# Module 2: Expression Engineering

20.109

Lecture 7

November 1st, 2007

Photo courtesy of Noll Steinweg.

# Sparks of inspiration

Photo courtesy of Children's Hospital Boston. Used with permission.

Judah Folkman

1950s Surgical Resident MGH

"the warmth of tumors made him wonder whether they, like the other tissue in the body, needed their own blood supply and whether they could be starved if that blood supply was cut off"

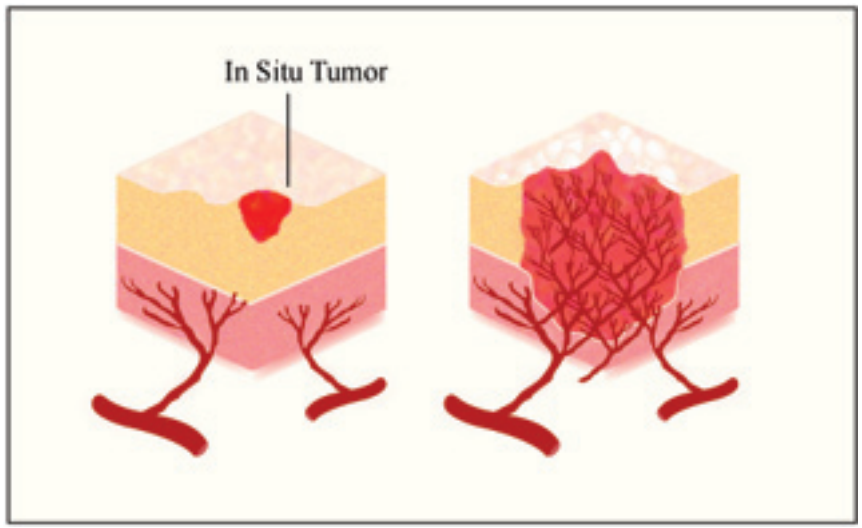
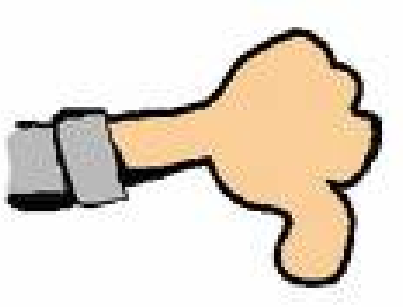



Figure by MIT OpenCourseWare.

# Sparks of inspiration



- existing blood supply enough
- no such molecules known
- tumors red from inflammation

Photo courtesy of Children's Hospital Boston.
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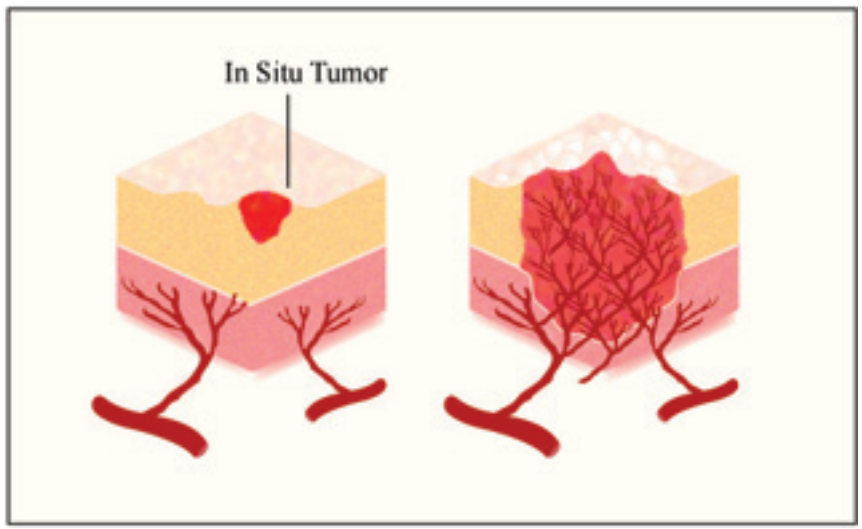



Figure by MIT OpenCourseWare.

# Sparks of inspiration

Photo courtesy of Children's Hospital Boston.
Used with permission.

1: Science. 1976 Jul 2;193(4247):70-2.

**Isolations of a cartilage factor that inhibits tumor neovascularization.**

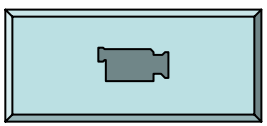
Langer R, Brem H, Falterman K, Klein M, Folkman J.

A cartilage fraction isolated by guanidine extraction and purified by affinity chromatography inhibits tumor-induced vascular proliferation and consequently restricts tumor growth. This fraction contains several different proteins; the major one has a molecular weight of about 16,000. The fraction strongly inhibits protease activity.

rabbit
cornea

Three photos removed due to copyright restrictions.

tumor + polymer + inhibitor -inhibitor



http://www.childrenshospital.org/research/videos/3_Isolating.mov

# Sparks of inspiration

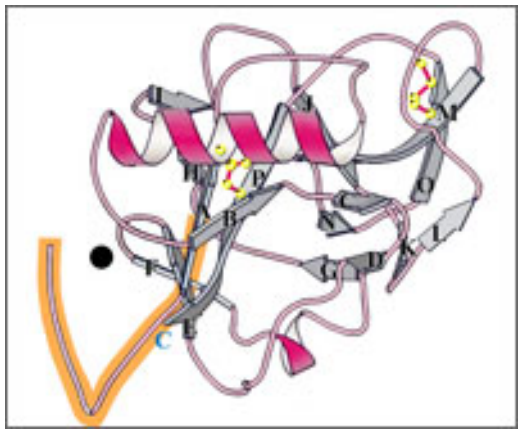


Image courtesy of Children's Hospital Boston.
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## Clinical translation

Photo courtesy of Children's Hospital Boston.
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2004

FDA approval for endostatin

# Sparks of inspiration

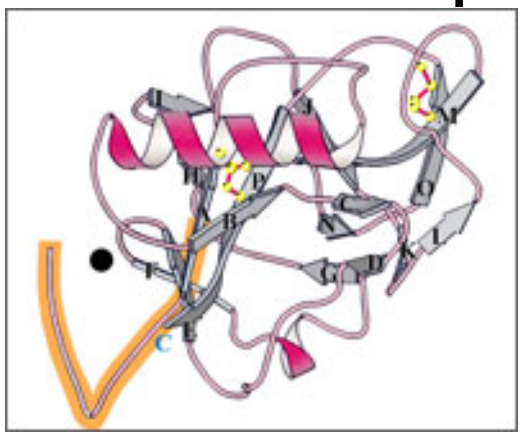

Image courtesy of Children's Hospital Boston.
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Photo courtesy of Children's Hospital Boston.
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## Clinical translation

2004

FDA approval for endostatin

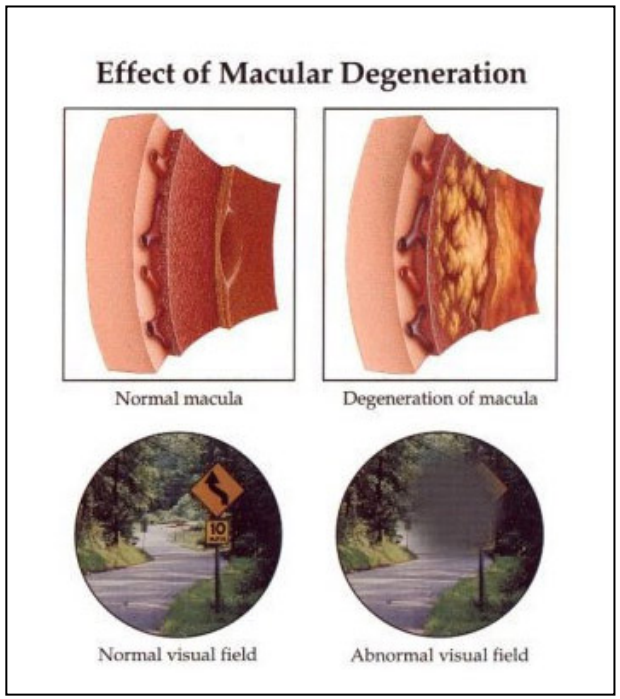
2007

10 anti-angiogenesis treatments approved

32 countries

cancer + macular degeneration

http://www.affymetrix.com/community/wayahead/macular_degeneration.affx




Courtesy of Affymetrix. Used with permission.

# Genome-wide genotype analysis

AMD: Age-related Macular Degeneration

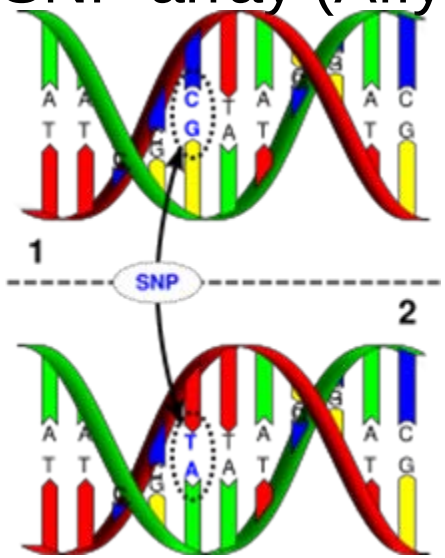
*15 million people in the US suffer from AMD*

*# is expected to double as baby boomers age*

50 unaffected

96 patients

100K SNP array (Affymetrix)




Courtesy of Affymetrix. Used with permission.

# Genome-wide genotype analysis

Both SNPs in gene for Complement Factor H

Figure removed due to copyright restrictions. Figure 1 in Klein, R., et al. “Complement Factor H Polymorphism in Age-Related Macular Degeneration.” Science 308 (2005): 385.

SNP encodes Y402H mutation

Science (2005) 308:419

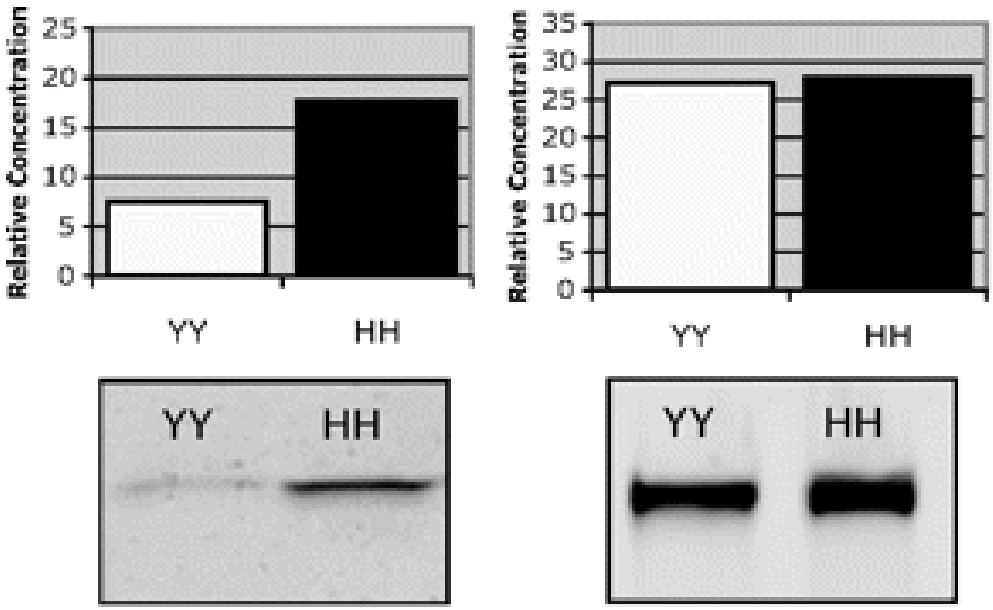
Science (2005) 308:421

Science (2005) 308:385

# Genome-wide genotype analysis

## Western

C-reactive protein (CRP)  Complement Factor H (CFH)






YY= homozygous for Y402

HH = homozygous for Y402H SNP

# Genome-wide phenotype analysis

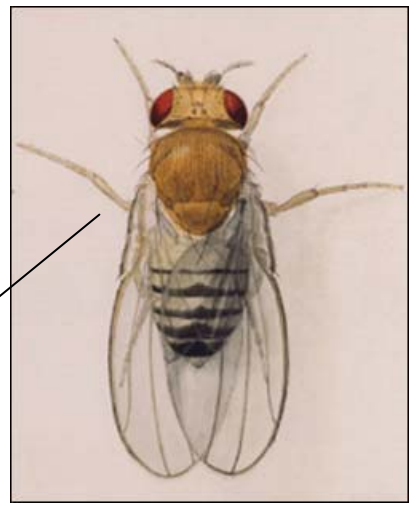
## Arrays for RNAi drug discovery

BROAD INSTITUTE The RNAi Consortium

**"The researchers involved in the three-year, $18 million initiative successfully built a library of 160,000 custom-designed RNAi constructs targeting 15,000 human genes and 15,000 mouse genes. They also developed methods to apply this library effectively for loss-of-function genetic screens. This fundamental resource is available to scientists worldwide through Sigma-Aldrich and Open Biosystems."**

Souce: http://www.broad.mit.edu/genome_bio/trc/ [Accessed June 12, 2008]
Courtesy of the Broad Institute. Used with permission.

grow at RT
ambient CO2
no lipid needed for txn



Watercolor illustration by Thomas Hunt Morgan, 1919.

Features:

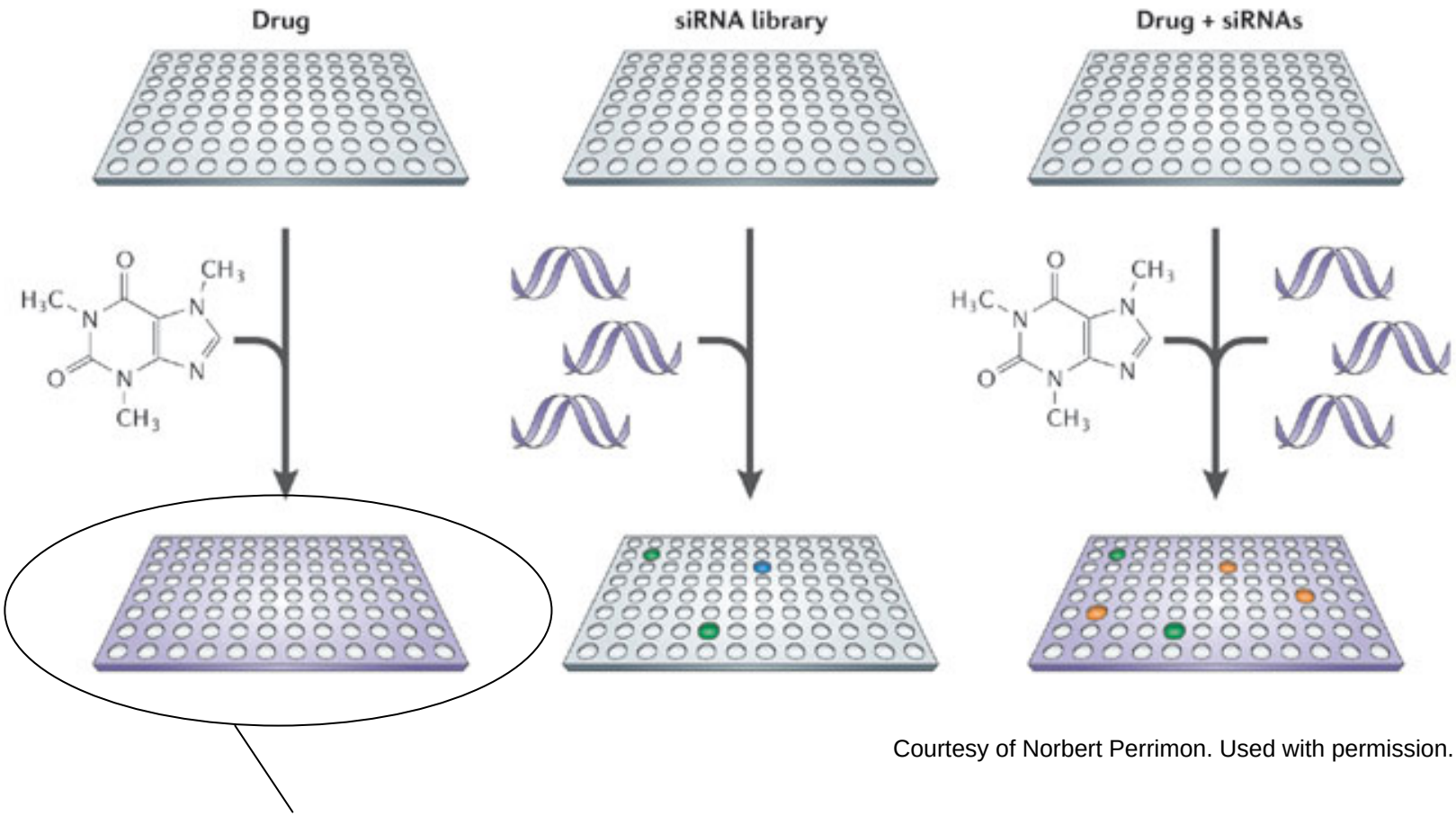
$$$

automated

collaborative

# Genome-wide phenotype analysis

## One possible screening paradigm

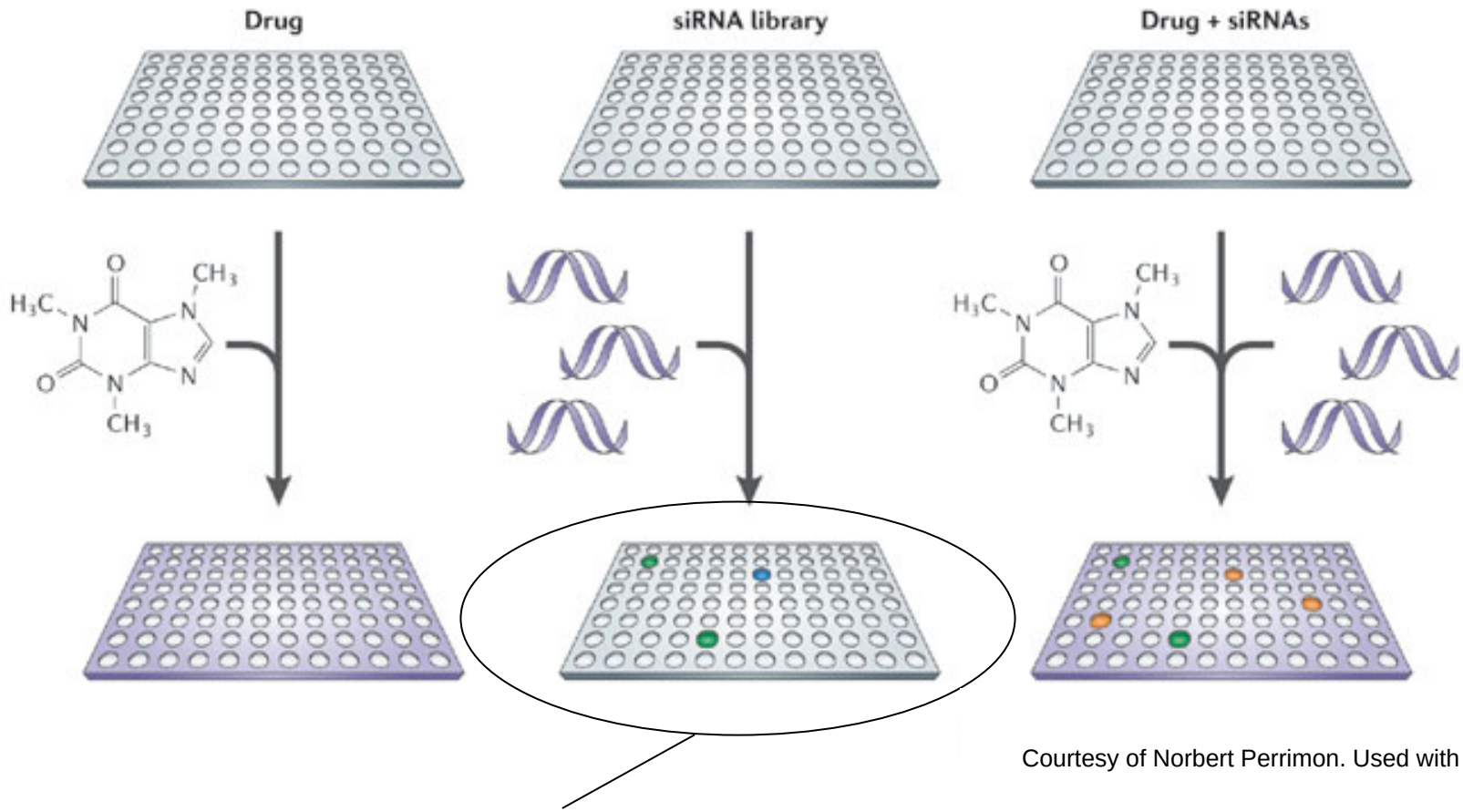



Courtesy of Norbert Perrimon. Used with permission.

Nature Reviews Genetics 2006 7: 373

# Genome-wide phenotype analysis

## One possible screening paradigm

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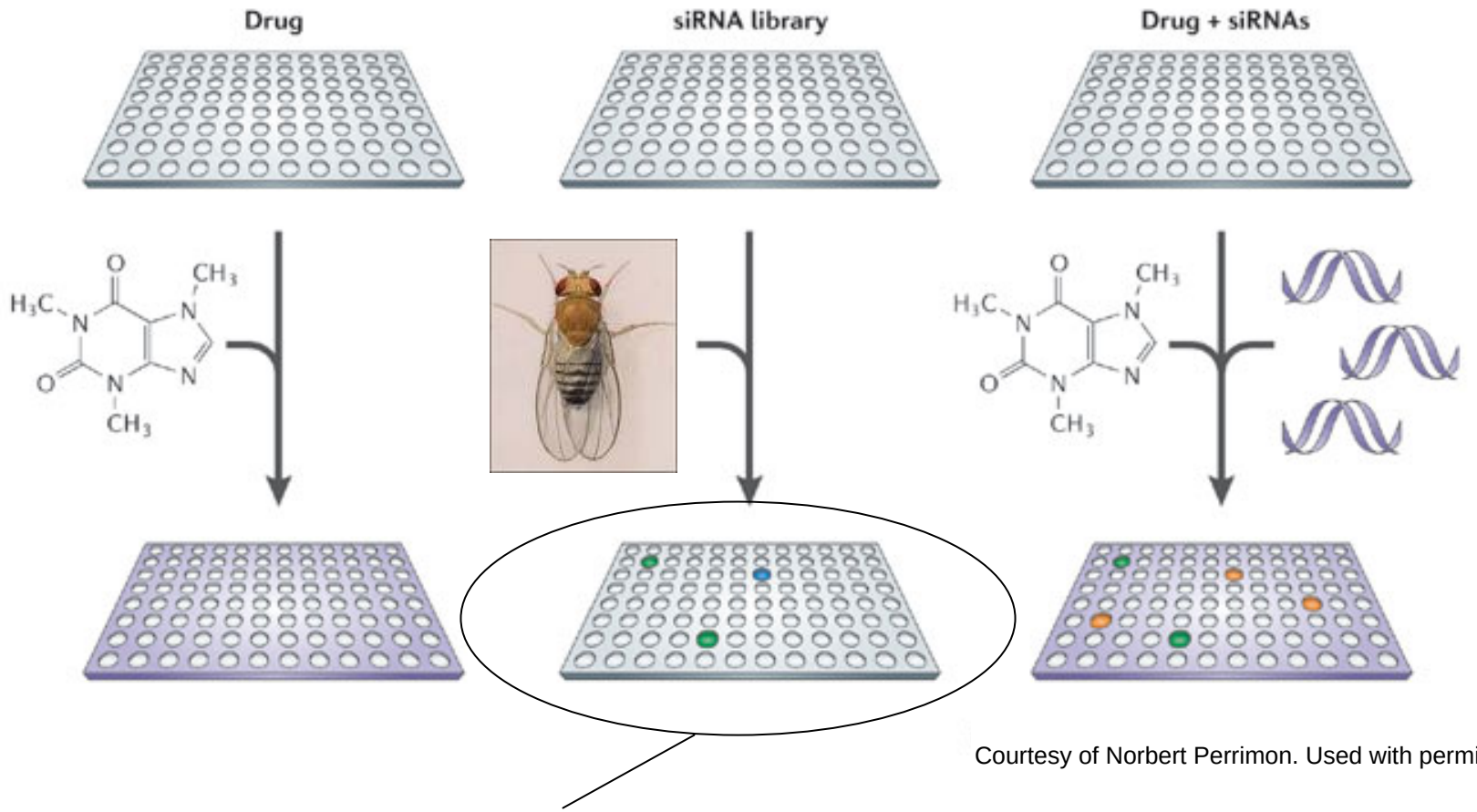
screen to find siRNAs with some effect on cells

Nature Reviews Genetics 2006 7: 373

# Genome-wide phenotype analysis

## One possible screening paradigm




Courtesy of Norbert Perrimon. Used with permission.

screen to find siRNAs with some effect on cells

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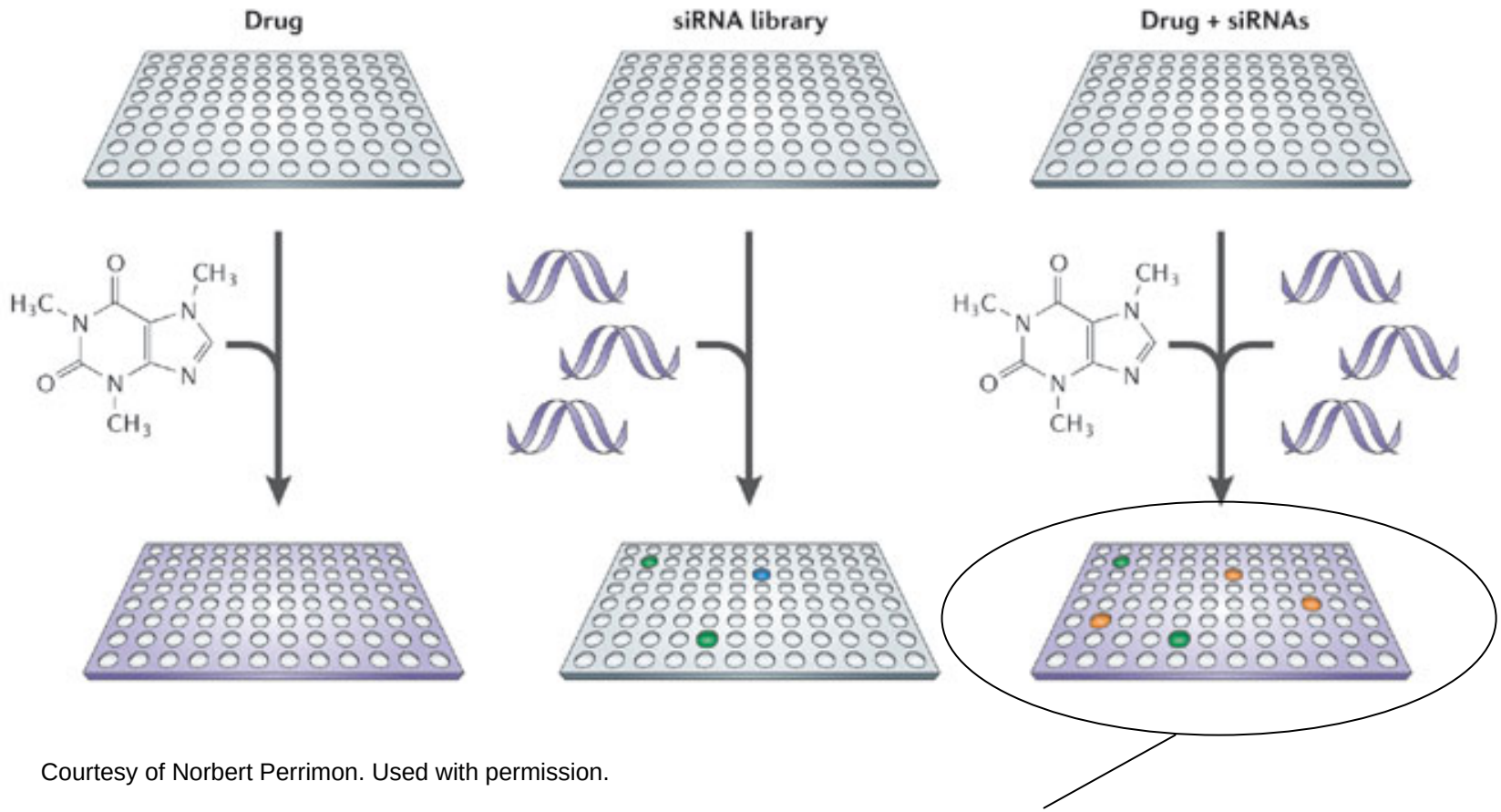
# Genome-wide phenotype analysis

## One possible screening paradigm



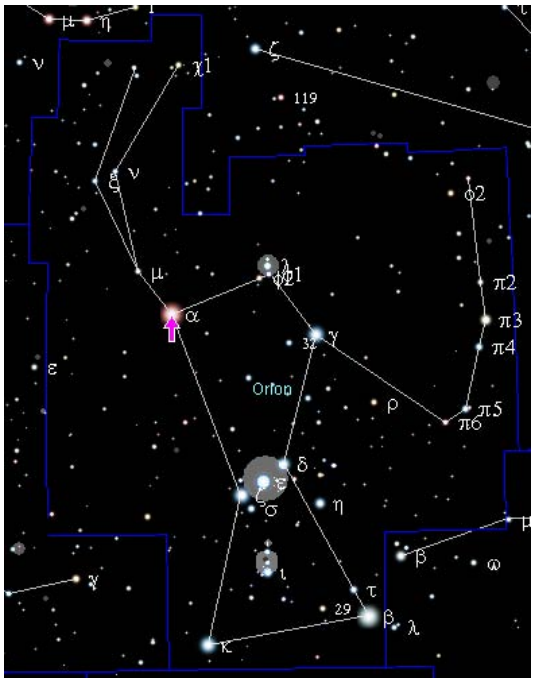

Courtesy of Norbert Perrimon. Used with permission.

screen for synthetic effects/biomarkers of drug with siRNA

Nature Reviews Genetics 2006 7: 373

# Genome-wide phenotype analysis

## Alnylam Pharmaceuticals




Constellation “Orion the Hunter”
Source: Wikipedia

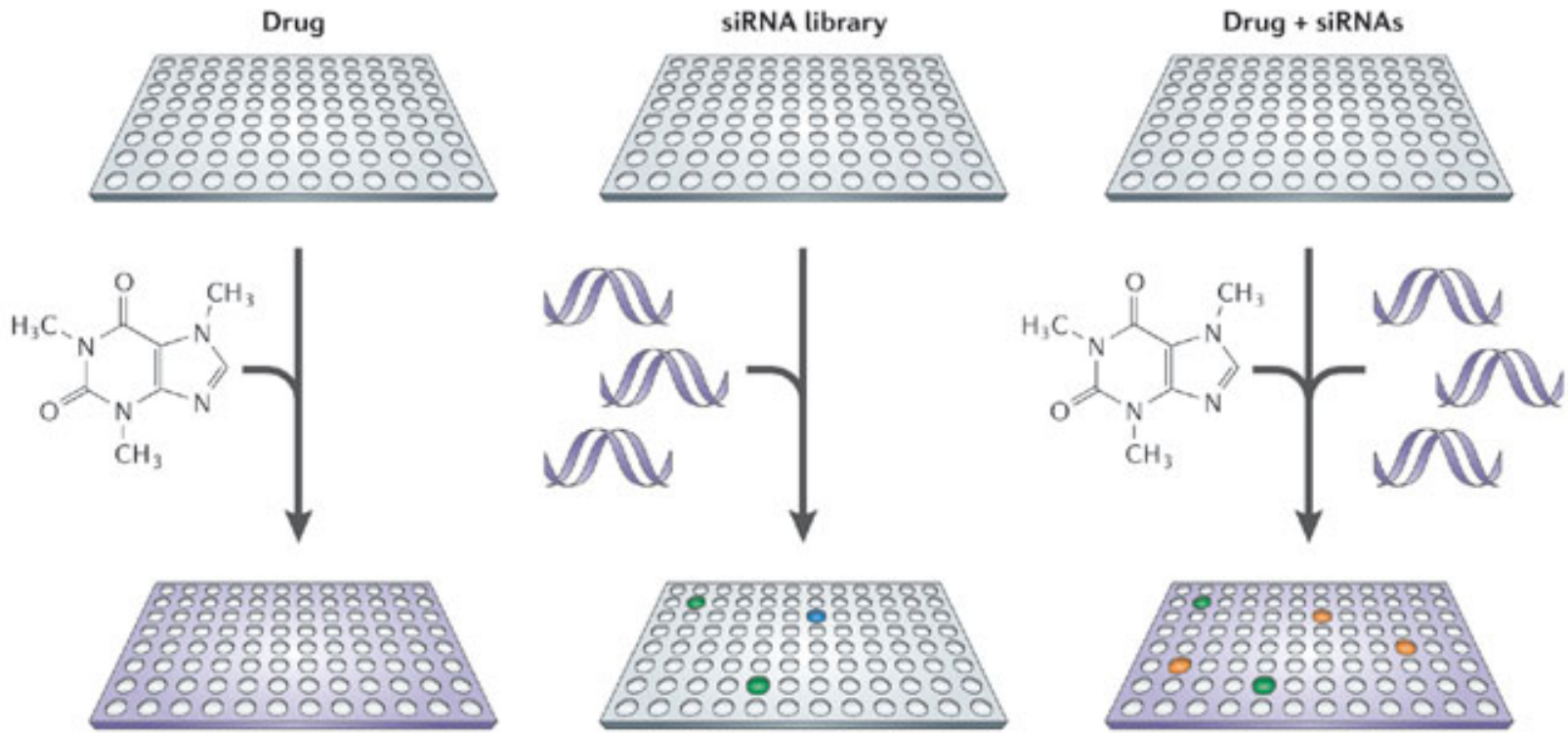
## Respiratory Syncytial Virus (RSV)

- ssRNA enveloped virus of the Paramyxovirus family
- Responsible for 50-90% of Bronchiolitis and 5-40% of Bronchopneumonia in infants
- RSV fusion protein allows virus to fuse with host cells, and then fuses neighboring cells into multinucleated masses

RSV pathology photo removed due to copyright restrictions.
See http://web.uct.ac.za/depts/mmi/stannard/syncytia.html.

# Genome-wide phenotype analysis

## One possible screening paradigm

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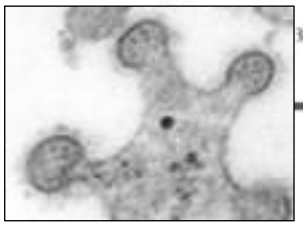
# Genome-wide phenotype analysis

## One possible screening paradigm

RSV



Courtesy of CDC.




Courtesy of Norbert Perrimon. Used with permission.

Alnylam PHARMACEUTICALS

"ALN-RSV01"

in Phase II clinical trials

RSV pathology photo removed due to copyright restrictions.
See http://web.uct.ac.za/depts/mmi/stannard/syncytia.html.

Photo courtesy of Noll Steinweg.

# Summary

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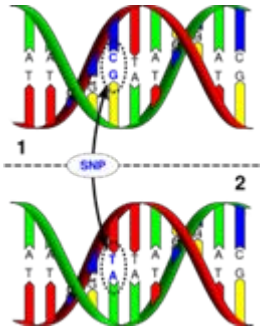
## Arrays for

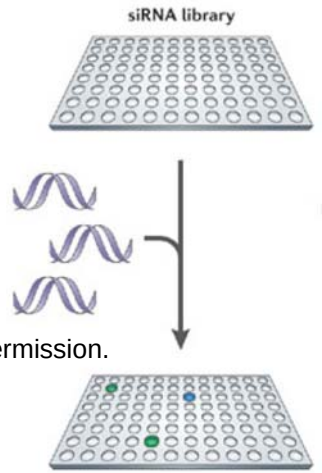
## 1. Genotyping: SNP ID --> AMD




Courtesy of Affymetrix. Used with permission.

Graph removed due to copyright restrictions. Figure 1 in Klein, R., et al. "Complement Factor H Polymorphism in Age-Related Macular Degeneration." Science 308 (2005): 385.

## 2. Phenotyping: TRC -->RSVs




Courtesy of Norbert Perrimon. Used with permission.

RSV pathology photo removed due to copyright restrictions. See http://web.uct.ac.za/depts/mmi/stannard/syncytia.html.