



ZF-MED

ZEBRAFISH AND OTHER AQUATIC
MODELS IN MEDITERRANEAN LABS



innovation through passion

Giornate studio sull'impiego dei Modelli Acquatici a fini scientifici
Istituto Zooprofilattico Sperimentale dell'Abruzzo e del Molise 'G. Caporale'



15-16 Ottobre 2018

BIOLOGIA MOLECOLARE IN ZEBRAFISH

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UNIVERSITÀ
DEGLI STUDI
DI PALERMO



Dept. of Biological, Chemical and
Pharmaceutical Sciences and Technologies

ATeN Center

Mediterranean Center
for Human Health
Advanced Biotechnologies



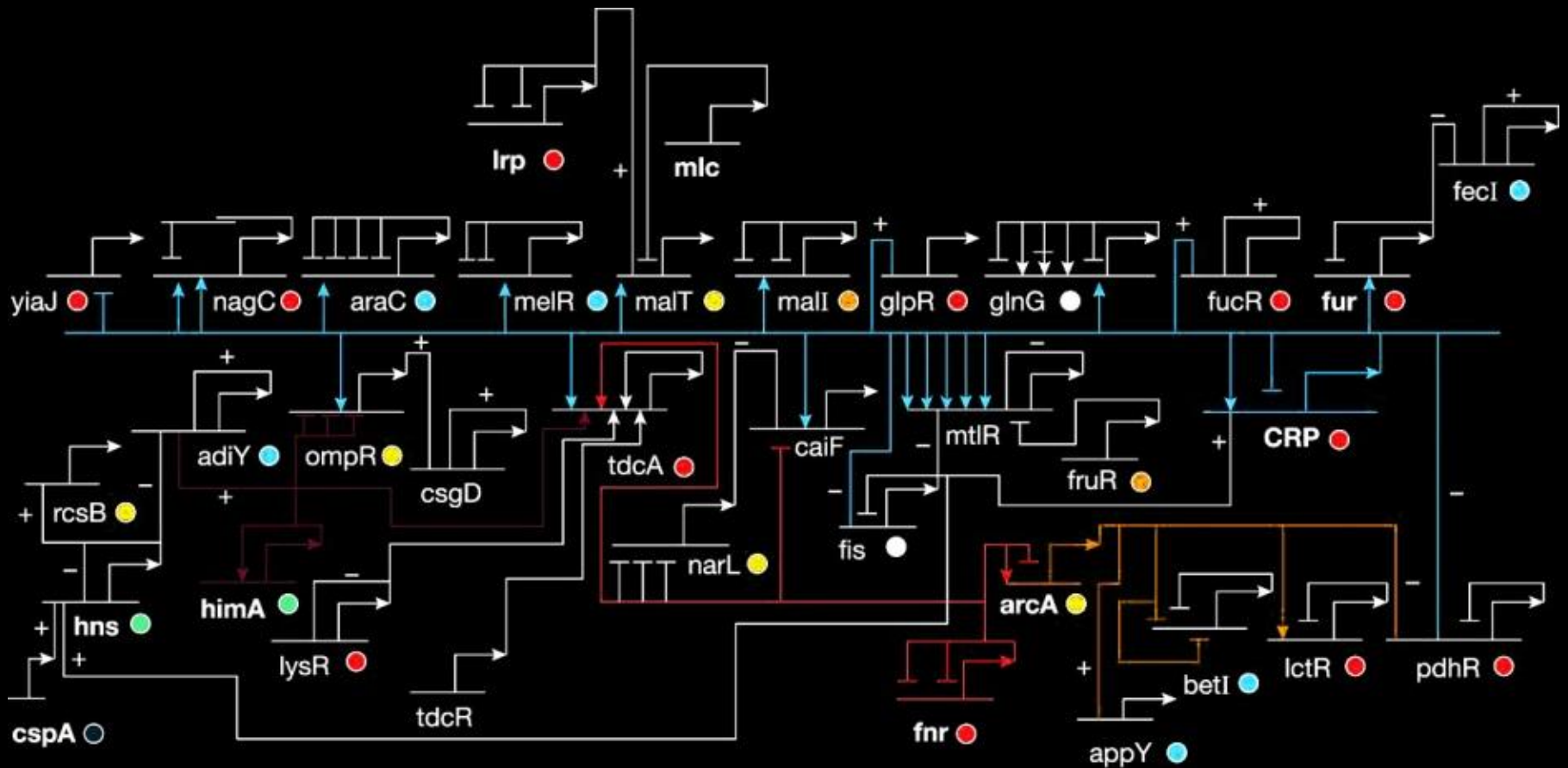
L'Era post-Genomica



Qual'è la funzione di ciascun gene?

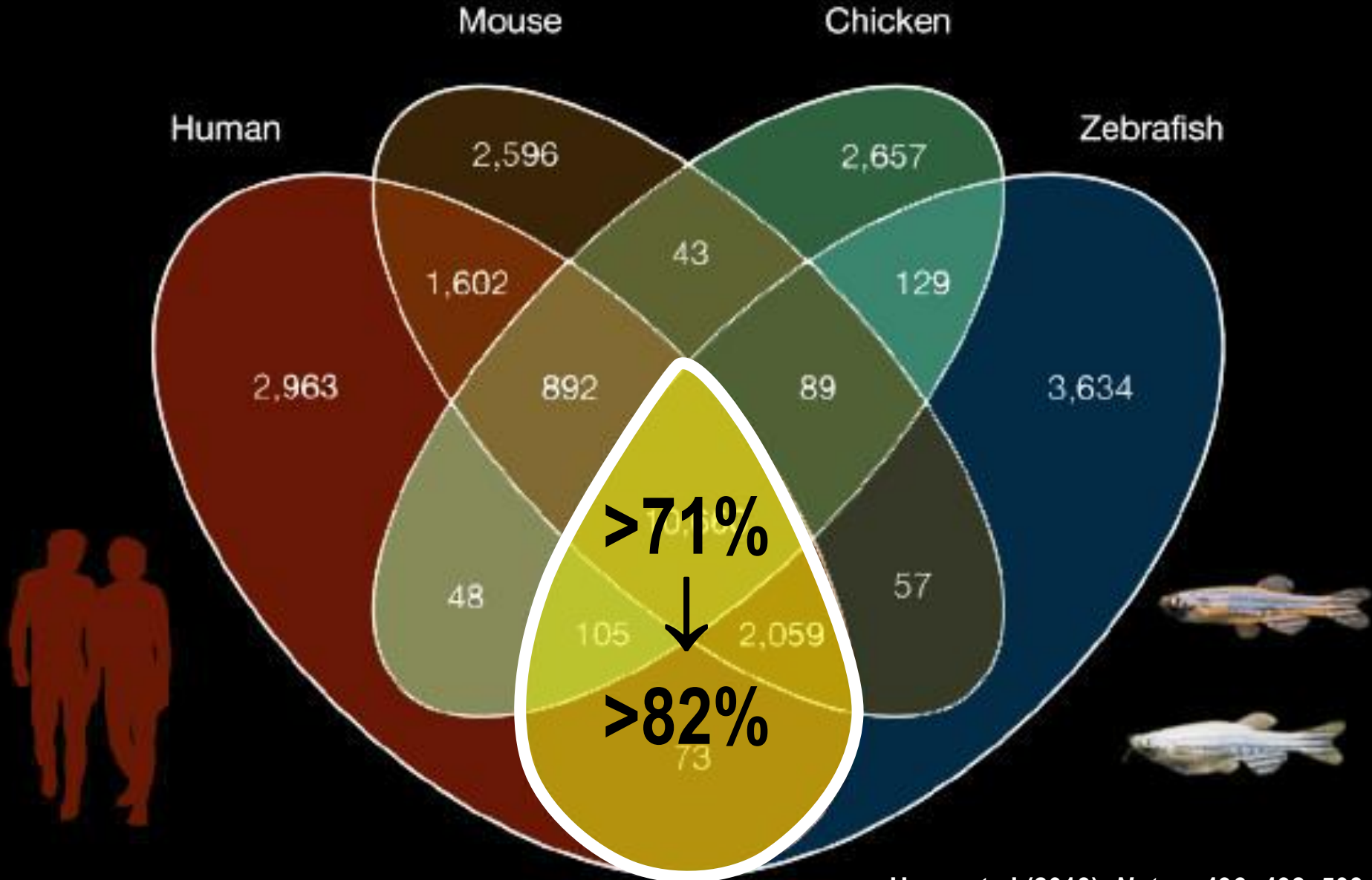
*Coloro che studiano sia i geni che i loro prodotti sono i **Biologi Molecolari***

L'approccio Sistemico



+ componente EPIGENETICA

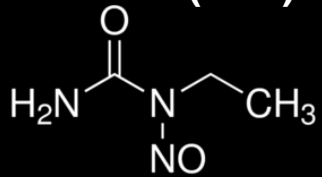
I Modelli Animali: Zebrafish (*Danio rerio*)



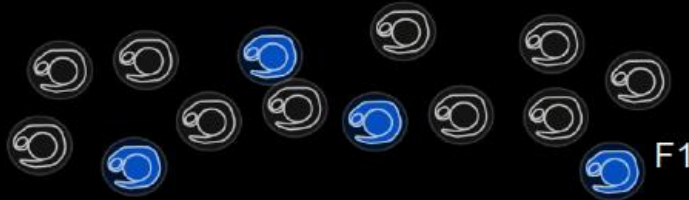
Identificazione della funzione di un gene

APPROCCIO CASUALE

n-ethyl-n-nitrosourea
(ENU)



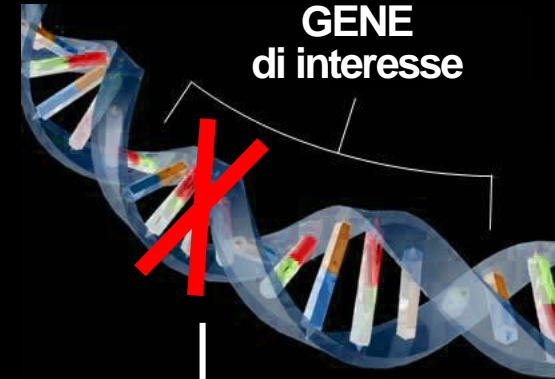
1993-95



FENOTIPO ?

>4,000 mutanti
~400 geni

APPROCCIO MIRATO

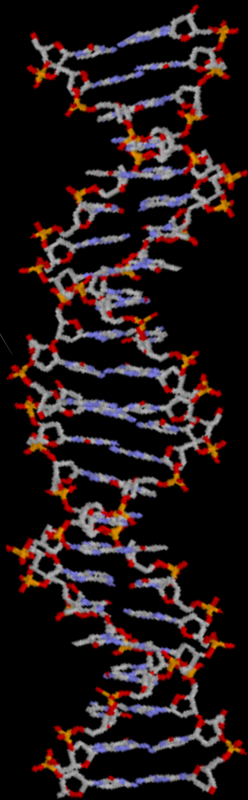


FENOTIPO ?

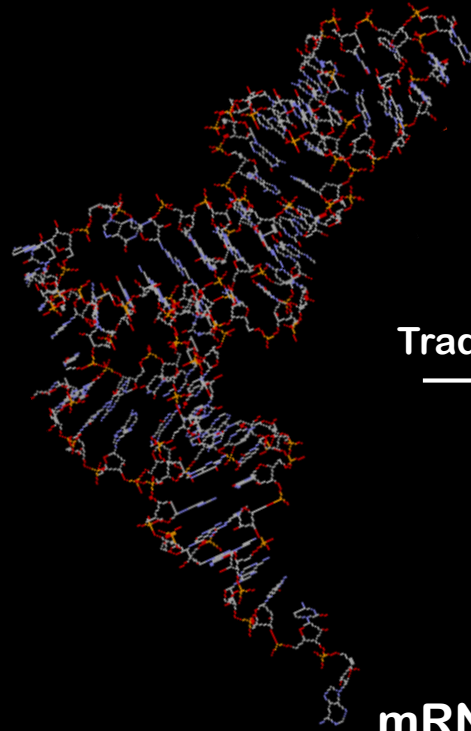
Approccio Mirato

Knock-out

Knock-down



Trascrizione



Traduzione

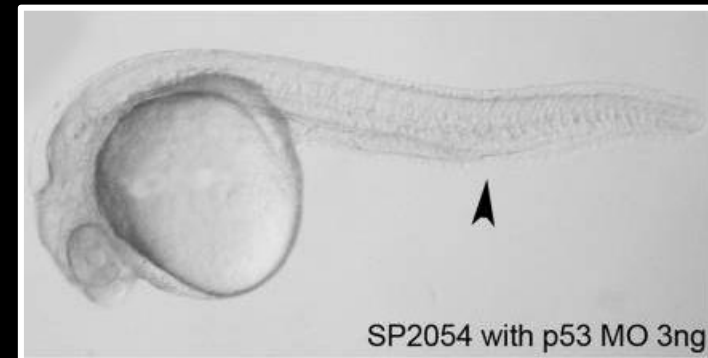
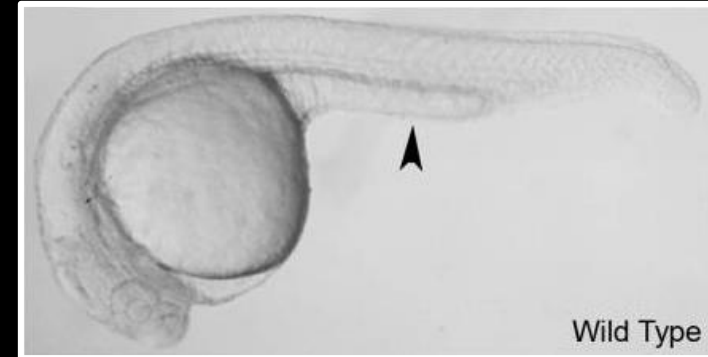
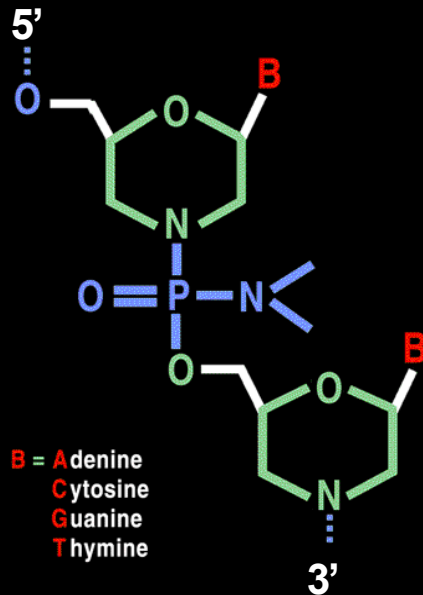
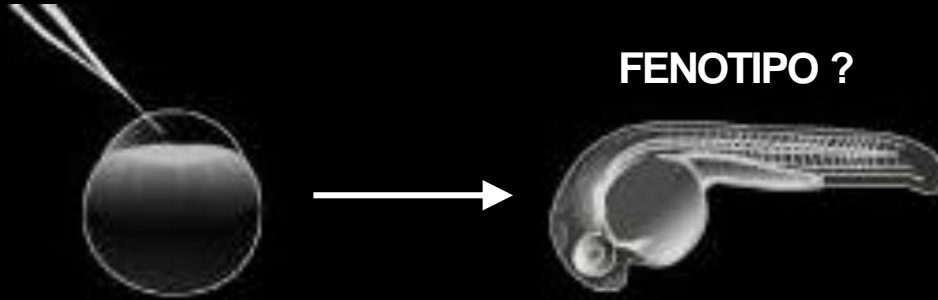


DNA

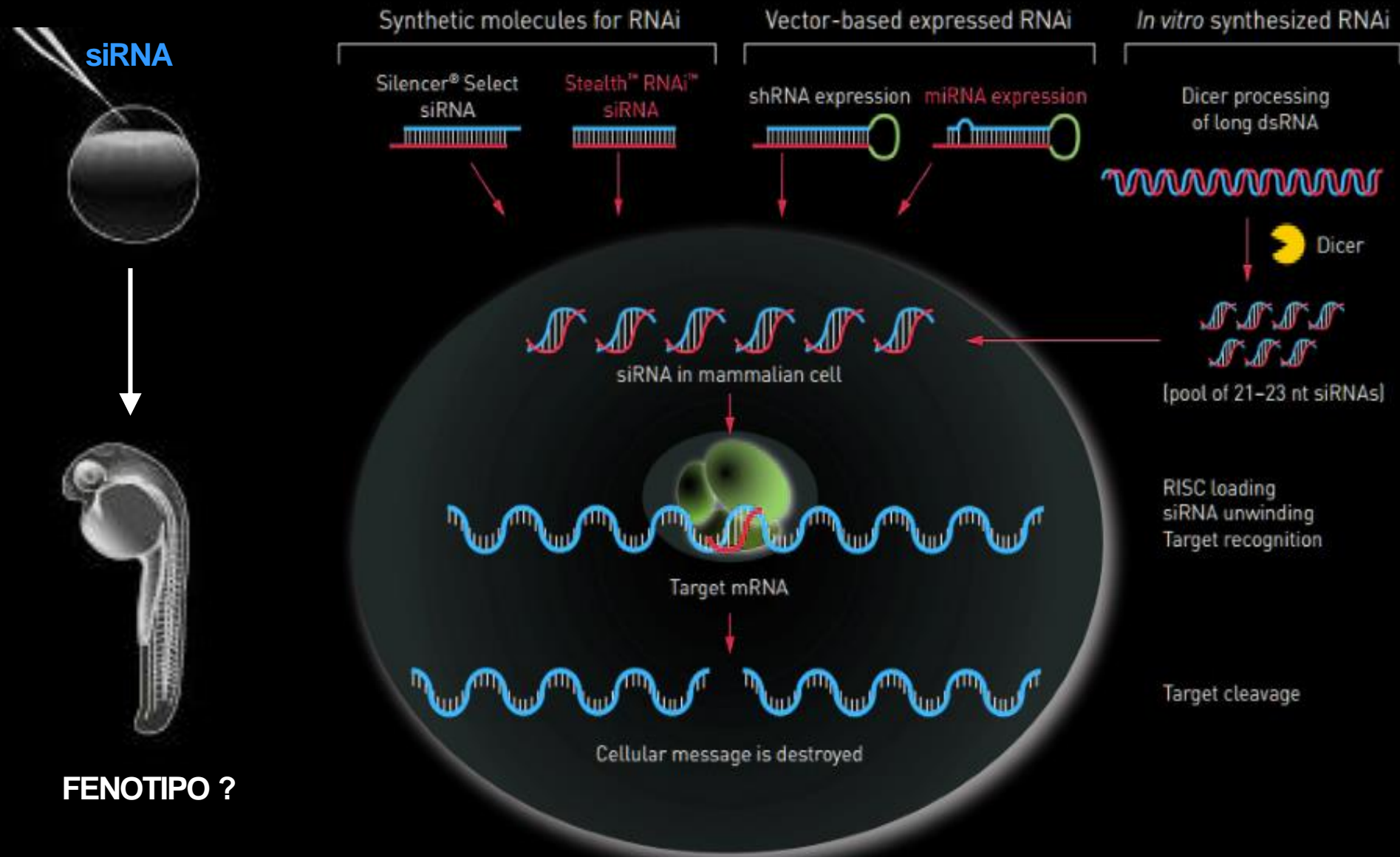
mRNA

Proteina

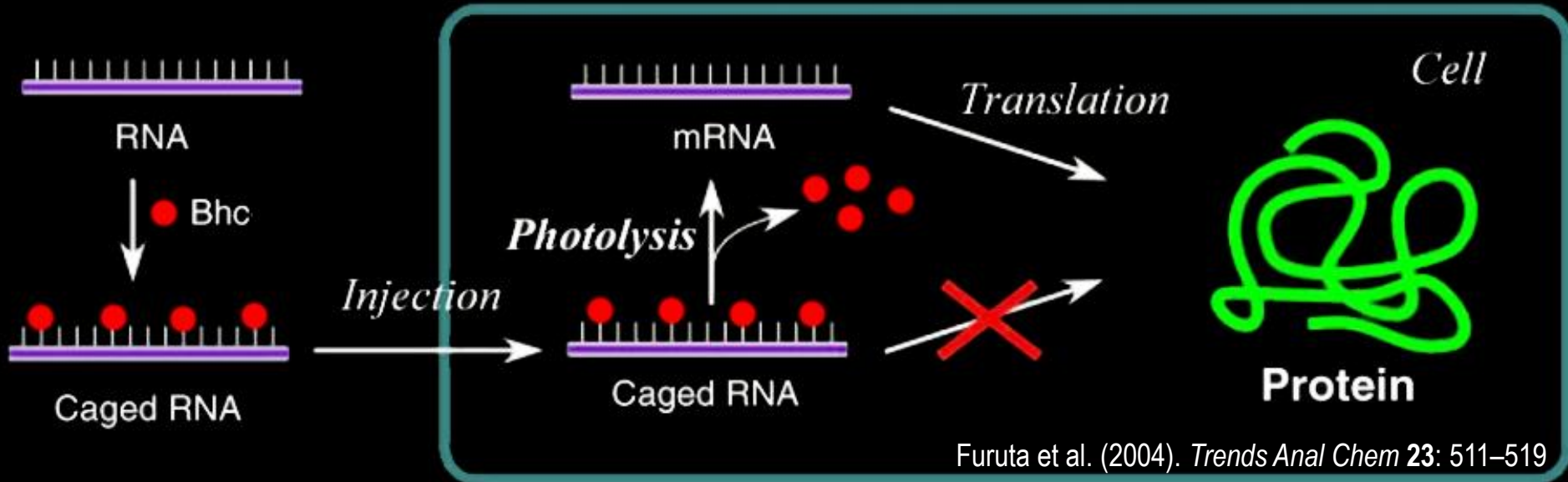
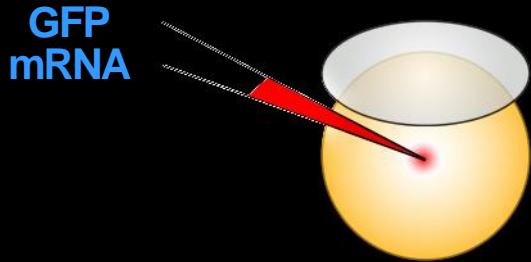
Knock-down funzionale: morpholino



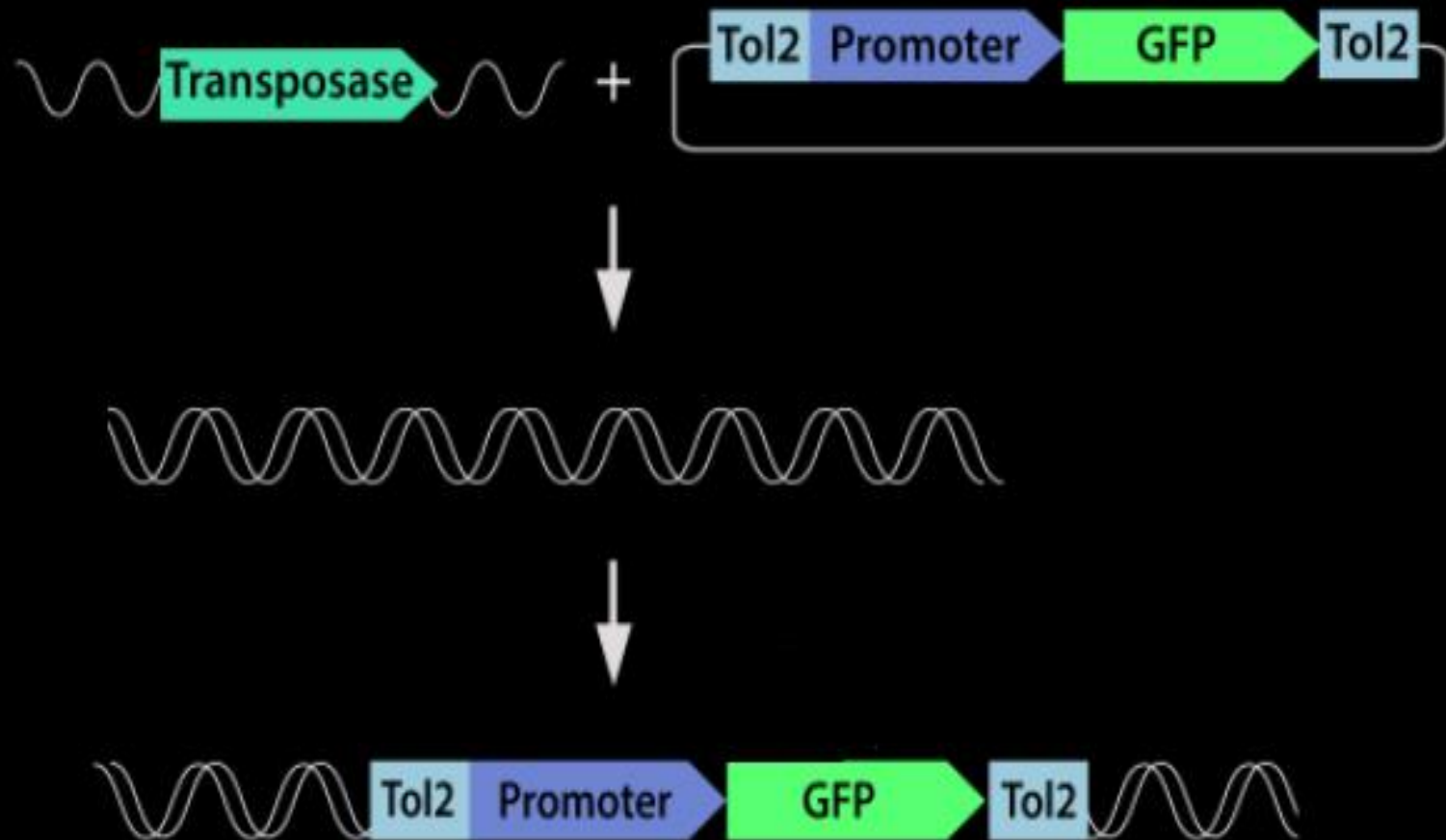
Knock-down funzionale: RNA interference



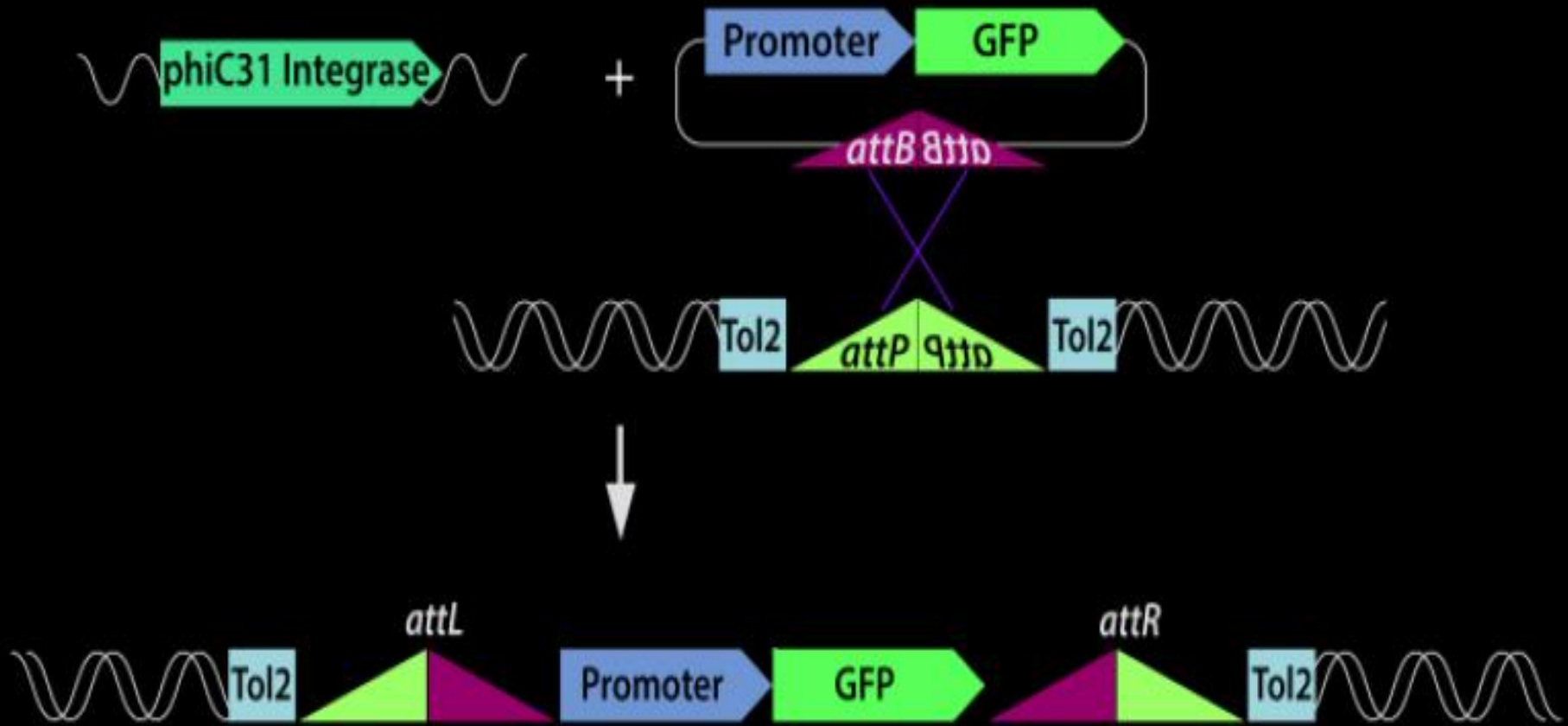
Espressione Ectopica tramite mRNA sintetici



Transgenesi: Trasposizione *Tol2*-dipendente

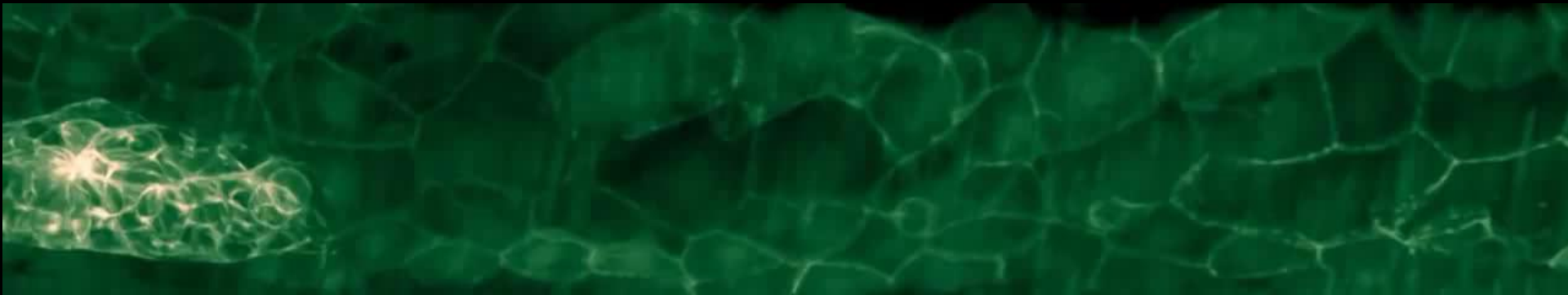
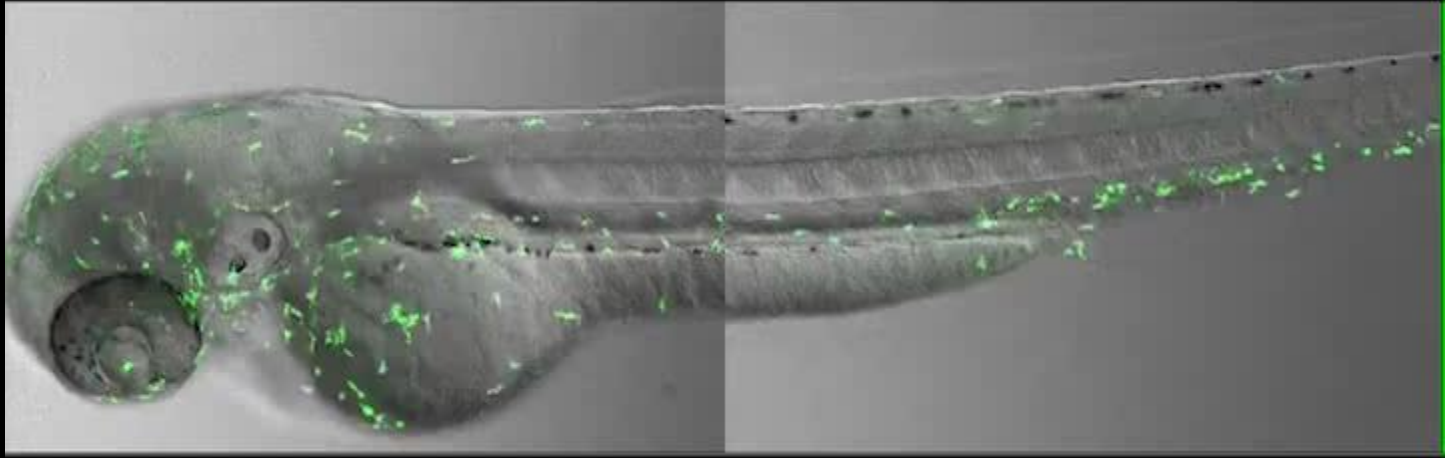


Transgenesis: Ricombinazione ϕ C31-dipendente



Transgenesi: Applicazioni

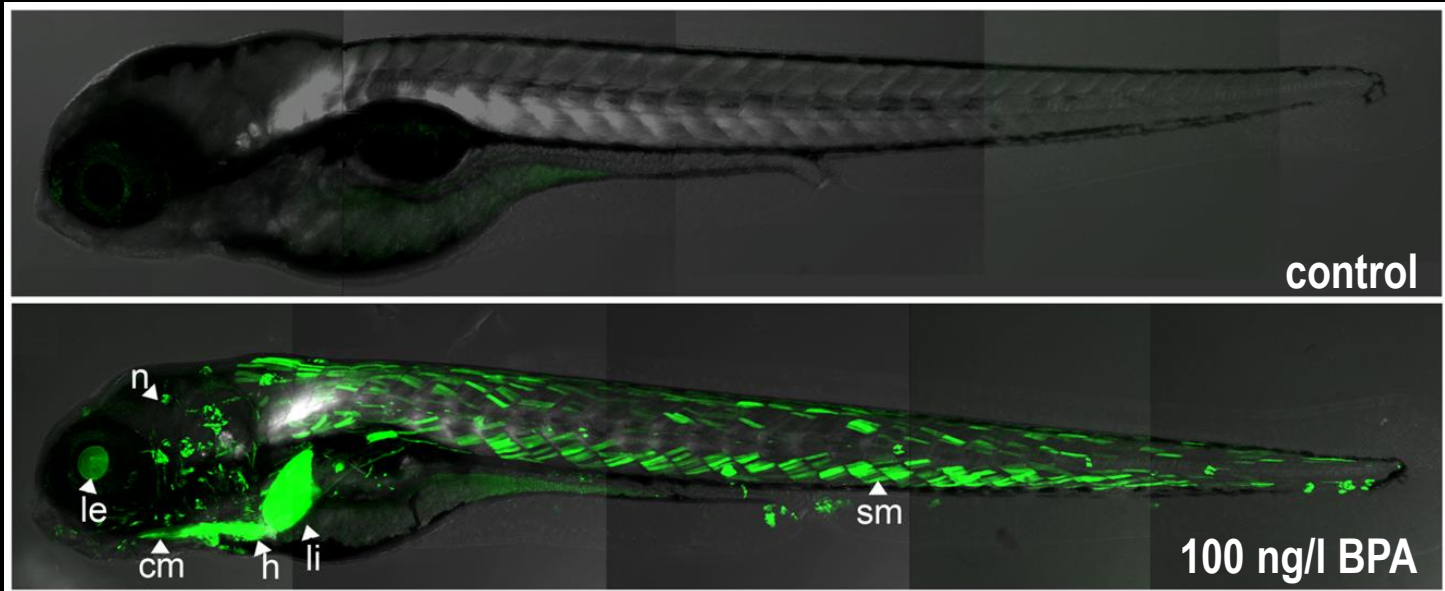
LINEE TRANSGENICHE FLUORESCENTI



Transgenesi: Applicazioni

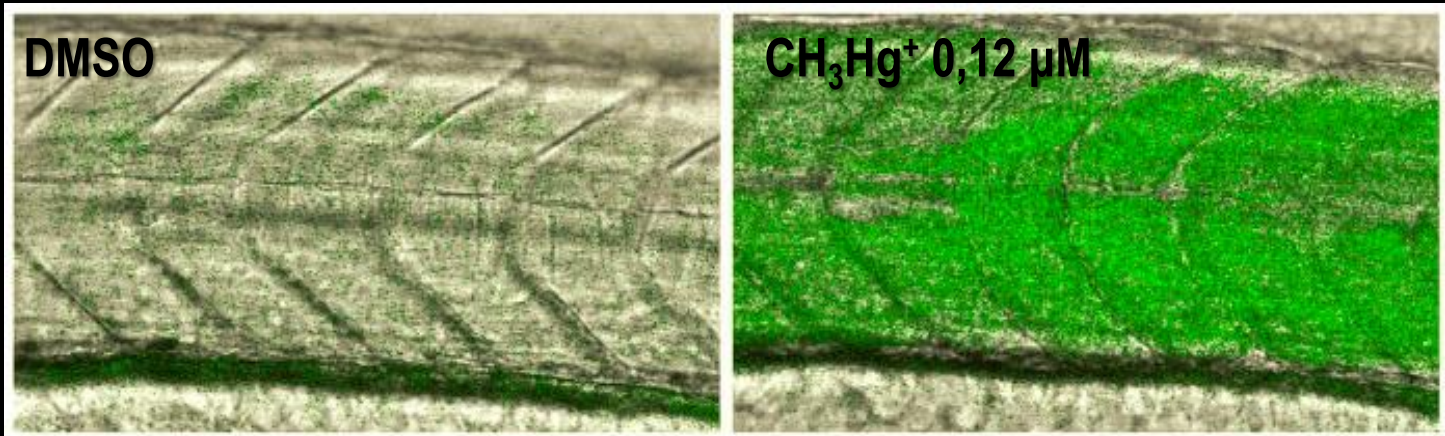
BIOSENSORI

ERE-GFP



Lee et al (2012). *Env Health Perspect* 120:990-6.

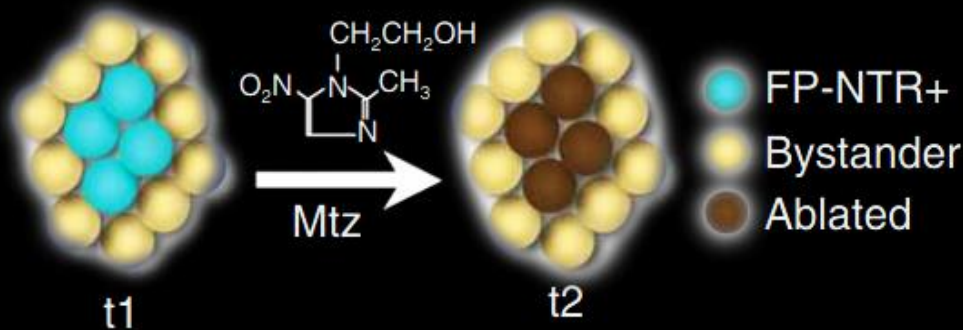
hspb11-GFP



Shadid et al (2016). *Sci Rep* 6:23768.

Transgenesi: Applicazioni

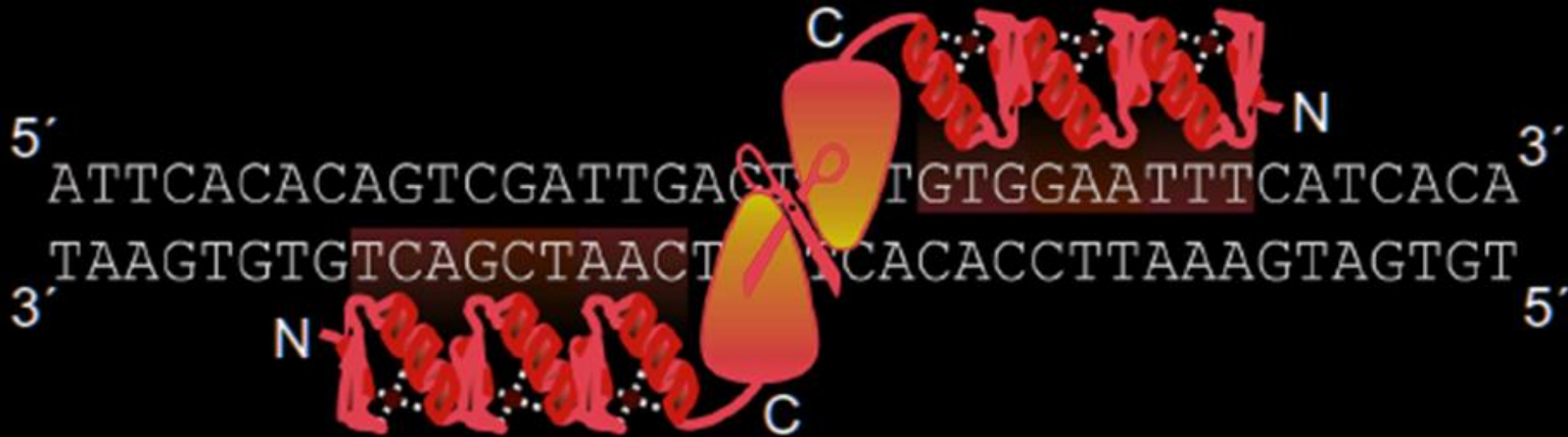
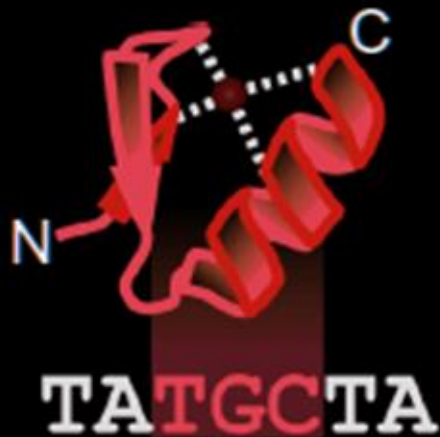
ABLAZIONE MIRATA DI CELLULE



t1: start of ablation

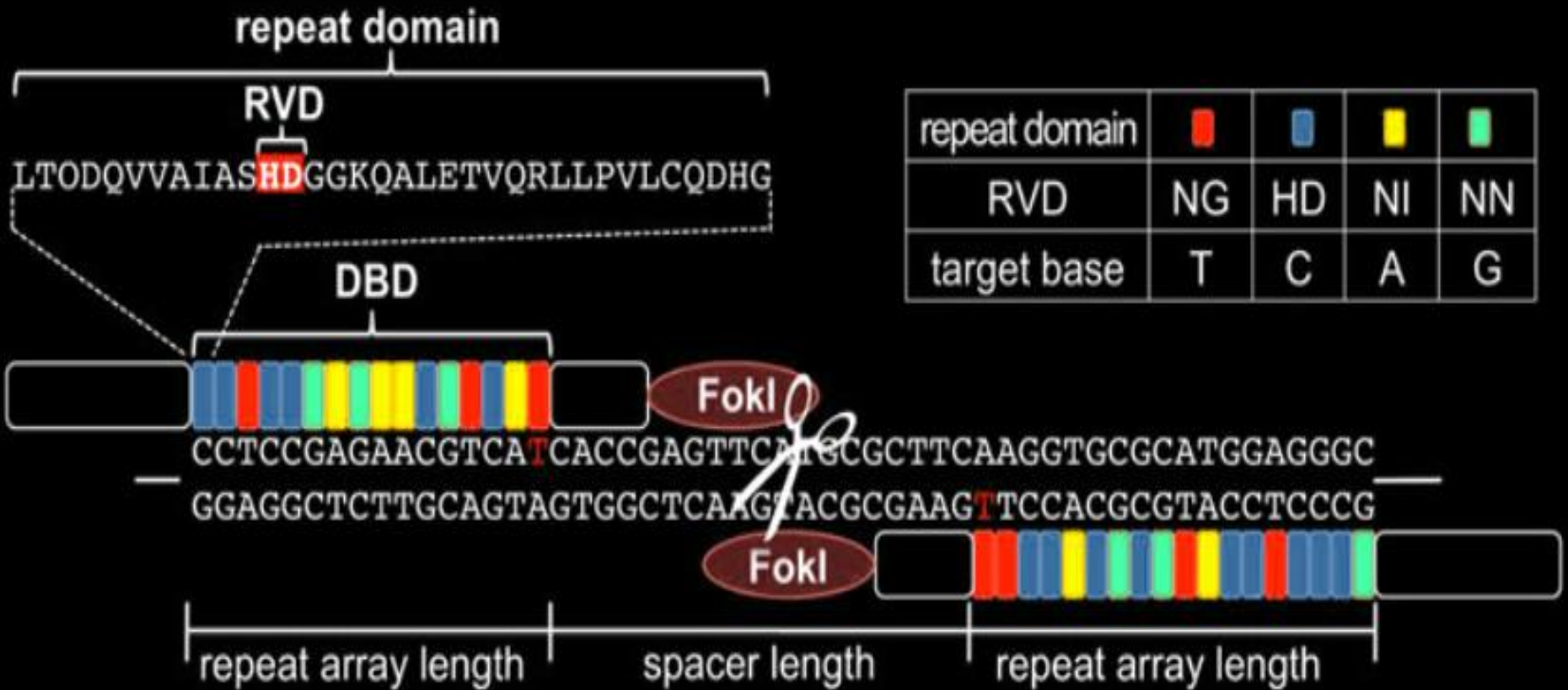
t2: end of ablation

Genome Editing: ZFNs (Zinc-Finger Nucleases)



Genome Editing: TALENs

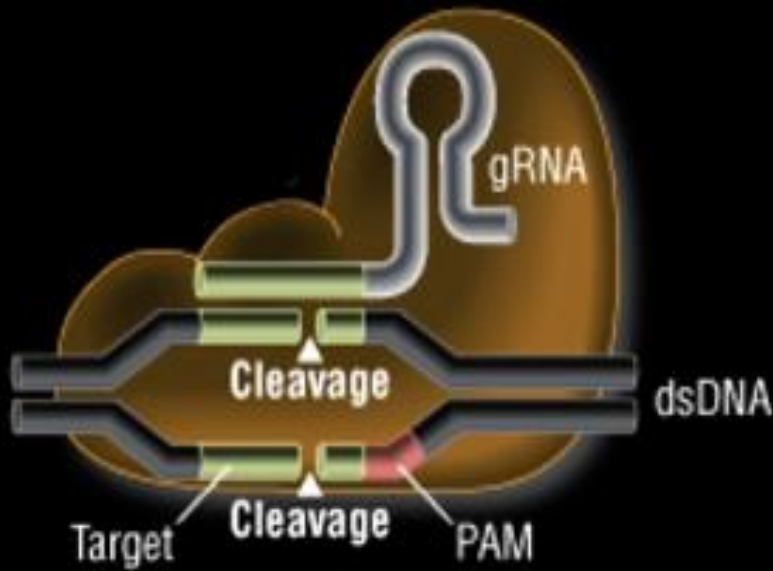
(Transcription Activator-Like Effector Nucleases)



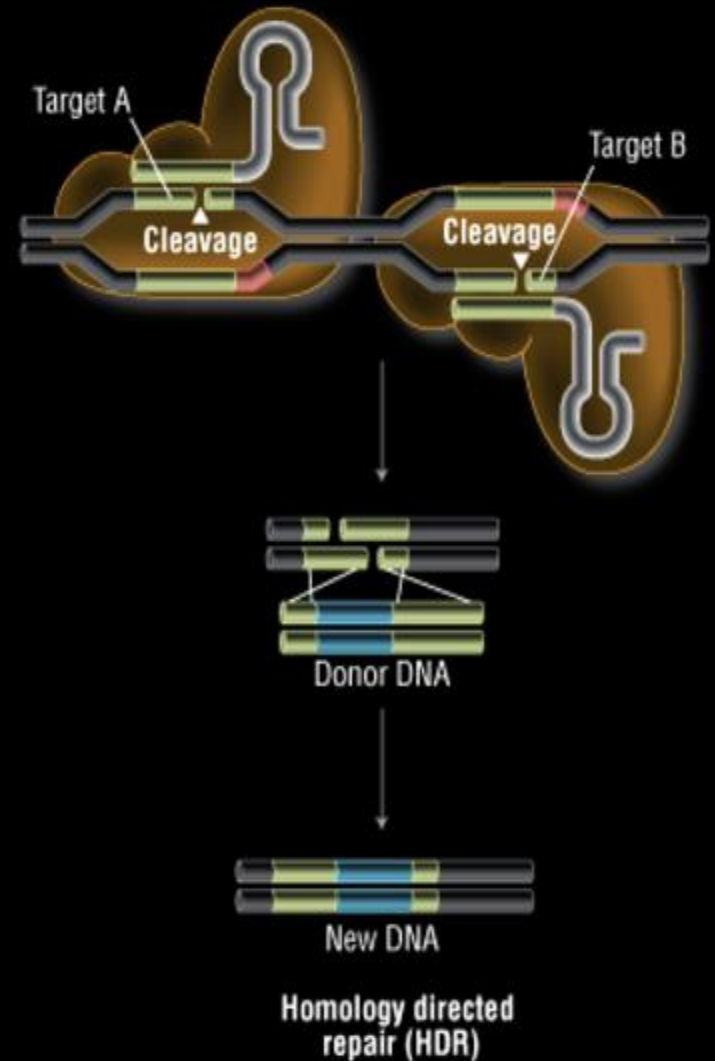
Genome Editing: CRISPR-Cas9

(Clustered Regularly Interspaced Short Palindromic Repeats)

Cas9 wild type



Cas9 D10A





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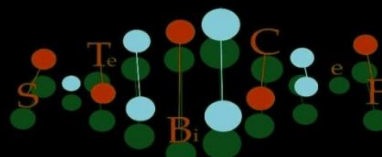
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Thank you

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