(Mei and Xiong, 2008)



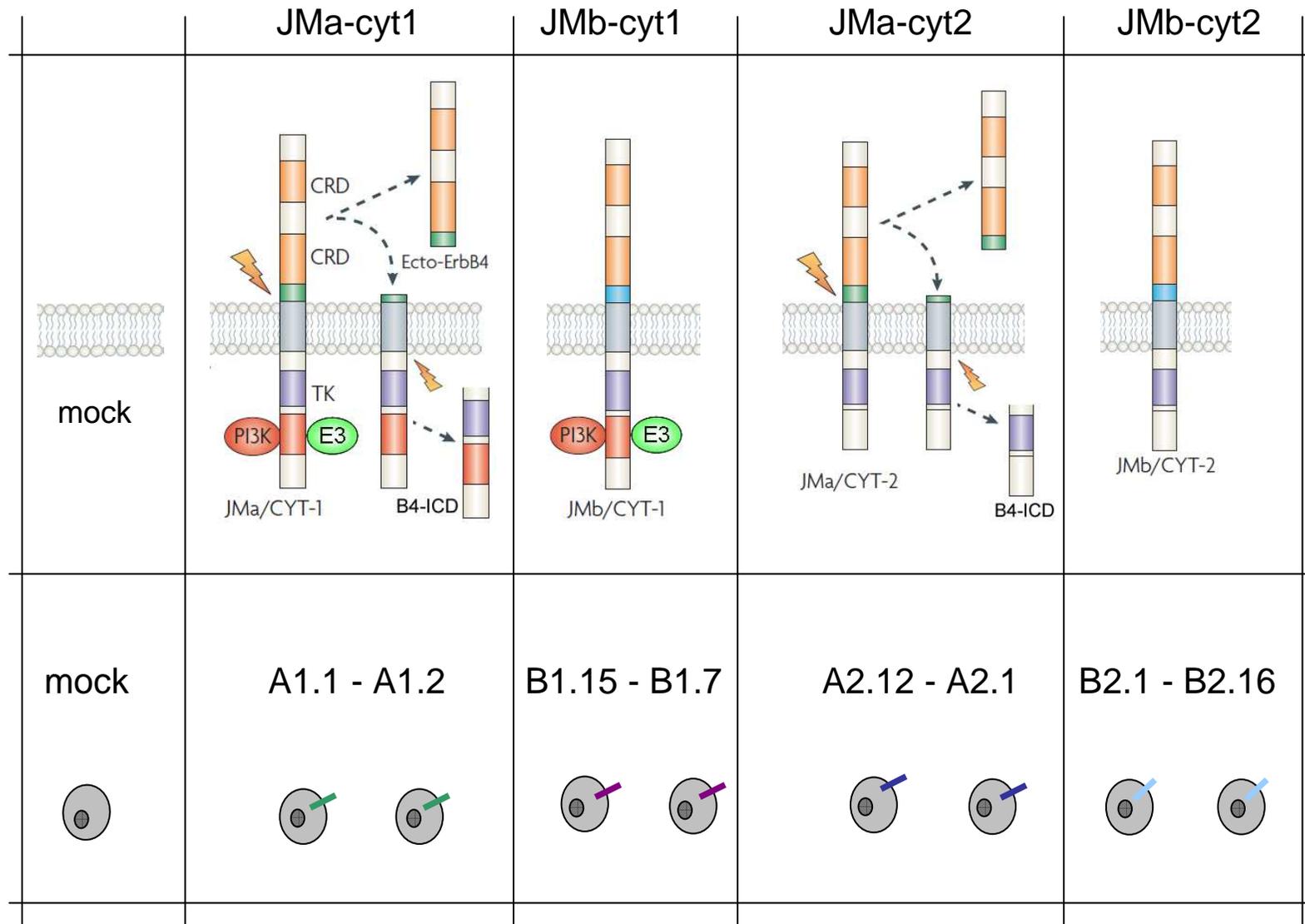
(Mei and Xiong, 2008)

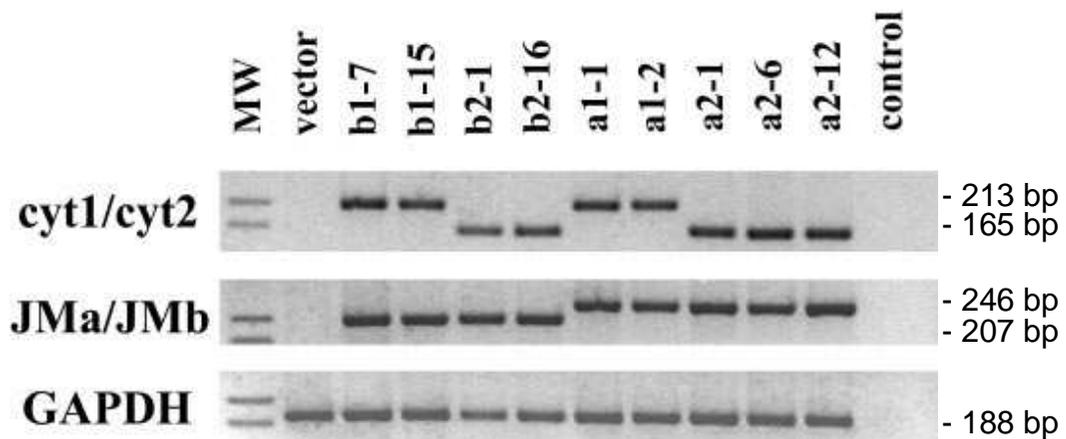
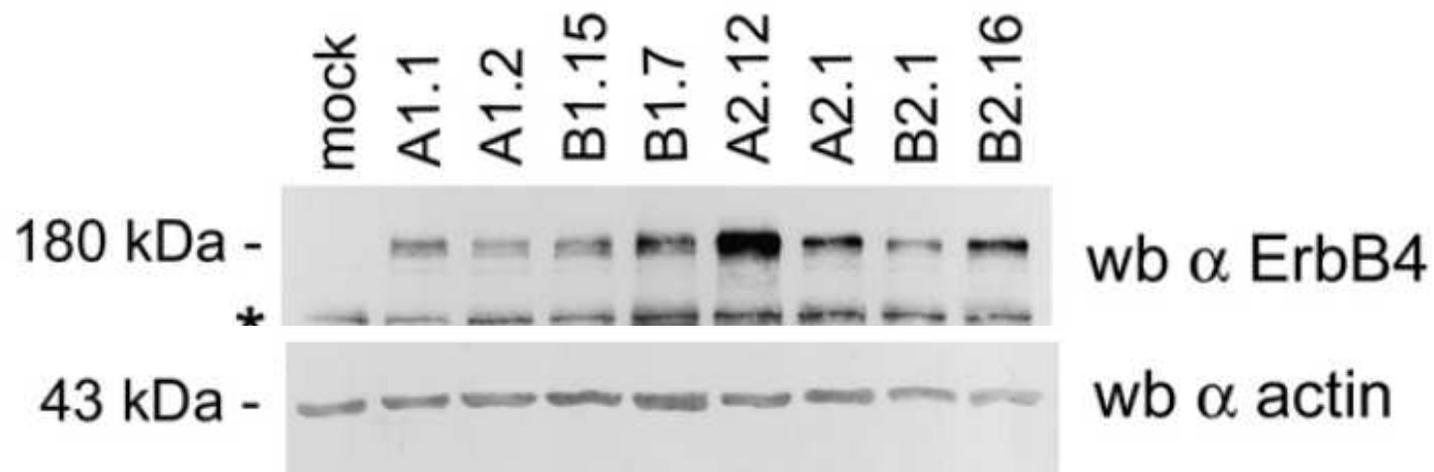
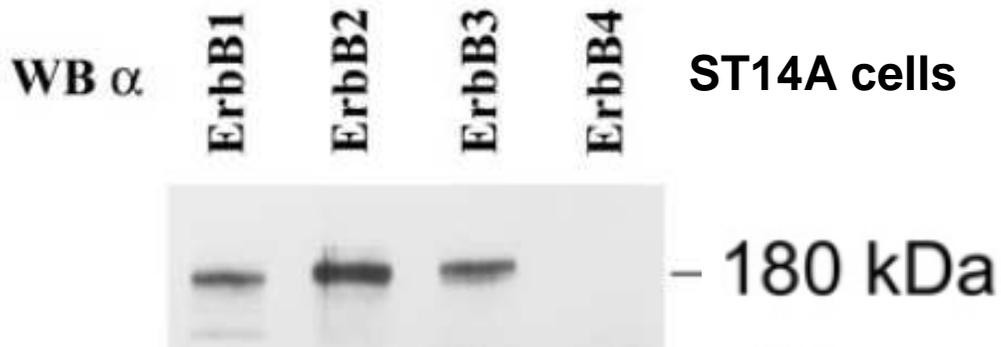


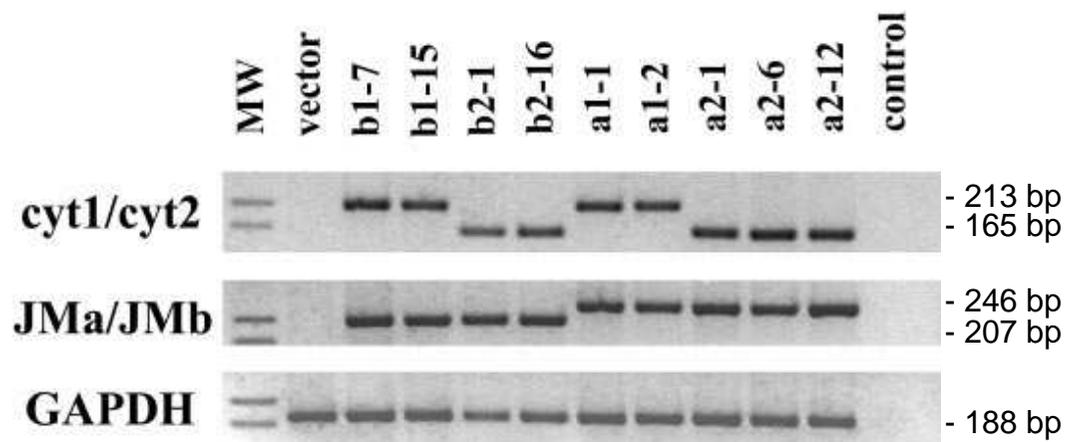
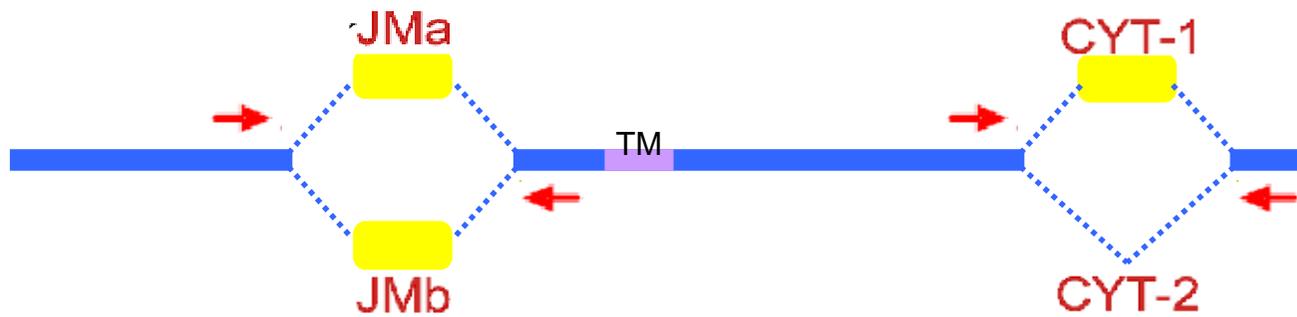
## The four ErbB4 isoforms and the neuronal migration

- *in vitro* analysis of substrate preference displayed by different ErbB4 isoforms through juxtacrine interactions
- *in vitro* analysis of migratory activity mediated by different ErbB4 isoforms through paracrine interactions
- *in vivo* expression of ErbB4 isoforms

# ErbB4 stable expression in neural progenitor cells (ST14A) endogenously expressing ErbB1, ErbB2, ErbB3





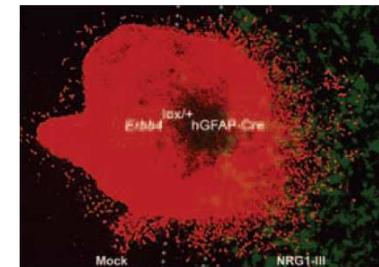
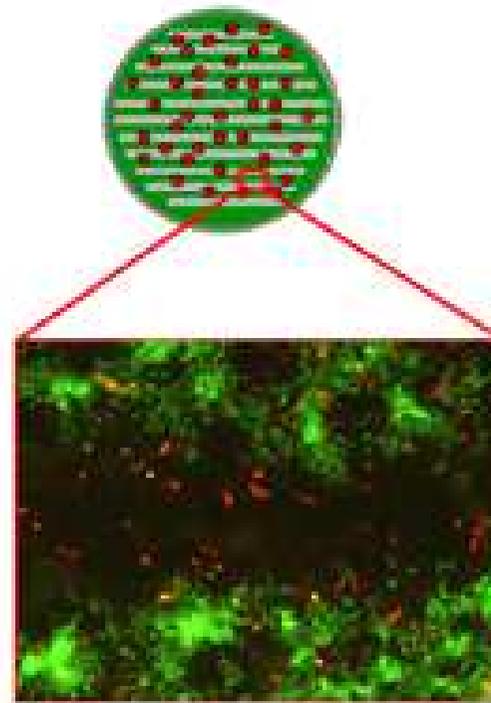
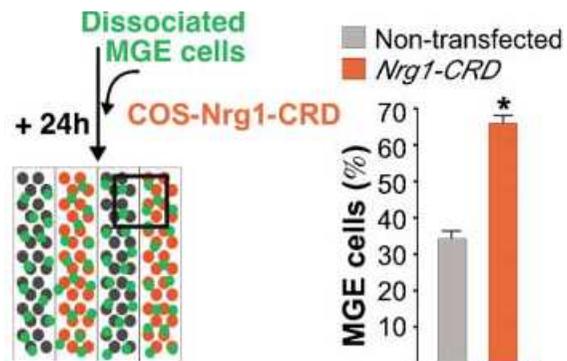


## The four ErbB4 isoforms and the neuronal migration

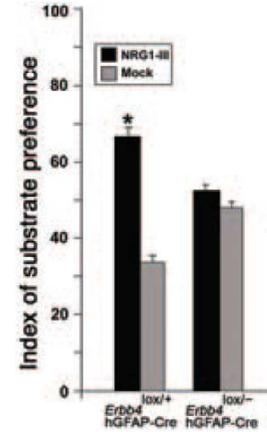
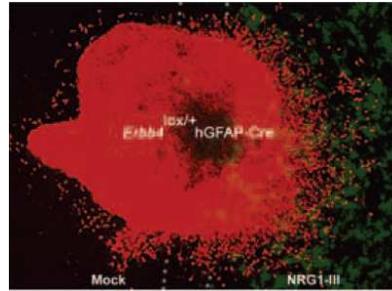
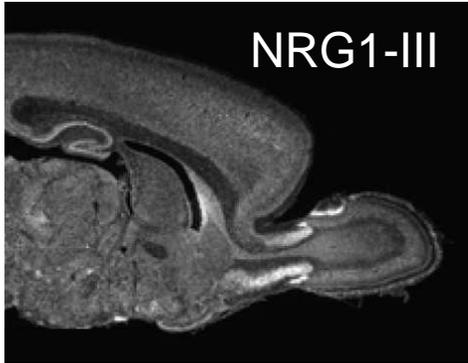
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# Qual è l'influenza del substrato su adesione & migrazione?

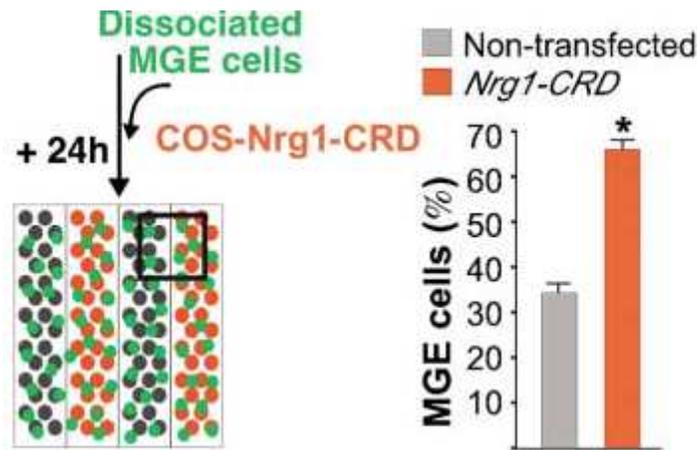
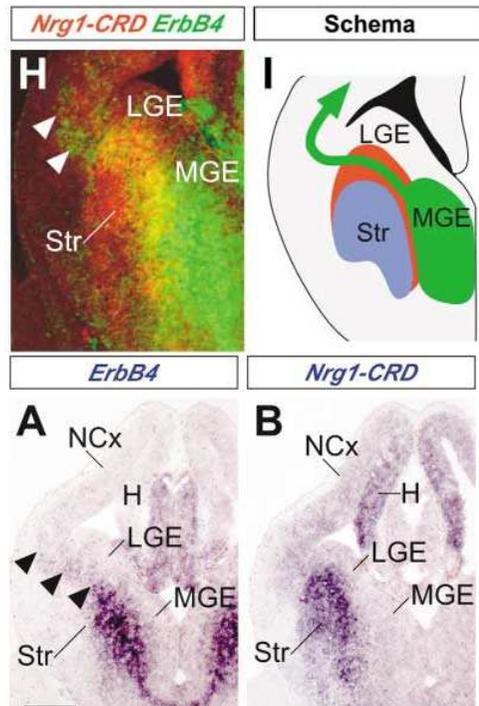
- approcci sperimentali per valutare la preferenza di adesione su un certo substrato



# *in vivo* ErbB4 expressing neurons adhere preferentially on a substrate expressing transmembrane NRG1

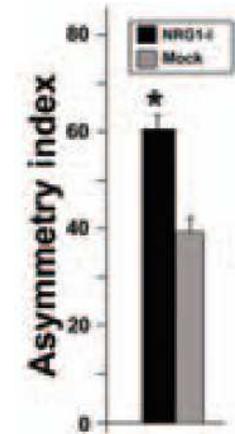
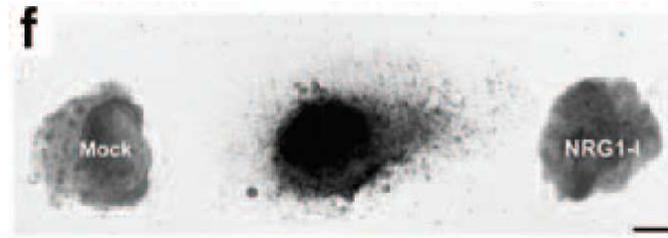


(Anton *et al*, 2004)

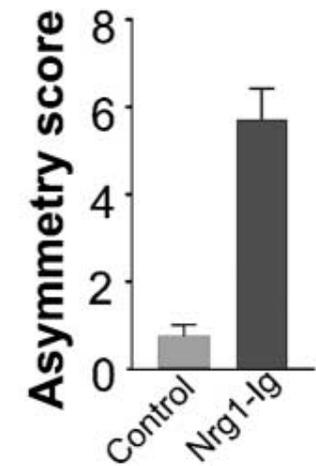
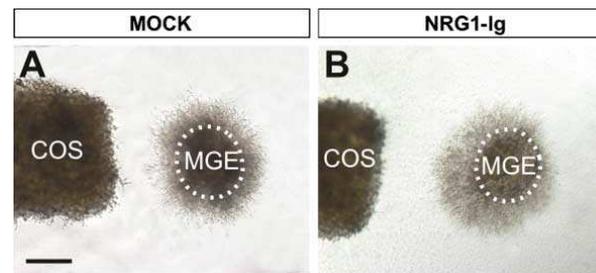
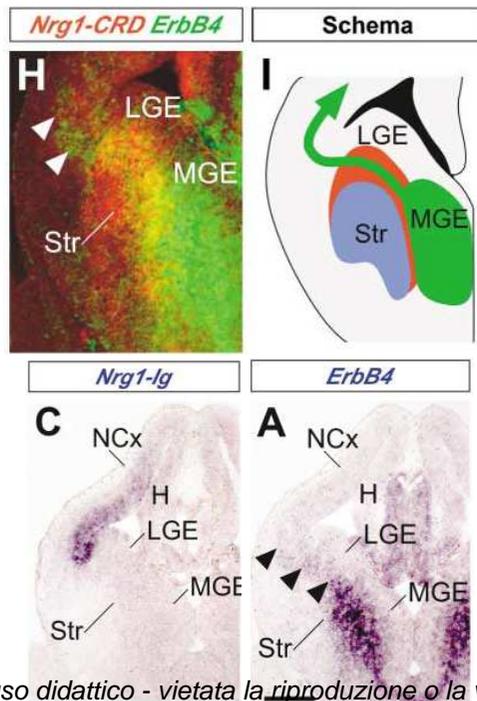


(Flames *et al*, 2004)

# *in vivo* ErbB4 expressing neurons are attracted by soluble NRG1

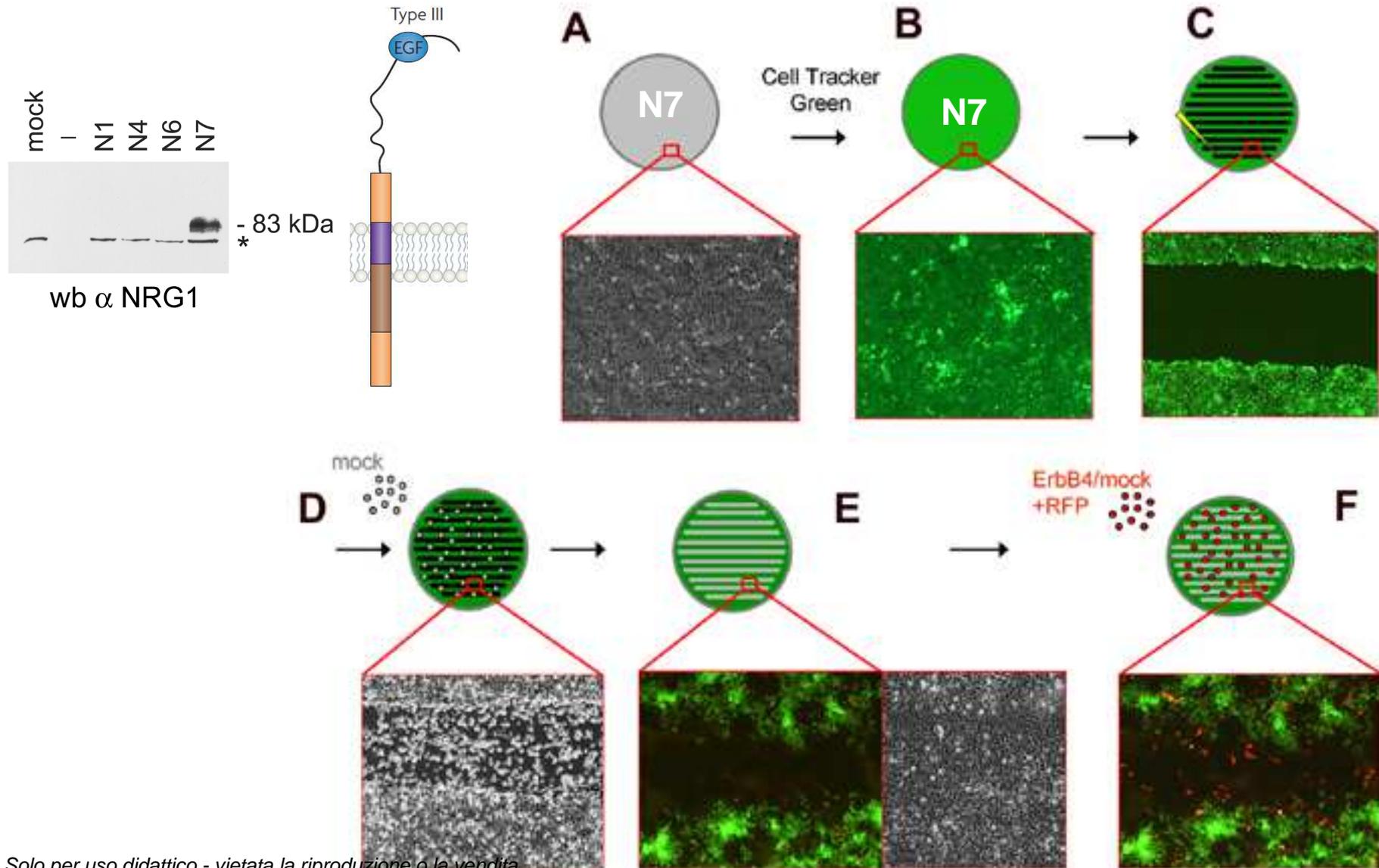


(Anton *et al*, 2004)

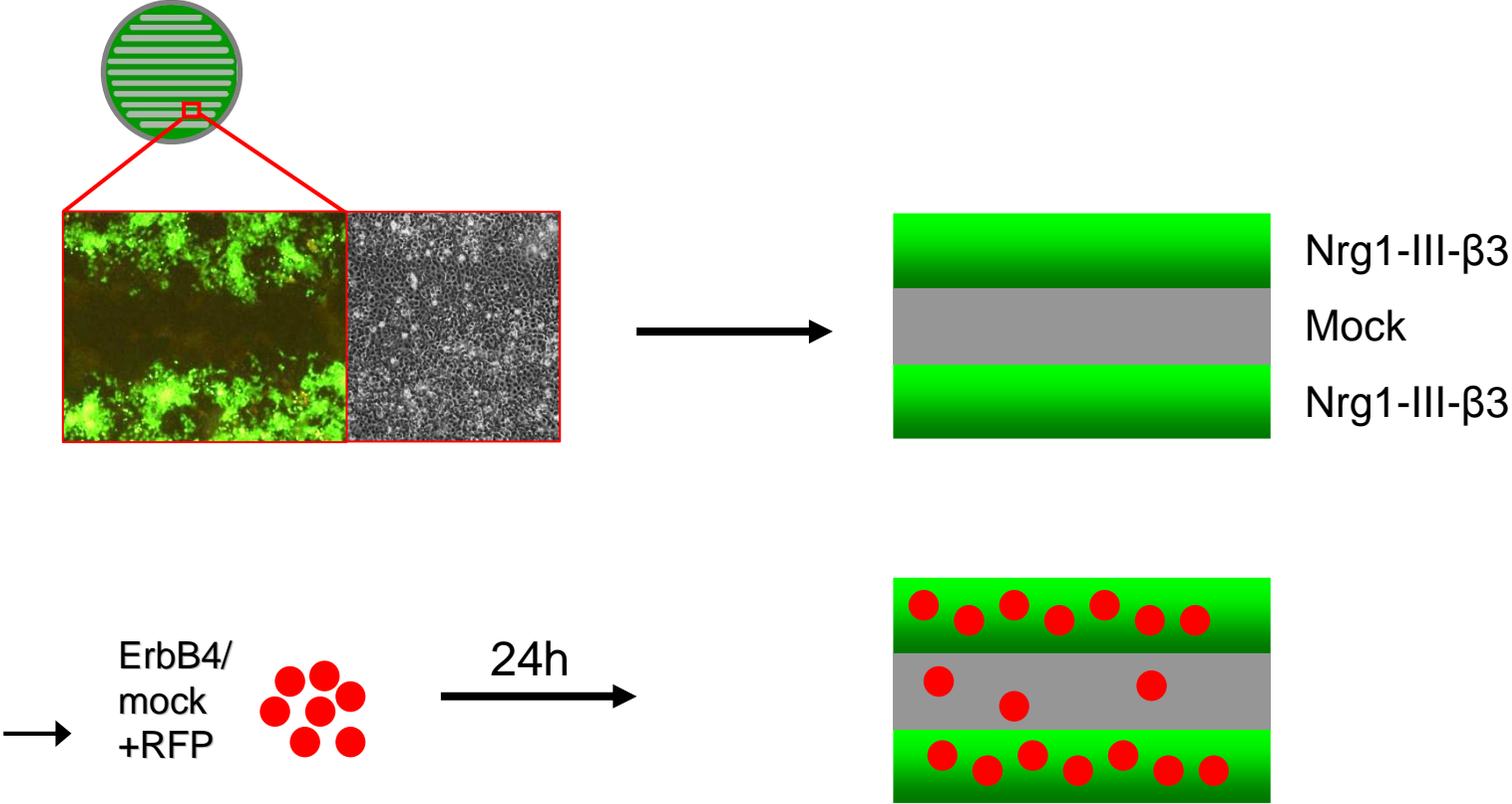


(Flames *et al*, 2004)

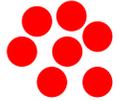
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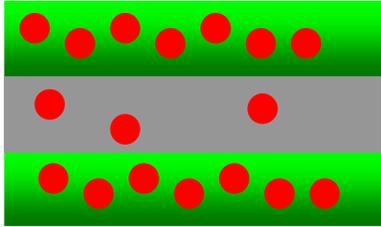
# Stripe Choice Assay



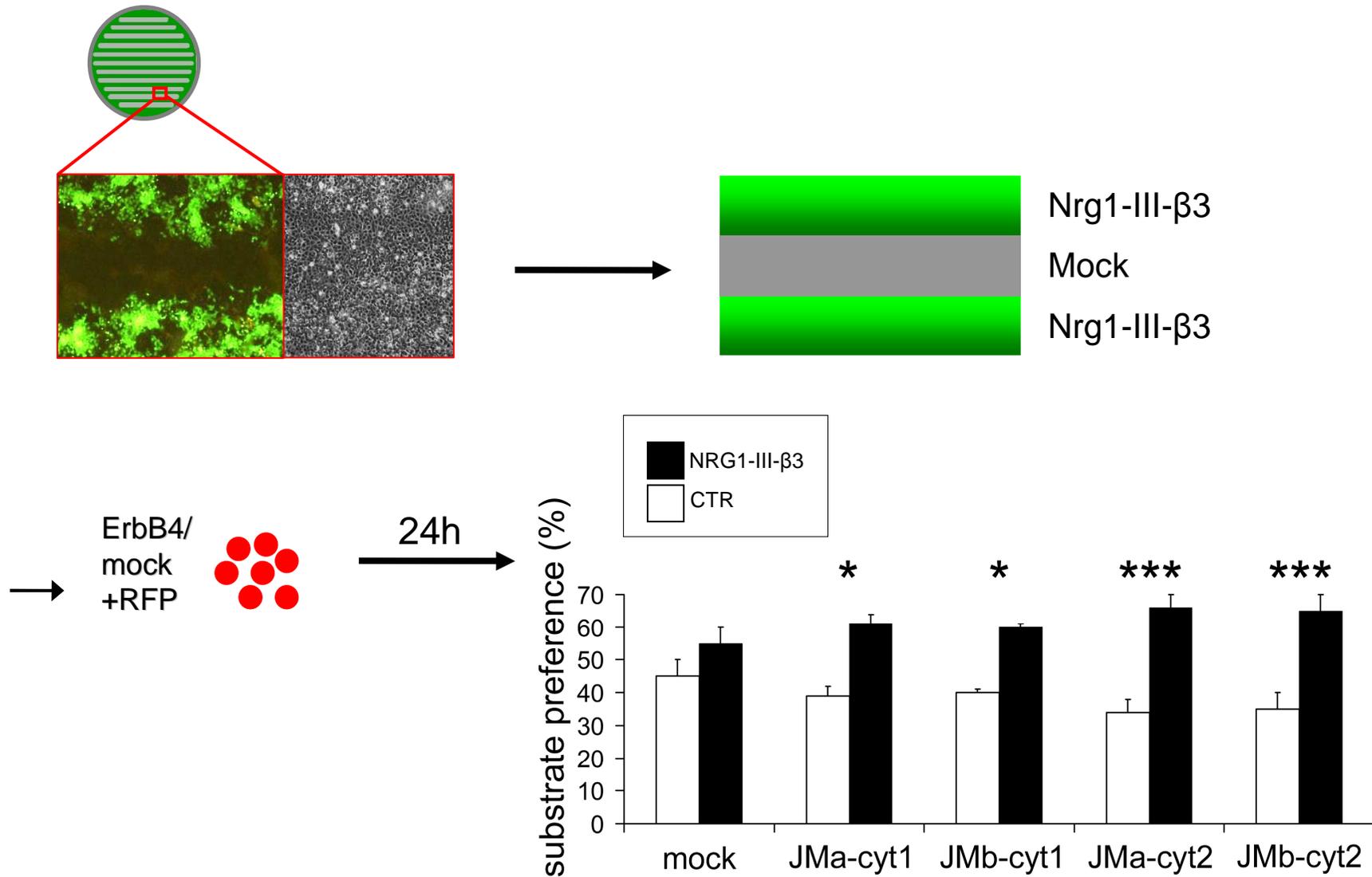
→ ErbB4/  
mock  
+RFP



24h →



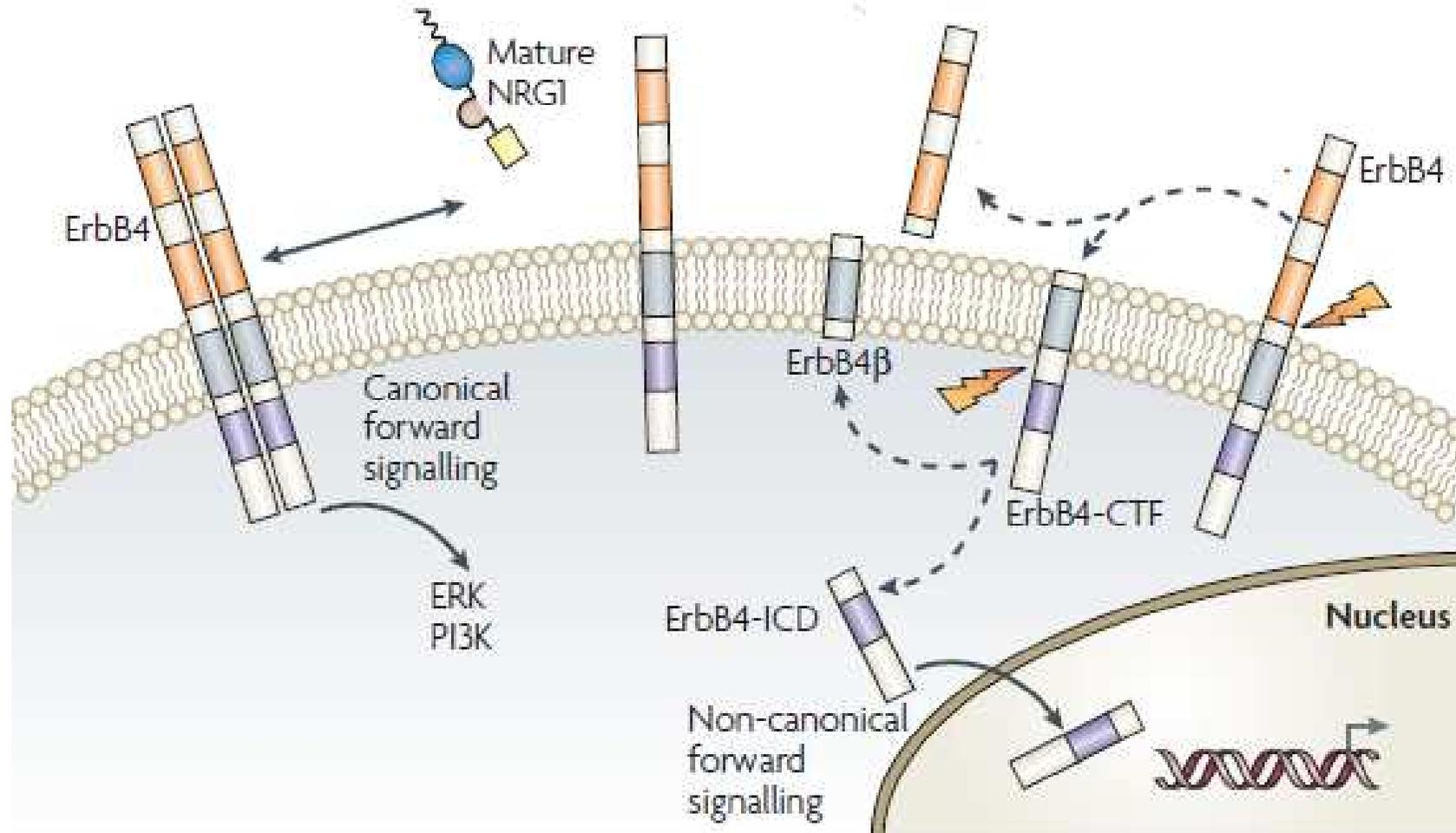
# Stripe Choice Assay



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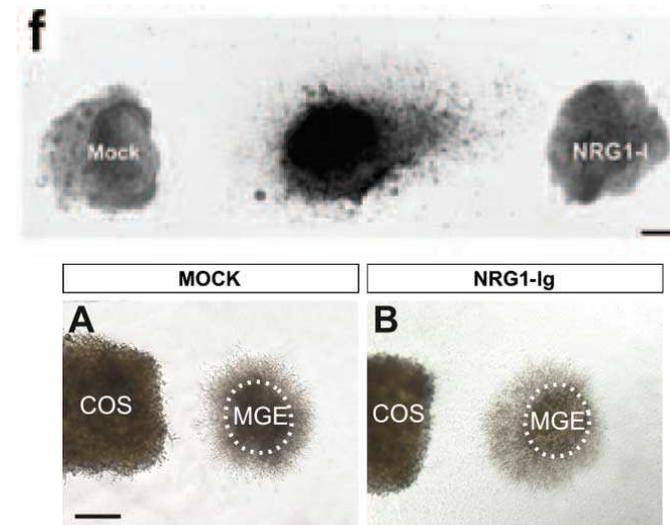
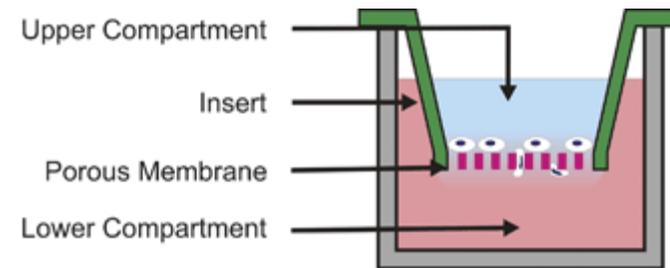
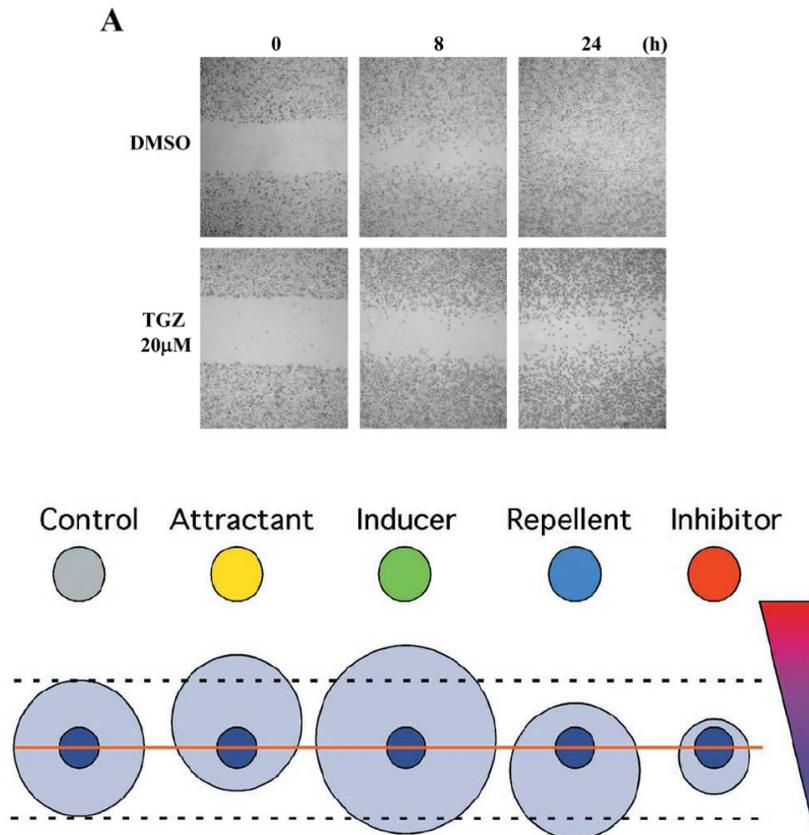
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# *In vitro* analysis of migratory activity mediated by different ErbB4 isoforms through paracrine interactions

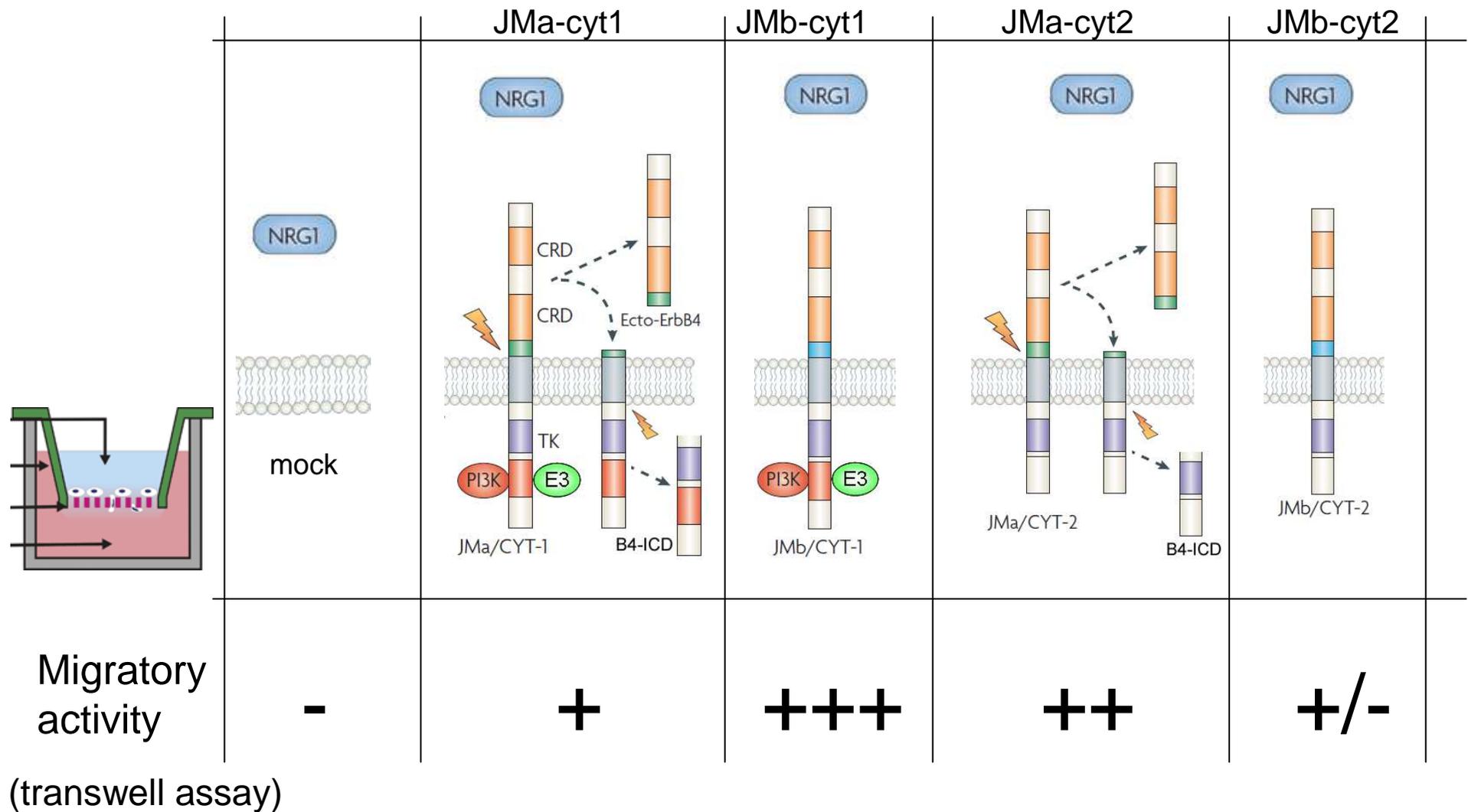


# Come si studia la migrazione cellulare?

- descrizione di diversi approcci sperimentali per studiare la migrazione cellulare, *in vitro* ed *in vivo*



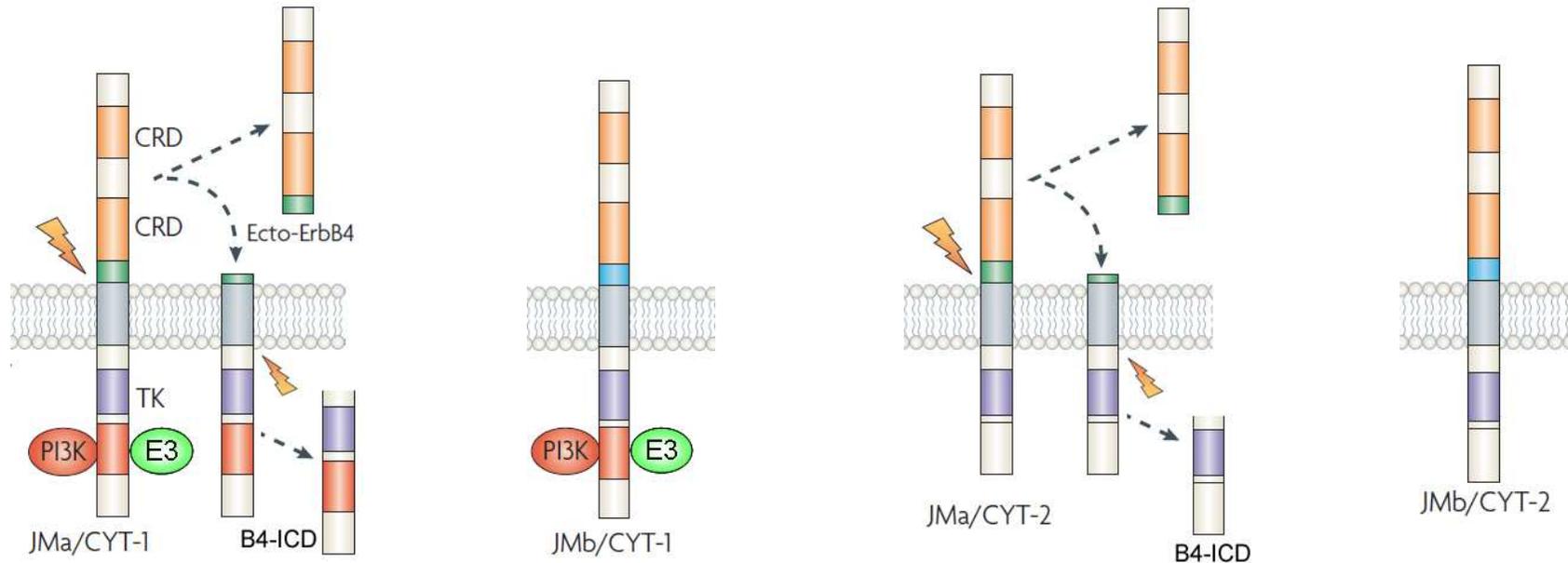
# ErbB4 expression in neural progenitor cells (ST14A) is necessary to mediate Neuregulin-1 $\beta$ 1-induced migration



## Conclusions

- ErbB4 isoforms confer the ability to preferentially adhere on a substrate stably expressing a transmembrane NRG1 isoform
- ErbB4 is necessary to mediate NRG1 $\beta$ 1-induced migration
- different ErbB4 isoforms confer different migratory activity

# Domande aperte

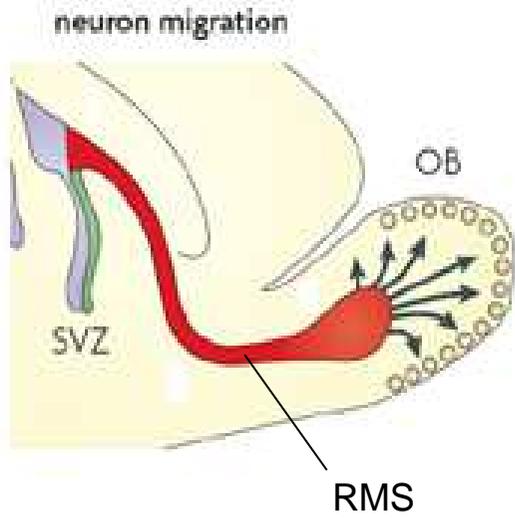


- quali vie di trasduzione sono attivate a valle delle diverse isoforme di ErbB4? (*akt, erk, ...?*)
- qual è la dinamica di attivazione di queste diverse vie? (*time course*)
- le diverse isoforme di ErbB4 manifestano diversa affinità per la NRG1? (*saggi dose/risposta*)
- le diverse isoforme di ErbB4 rispondono in maniera diversa allo stimolo con la NRG1  $\alpha$  o la  $\beta$ ?
- le diverse isoforme di ErbB4 manifestano diversa affinità per gli altri recettori ErbB? (*attivazione degli altri recettori, co-precipitazioni*)

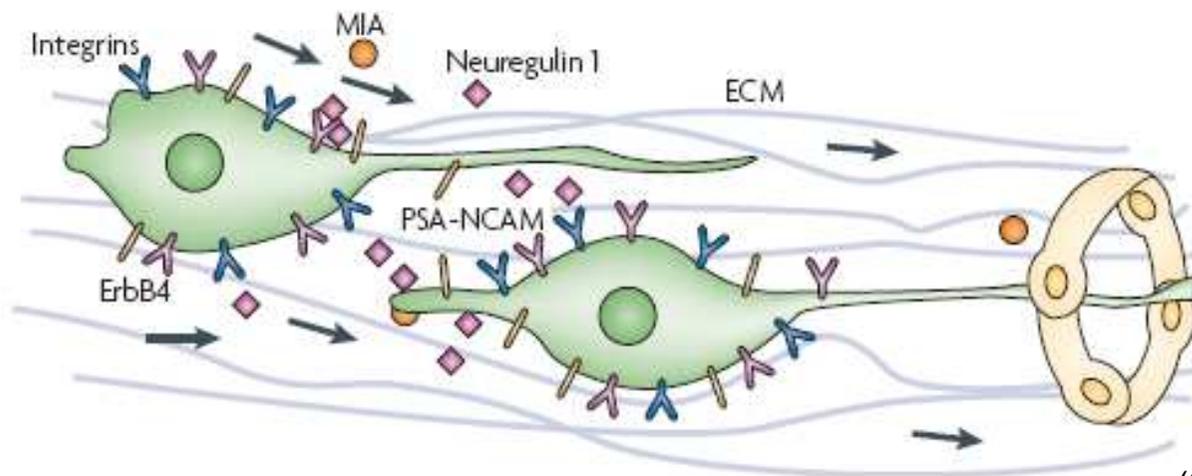
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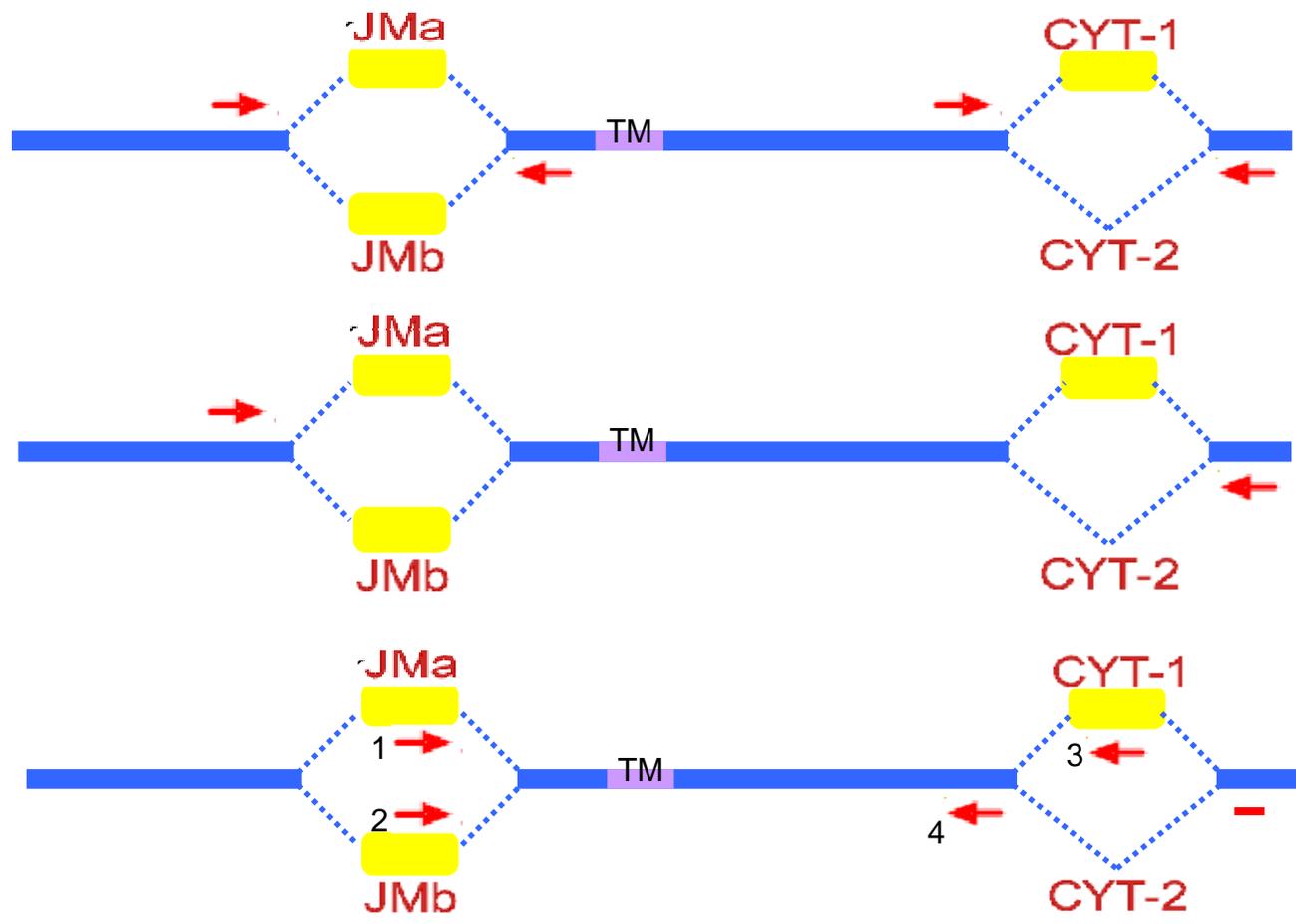
*in vivo*



(Anton *et al.*,2004)



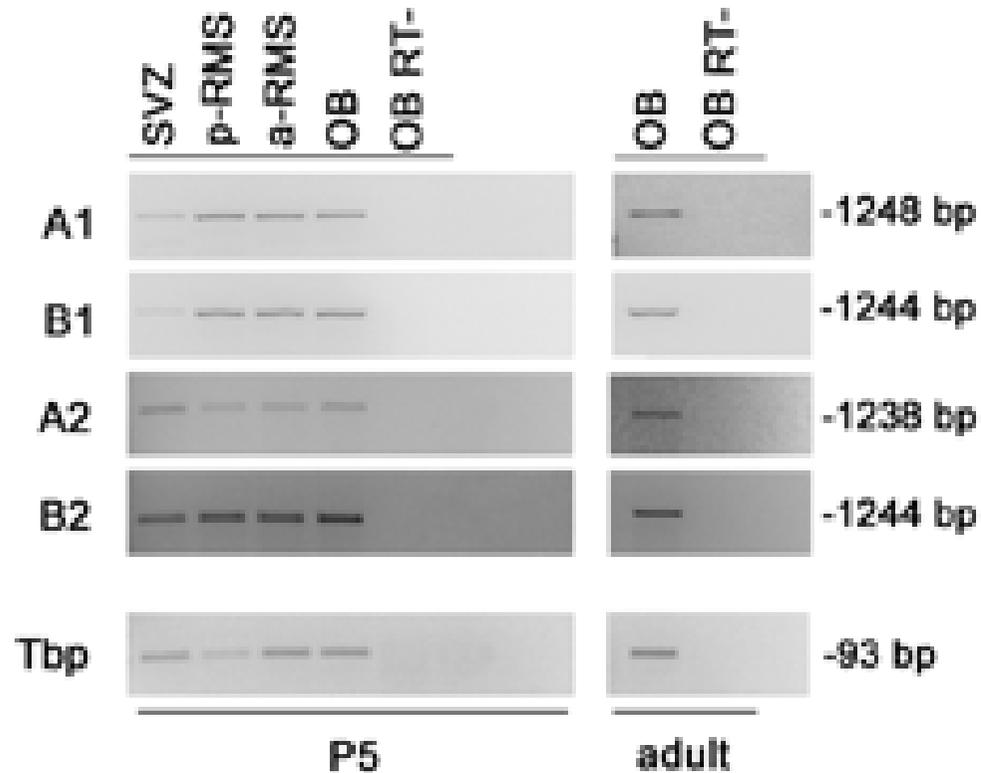
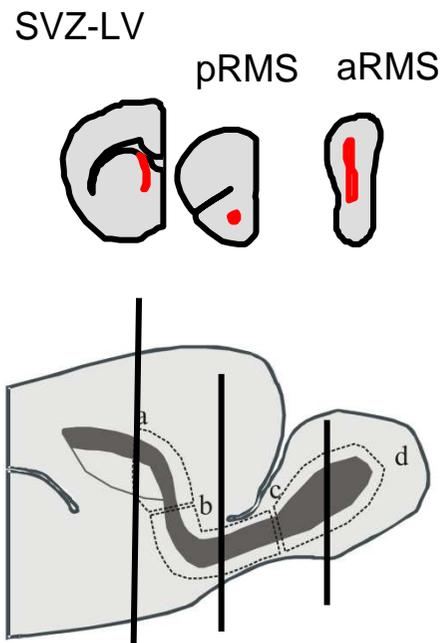
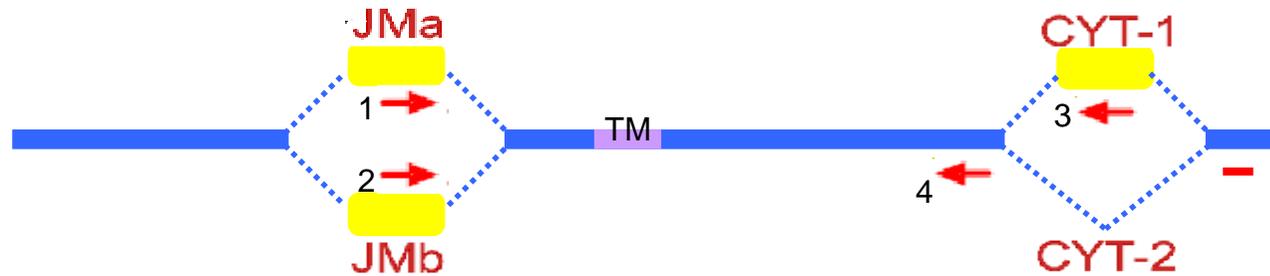
(Ghashghaei *et al.*,2007)

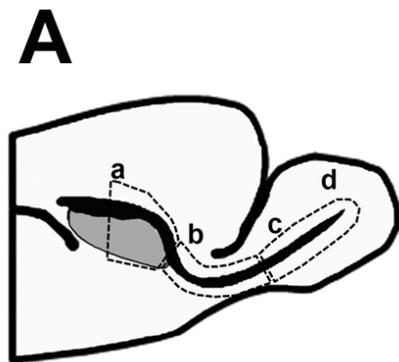
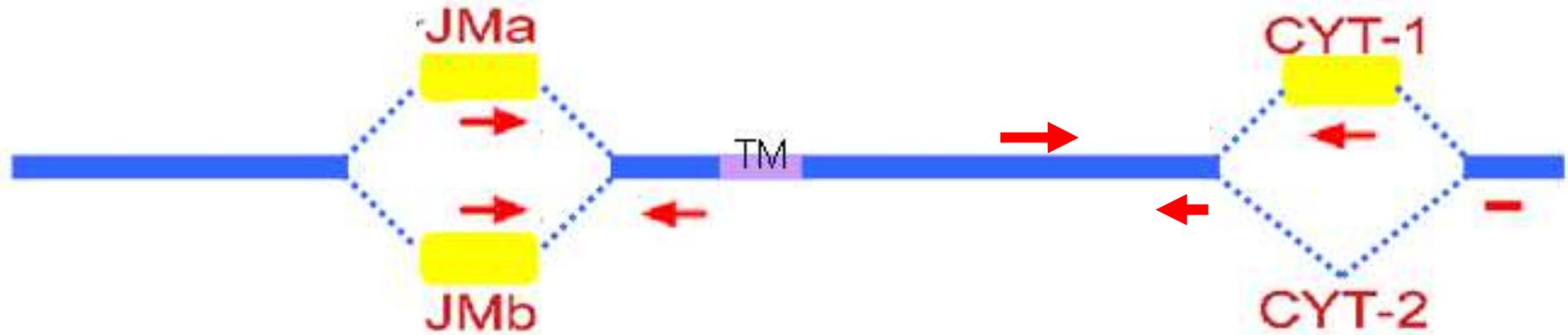


Primer	JMa-Cyt1					JMb-Cyt1					JMa-Cyt2					JMb-Cyt2				
cDNA	A1	B1	A2	B2	H <sub>2</sub> O	A1	B1	A2	B2	H <sub>2</sub> O	A1	B1	A2	B2	H <sub>2</sub> O	A1	B1	A2	B2	H <sub>2</sub> O



# The four ErbB4 isoforms are expressed in the SVZ-OB system





a: SVZ  
 b: pRMS  
 c: aRMS  
 d: sOB

