CSE 521: Algorithms

W. L. Ruzzo

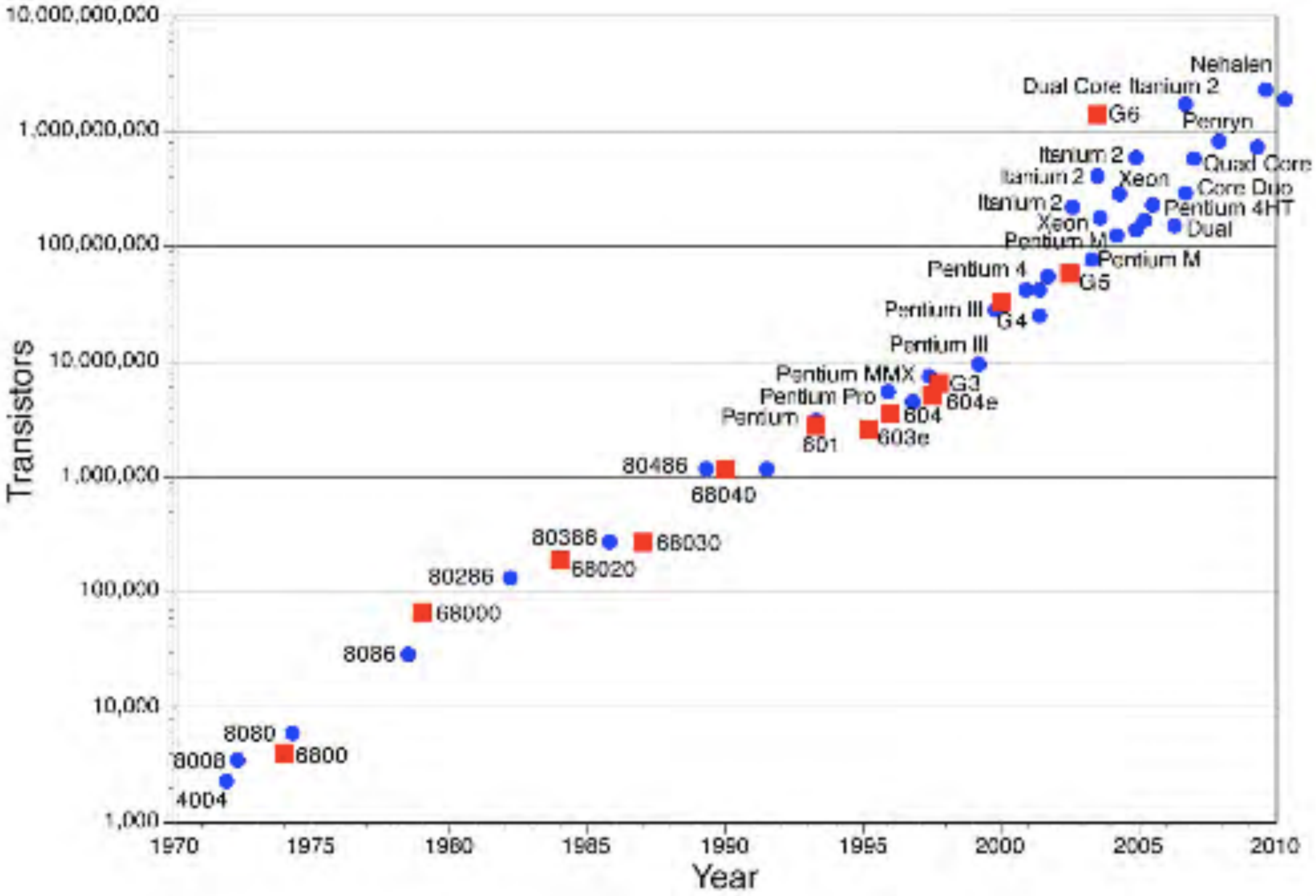
Dynamic Programming:
String alignment and RNA Folding

Outline

- A few slides on *applications* of dynamic programming in biology
    - Sequence alignment
    - RNA structure
- Algorithms for RNA structure

Application: Sequence Search

Moore's Law



10,000,000,000
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Transistors
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Quad Core
Itanium 2
Xeon
Core Duo
Itanium 2
Pentium 4HT
Xeon
Dual
Pentium M
Pentium M
Pentium 4
G5
Pentium III
G4
Pentium III
Pentium MMX
G3
Pentium Pro
604e
Pentium
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603e
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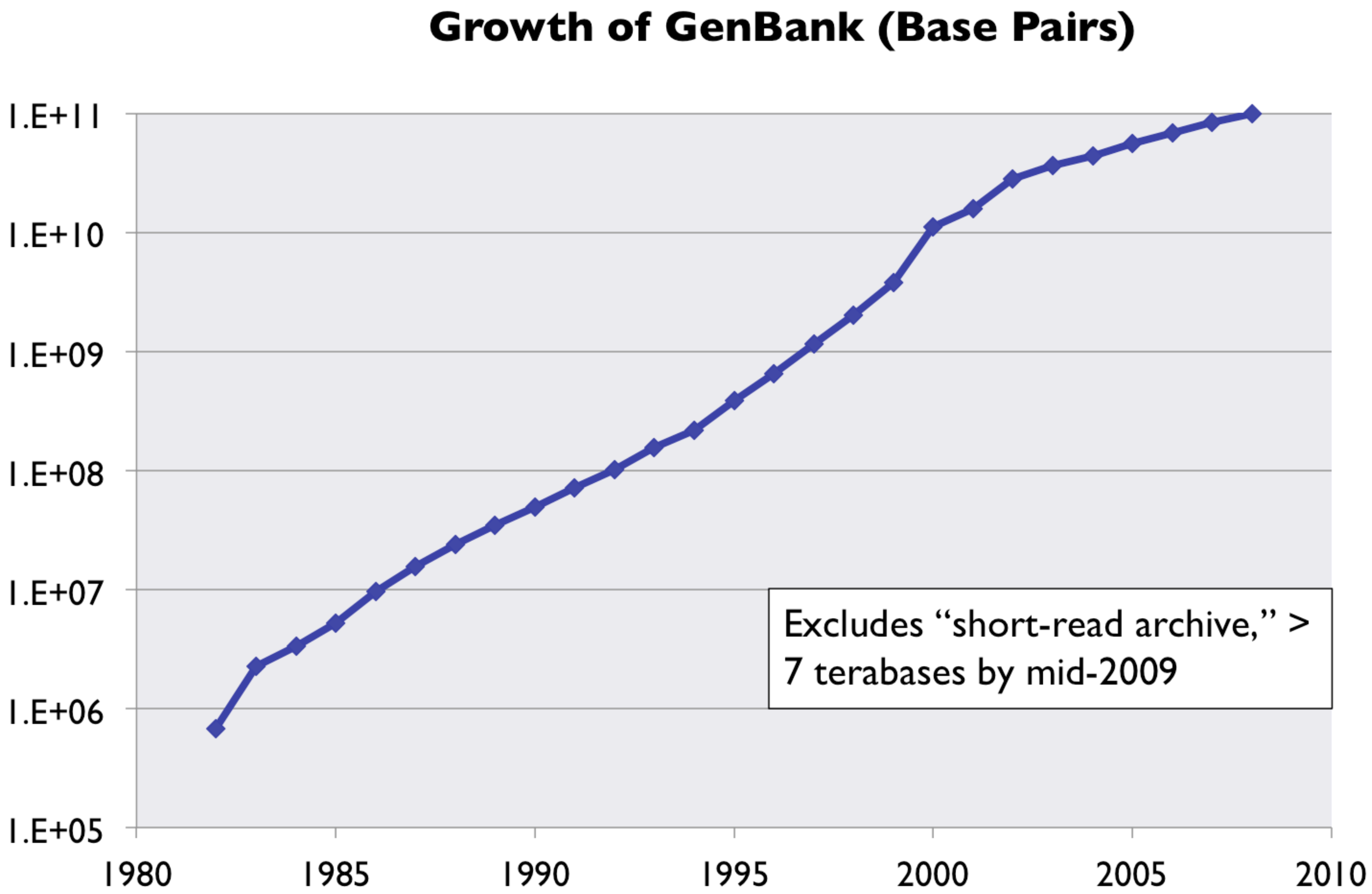
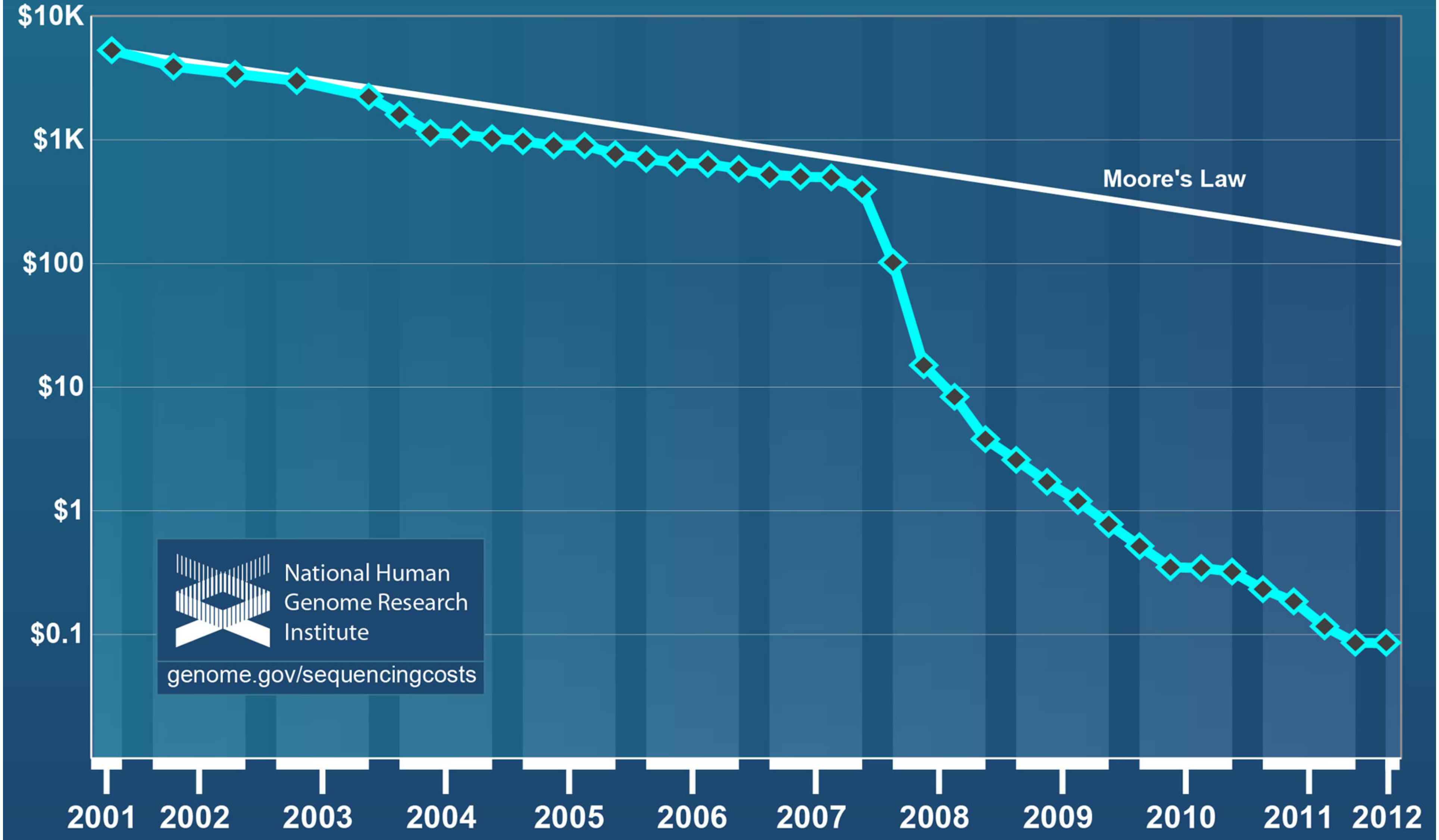


Excludes “short-read archive,” > 7 terabases by mid-2009

Source: http://www.ncbi.nlm.nih.gov/Genbank/genbankstats.html

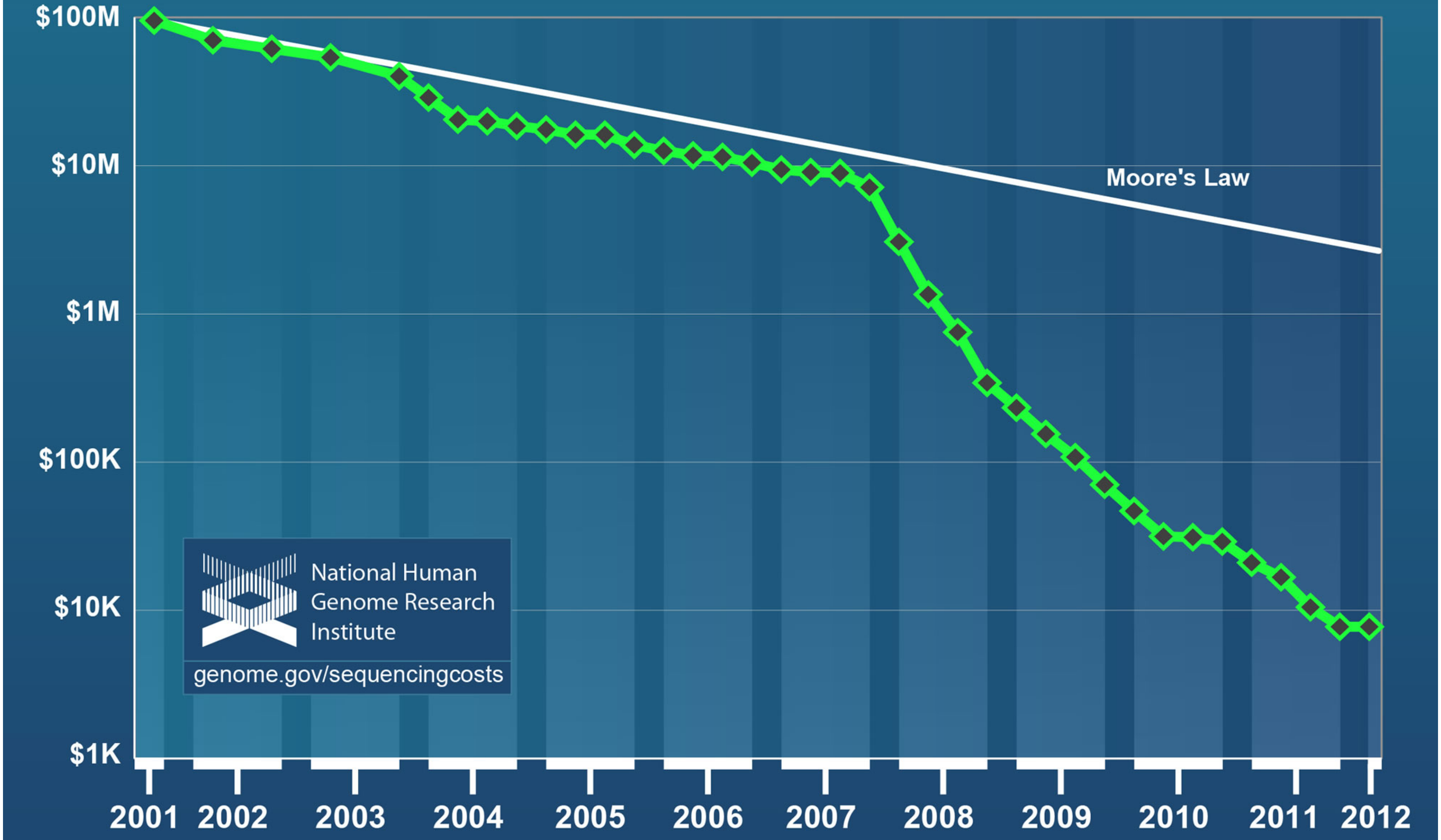
Cost per Raw Megabase of DNA Sequence



National Human Genome Research Institute

genome.gov/sequencingcosts

Cost per Genome



National Human
Genome Research
Institute

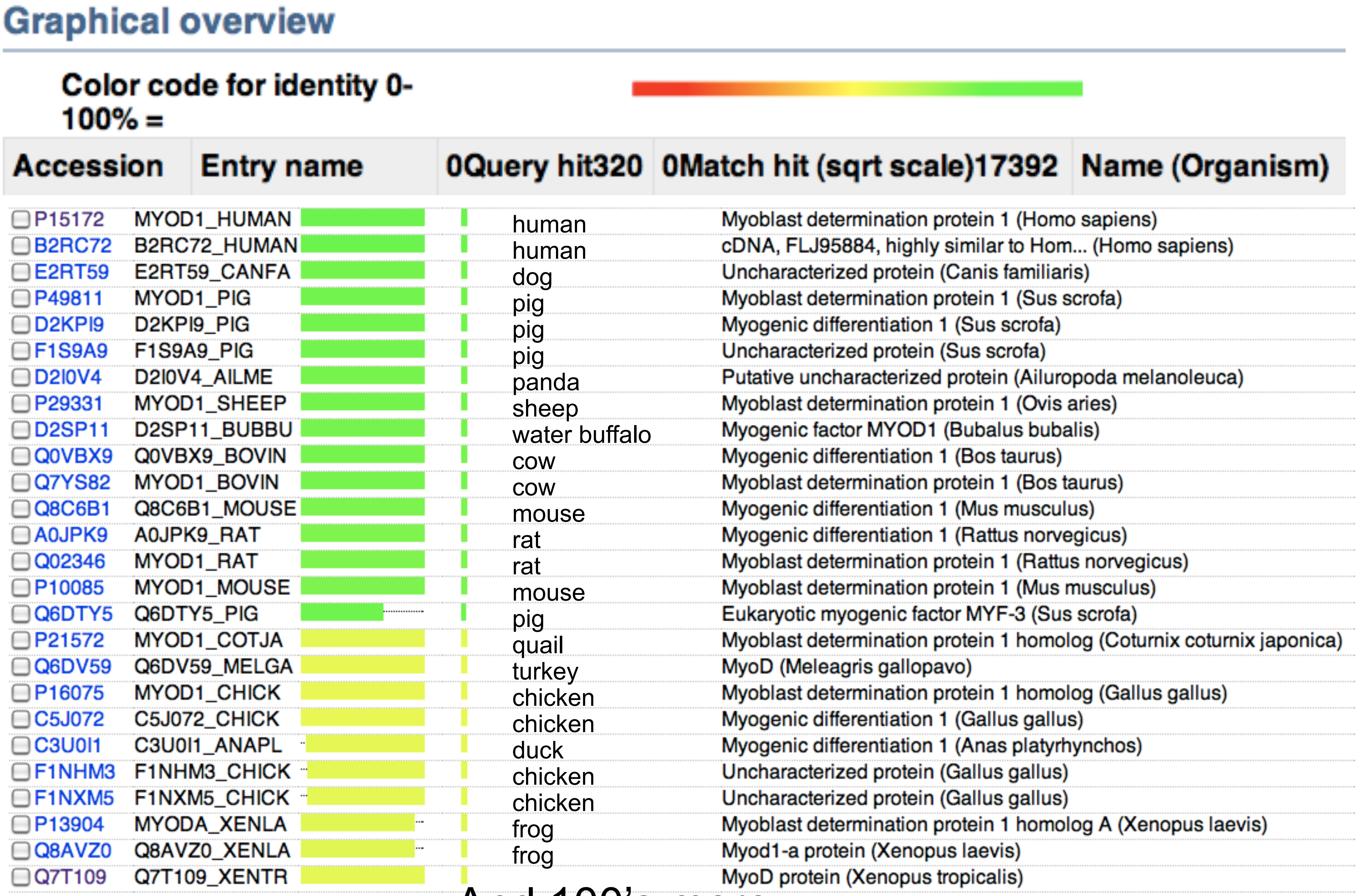
genome.gov/sequencingcosts

A Database Search

go to, e.g., http://www.uniprot.org/, “blast” tab, and paste in this:

```
>sp|P15172|MYOD1_HUMAN Myoblast determination protein 1 OS=Homo
  sapiens GN=MYOD1 PE=1 SV=3
MELLSPPLRDVDLTAPDGSLCSFATTDDFYDDPCFDSPDLRFFEDLDPRLMHVGALLKPE
EHSHFPAAVHPAPGAREDEHVRAPSGHHQAGRCLLWACKACKRKTTNADRRKAATMRERR
RLSKVNEAFETLKRCTSSNPNQRLPKVEILRNAIRYIEGLQALLRDQDAAPPGAAAAFYA
PGPLPPGRGGEHYSGDSDASSPRSNCSDGMMDYSGPPSGARRRNCYEGAYYNEAPSEPRP
GKSAAVSSLDCLSSIVERISTESPAAPALLLADVPSESPPRRQEAAAPSEGESSGDPTQS
PDAAPQCPAGANPNPIYQVL
```

A Few seconds Later…



Graphical overview

Color code for identity 0-100% =

Accession	Entry name	0Query hit320	0Match hit (sqrt scale)17392		Name (Organism)
☐ P15172	MYOD1_HUMAN	🟩	🟩	human	Myoblast determination protein 1 (Homo sapiens)
☐ B2RC72	B2RC72_HUMAN	🟩	🟩	human	cDNA, FLJ95884, highly similar to Hom... (Homo sapiens)
☐ E2RT59	E2RT59_CANFA	🟩	🟩	dog	Uncharacterized protein (Canis familiaris)
☐ P49811	MYOD1_PIG	🟩	🟩	pig	Myoblast determination protein 1 (Sus scrofa)
☐ D2KPI9	D2KPI9_PIG	🟩	🟩	pig	Myogenic differentiation 1 (Sus scrofa)
☐ F1S9A9	F1S9A9_PIG	🟩	🟩	pig	Uncharacterized protein (Sus scrofa)
☐ D2I0V4	D2I0V4_AILME	🟩	🟩	panda	Putative uncharacterized protein (Ailuropoda melanoleuca)
☐ P29331	MYOD1_SHEEP	🟩	🟩	sheep	Myoblast determination protein 1 (Ovis aries)
☐ D2SP11	D2SP11_BUBBU	🟩	🟩	water buffalo	Myogenic factor MYOD1 (Bubalus bubalis)
☐ Q0VBX9	Q0VBX9_BOVIN	🟩	🟩	cow	Myogenic differentiation 1 (Bos taurus)
☐ Q7YS82	MYOD1_BOVIN	🟩	🟩	cow	Myoblast determination protein 1 (Bos taurus)
☐ Q8C6B1	Q8C6B1_MOUSE	🟩	🟩	mouse	Myogenic differentiation 1 (Mus musculus)
☐ A0JPK9	A0JPK9_RAT	🟩	🟩	rat	Myogenic differentiation 1 (Rattus norvegicus)
☐ Q02346	MYOD1_RAT	🟩	🟩	rat	Myoblast determination protein 1 (Rattus norvegicus)
☐ P10085	MYOD1_MOUSE	🟩	🟩	mouse	Myoblast determination protein 1 (Mus musculus)
☐ Q6DTY5	Q6DTY5_PIG	🟩	🟩	pig	Eukaryotic myogenic factor MYF-3 (Sus scrofa)
☐ P21572	MYOD1_COTJA	🟨	🟨	quail	Myoblast determination protein 1 homolog (Coturnix coturnix japonica)
☐ Q6DV59	Q6DV59_MELGA	🟨	🟨	turkey	MyoD (Meleagris gallopavo)
☐ P16075	MYOD1_CHICK	🟨	🟨	chicken	Myoblast determination protein 1 homolog (Gallus gallus)
☐ C5J072	C5J072_CHICK	🟨	🟨	chicken	Myogenic differentiation 1 (Gallus gallus)
☐ C3U0I1	C3U0I1_ANAPL	🟨	🟨	duck	Myogenic differentiation 1 (Anas platyrhynchos)
☐ F1NHM3	F1NHM3_CHICK	🟨	🟨	chicken	Uncharacterized protein (Gallus gallus)
☐ F1NXM5	F1NXM5_CHICK	🟨	🟨	chicken	Uncharacterized protein (Gallus gallus)
☐ P13904	MYODA_XENLA	🟨	🟨	frog	Myoblast determination protein 1 homolog A (Xenopus laevis)
☐ Q8AVZ0	Q8AVZ0_XENLA	🟨	🟨	frog	Myod1-a protein (Xenopus laevis)
☐ Q7T109	Q7T109_XENTR	🟨	🟨		MyoD protein (Xenopus tropicalis)

…And 100's more…

Accession	Entry name	Status	Protein names	Organism	Length
Q7T109	Q7T109_XENTR	★	**MyoD protein**	Xenopus tropicalis (Western clawed frog) (Silurana tropicalis)	288

Some Details from #25

Alignment 1 against Q7T109

Score	964	**E-value**	1.0 ×10 $^{-102}$
Identity	64.0%	**Positives**	74.0%
Query length	320	**Match length**	288
Position	**Q7T109** matches from 1 to 288 (288AA), in the query sequence from 1 to 320 (320AA)		
Graphical	🟨		

```
1      MELLSPPLRDVDLTAPDGSLCSFATTDDFYDDPCFDSPDLRFFEDLDPRLMHVGALLKPE    60 P15172
       MELL PPLRD+++T  +GSLCSF T DDFYDDPCF++ D+ FFEDLDPRL+HV ALLKPE
1      MELLPPPLRDMEVT--EGSLCSFPTPDDFYDDPCFNTSDMSFFEDLDPRLVHV-ALLKPE    57 Q7T109

61     EHSHFPAAVHPAPGAREDEHVRAPSGHHQAGRCLLWACKACKRKTTNADRRKAATMRERR   120 P15172
       +  H            EDEHVRAPSGHHQAGRCLLWACKACKRKTTNADRRKAATMRERR
58     DPHH-----------NEDEHVRAPSGHHQAGRCLLWACKACKRKTTNADRRKAATMRERR   106 Q7T109

121    RLSKVNEAFETLKRCTSSNPNQRLPKVEILRNAIRYIEGLQALLRDQDAAPGAAAAFYA   180 P15172
       RLSKVNEAFETLKRCTS+NPNQRLPKVEILRNAIRYIE LQ+LLR Q+        +FY
107    RLSKVNEAFETLKRCTSTNPNQRLPKVEILRNAIRYIESLQSLLRGQE-------ESFY-   158 Q7T109

181    PGPLPPGRGGEHYSGDSDASSPRSNCSDGMMDYSGPPSGARRRNCYEGAYYNEAPSEPRP   240 P15172
         P+      EHYSGDSDASSPRSNCSDGM DYS PP G+RRRN Y+ ++Y+++P+  R
159    --PVL-----EHYSGDSDASSPRSNCSDGMTDYS-PPCGSRRRNSYDSSFYSDSPNGLRL   210 Q7T109

241    GKSAAVSSLDCLSSIVERISTESPAAPALLLADVPSESPPRRQEAAAPSEGES---SGDP   297 P15172
       GKS+ +SSLDCLSSIVERISTESP  P +  AD  SE  P      +P +GE+   SG
211    GKSSVISSLDCLSSIVERISTESPVCPVIPAADSGSEGSP-----CSPLQGETLSESGII   265 Q7T109
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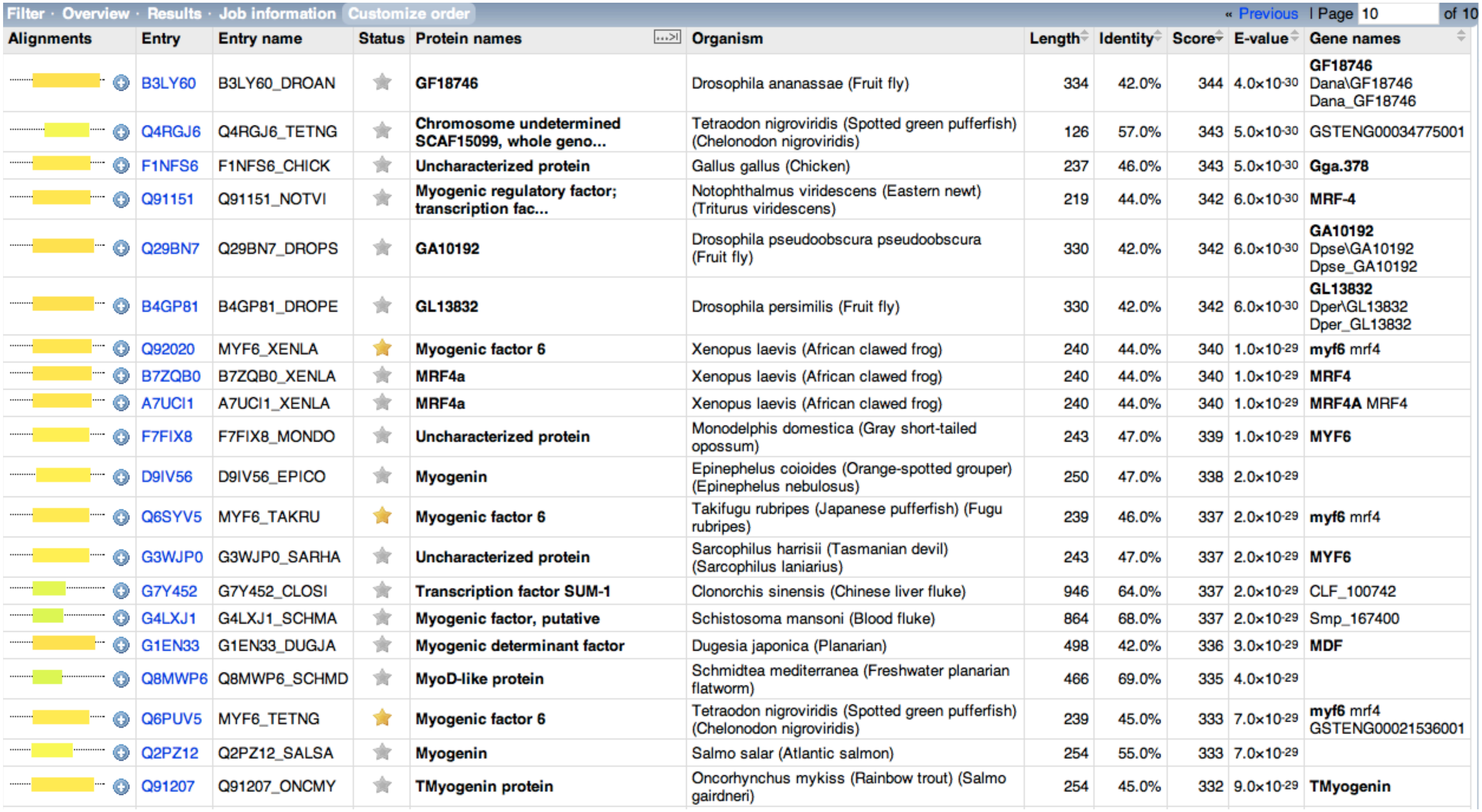


Alignments	Entry	Entry name	Status	Protein names	Organism	Length	Identity	Score	E-value	Gene names
	B3LY60	B3LY60_DROAN	☆	**GF18746**	Drosophila ananassae (Fruit fly)	334	42.0%	344	4.0×10^{-30}	**GF18746** Dana\GF18746 Dana_GF18746
	Q4RGJ6	Q4RGJ6_TETNG	☆	**Chromosome undetermined SCAF15099, whole geno...**	Tetraodon nigroviridis (Spotted green pufferfish) (Chelonodon nigroviridis)	126	57.0%	343	5.0×10^{-30}	GSTENG00034775001
	F1NFS6	F1NFS6_CHICK	☆	**Uncharacterized protein**	Gallus gallus (Chicken)	237	46.0%	343	5.0×10^{-30}	**Gga.378**
	Q91151	Q91151_NOTVI	☆	**Myogenic regulatory factor; transcription fac...**	Notophthalmus viridescens (Eastern newt) (Triturus viridescens)	219	44.0%	342	6.0×10^{-30}	**MRF-4**
	Q29BN7	Q29BN7_DROPS	☆	**GA10192**	Drosophila pseudoobscura pseudoobscura (Fruit fly)	330	42.0%	342	6.0×10^{-30}	**GA10192** Dpse\GA10192 Dpse_GA10192
	B4GP81	B4GP81_DROPE	☆	**GL13832**	Drosophila persimilis (Fruit fly)	330	42.0%	342	6.0×10^{-30}	**GL13832** Dper\GL13832 Dper_GL13832
	Q92020	MYF6_XENLA	★	**Myogenic factor 6**	Xenopus laevis (African clawed frog)	240	44.0%	340	1.0×10^{-29}	**myf6** mrf4
	B7ZQB0	B7ZQB0_XENLA	☆	**MRF4a**	Xenopus laevis (African clawed frog)	240	44.0%	340	1.0×10^{-29}	**MRF4**
	A7UCI1	A7UCI1_XENLA	☆	**MRF4a**	Xenopus laevis (African clawed frog)	240	44.0%	340	1.0×10^{-29}	**MRF4A** MRF4
	F7FIX8	F7FIX8_MONDO	☆	**Uncharacterized protein**	Monodelphis domestica (Gray short-tailed opossum)	243	47.0%	339	1.0×10^{-29}	**MYF6**
	D9IV56	D9IV56_EPICO	☆	**Myogenin**	Epinephelus coioides (Orange-spotted grouper) (Epinephelus nebulosus)	250	47.0%	338	2.0×10^{-29}	
	Q6SYV5	MYF6_TAKRU	★	**Myogenic factor 6**	Takifugu rubripes (Japanese pufferfish) (Fugu rubripes)	239	46.0%	337	2.0×10^{-29}	**myf6** mrf4
	G3WJP0	G3WJP0_SARHA	☆	**Uncharacterized protein**	Sarcophilus harrisii (Tasmanian devil) (Sarcophilus laniarius)	243	47.0%	337	2.0×10^{-29}	**MYF6**
	G7Y452	G7Y452_CLOSI	☆	**Transcription factor SUM-1**	Clonorchis sinensis (Chinese liver fluke)	946	64.0%	337	2.0×10^{-29}	CLF_100742
	G4LXJ1	G4LXJ1_SCHMA	☆	**Myogenic factor, putative**	Schistosoma mansoni (Blood fluke)	864	68.0%	337	2.0×10^{-29}	Smp_167400
	G1EN33	G1EN33_DUGJA	☆	**Myogenic determinant factor**	Dugesia japonica (Planarian)	498	42.0%	336	3.0×10^{-29}	**MDF**
	Q8MWP6	Q8MWP6_SCHMD	☆	**MyoD-like protein**	Schmidtea mediterranea (Freshwater planarian flatworm)	466	69.0%	335	4.0×10^{-29}	
	Q6PUV5	MYF6_TETNG	★	**Myogenic factor 6**	Tetraodon nigroviridis (Spotted green pufferfish) (Chelonodon nigroviridis)	239	45.0%	333	7.0×10^{-29}	**myf6** mrf4 GSTENG00021536001
	Q2PZ12	Q2PZ12_SALSA	☆	**Myogenin**	Salmo salar (Atlantic salmon)	254	55.0%	333	7.0×10^{-29}	
	Q91207	Q91207_ONCMY	☆	**TMyogenin protein**	Oncorhynchus mykiss (Rainbow trout) (Salmo gairdneri)	254	45.0%	332	9.0×10^{-29}	**TMyogenin**

hits at rank ~ 250 still extremely good matches,
even though very distantly related organisms

The foregoing search capability is a *huge* deal

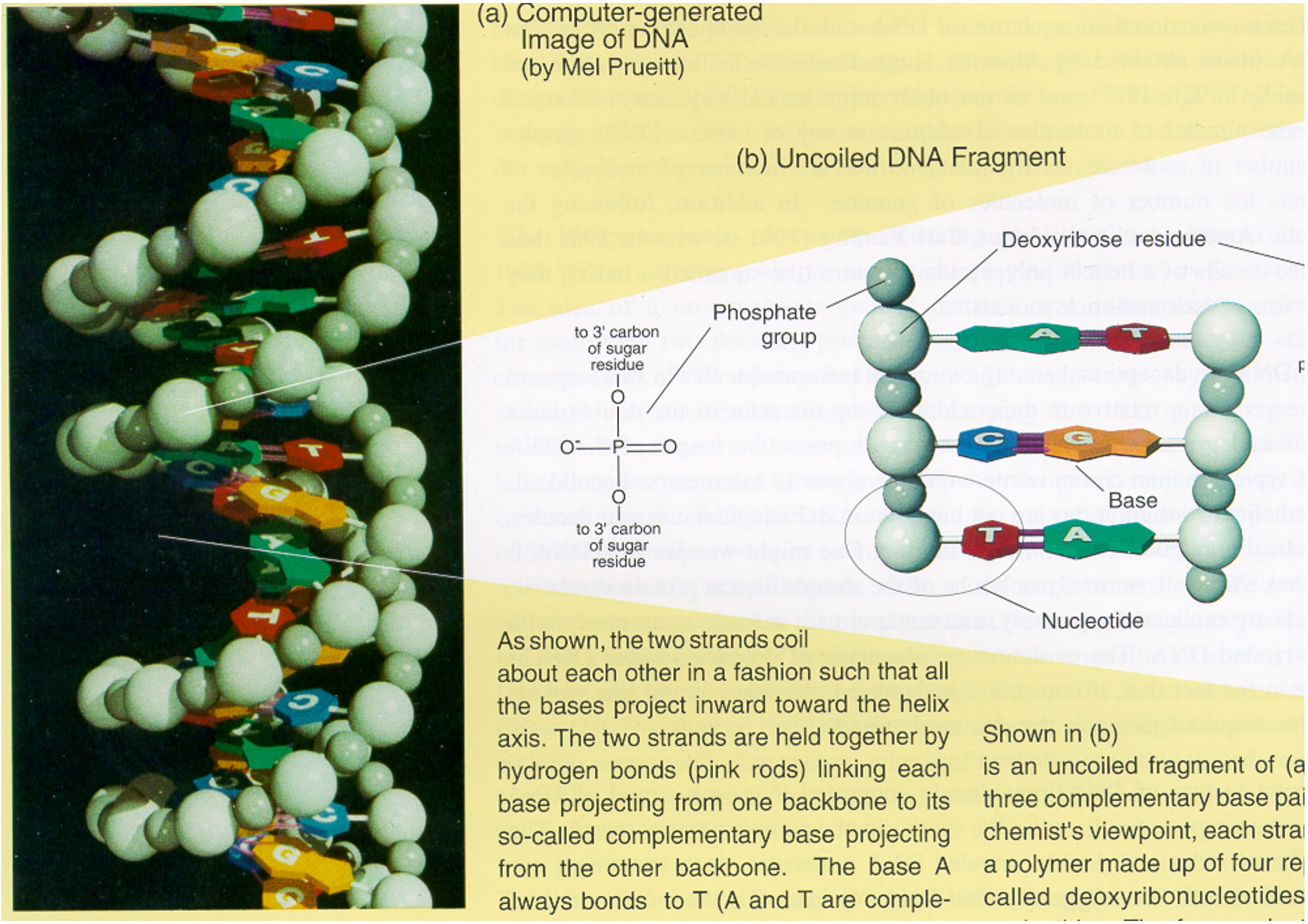
millions of searches

nearly all biologists (not just “computational biologists”) use this routinely

It connects information about *all* living things

Application: RNA structure

The Double Helix



(a) Computer-generated Image of DNA (by Mel Prueitt)

(b) Uncoiled DNA Fragment

As shown, the two strands coil about each other in a fashion such that all the bases project inward toward the helix axis. The two strands are held together by hydrogen bonds (pink rods) linking each base projecting from one backbone to its so-called complementary base projecting from the other backbone. The base A always bonds to T (A and T are comple-

Shown in (b)
is an uncoiled fragment of (a
three complementary base pai
chemist's viewpoint, each stra
a polymer made up of four re
called deoxyribonucleotides

Los Alamos Science

http://www.rcsb.org/pdb/explore.do?structureId=1GAT

NATURE VOL. 227 AUGUST 8 1970

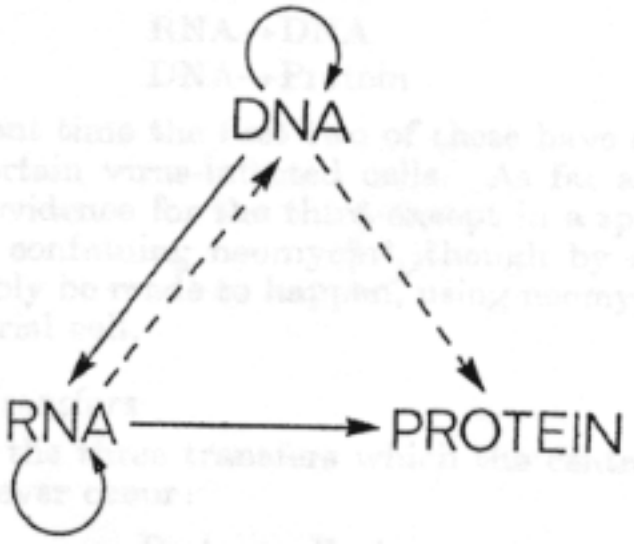
Central Dogma of Molecular Biology

by
FRANCIS CRICK
MRC Laboratory
Hills Road,
Cambridge CB2 2QH

The central dogma of molecular biology deals with the detailed residue-by-residue transfer of sequential information. It states that such information cannot be transferred from protein to either protein or nucleic acid.

"The central dogma, enunciated by Crick in 1958 and the keystone of molecular biology ever since, is likely to prove a considerable over-simplification."

Fig. 2. The arrows show the situation as it seemed in 1958. Solid arrows represent probable transfers, dotted arrows possible transfers. The absent arrows (compare Fig. 1) represent the impossible transfers postulated by the central dogma. They are the three possible arrows starting from protein.



Non-coding RNA

Messenger RNA - codes for proteins

Non-coding RNA - all the rest

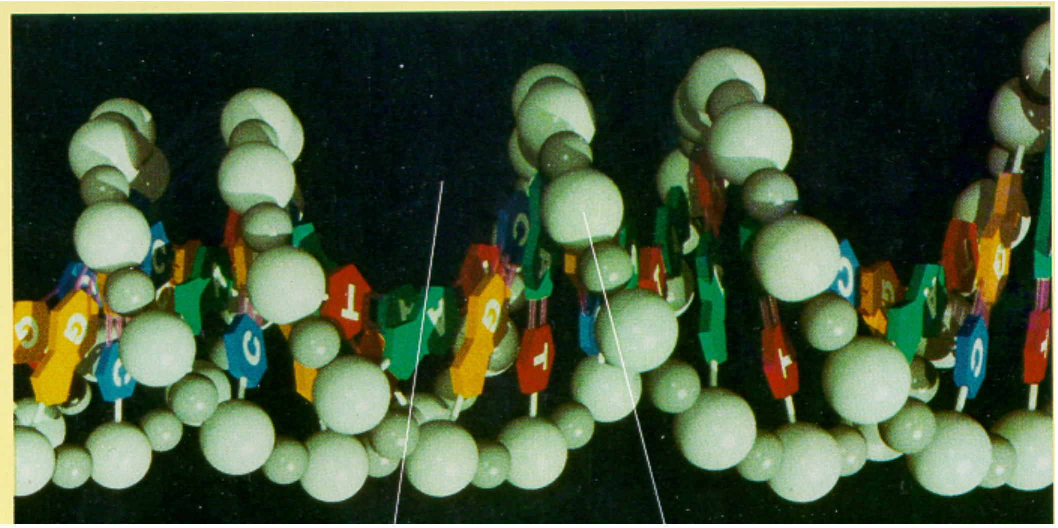
- Before, say, mid 1990's, 1-2 dozen known
  - (critically important, but narrow roles)

Since mid 90's dramatic discoveries

- Regulation, transport, stability/degradation
- E.g. "microRNA": 100s in humans => 50% of genes
- E.g. "riboswitches": 1000s in bacteria

DNA structure: dull

5'…ACCGCTAGATG…3'
| | | | | | | | | | |
3'…TGGCGATCTAC…5'

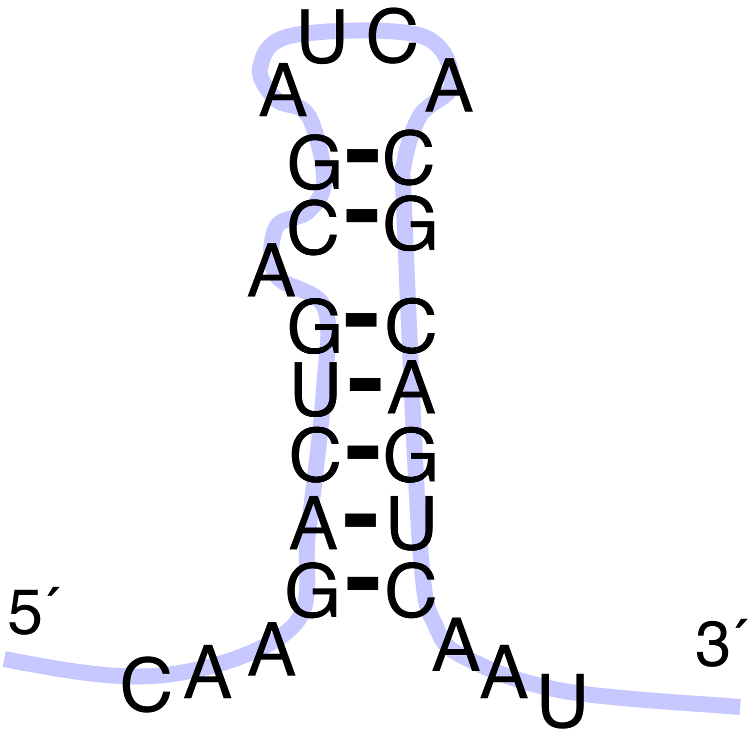


RNA Secondary Structure:
RNA makes helices too

Base pairs

A-U

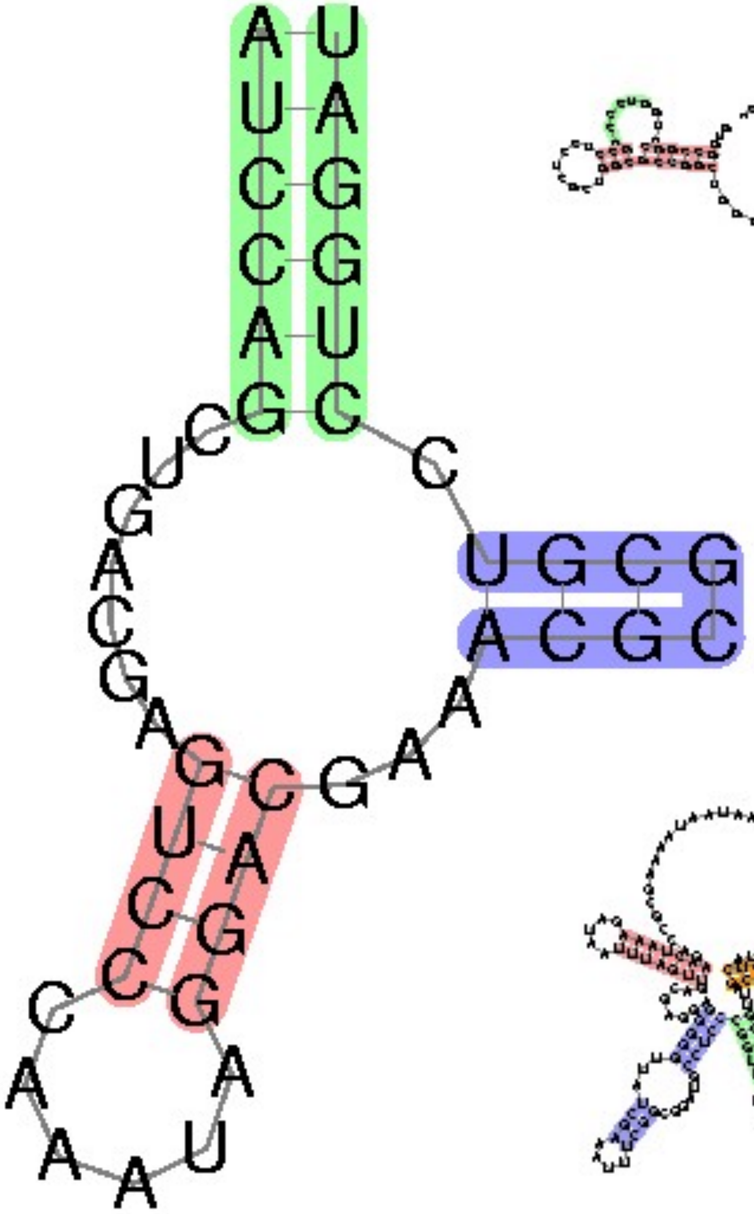
C-G

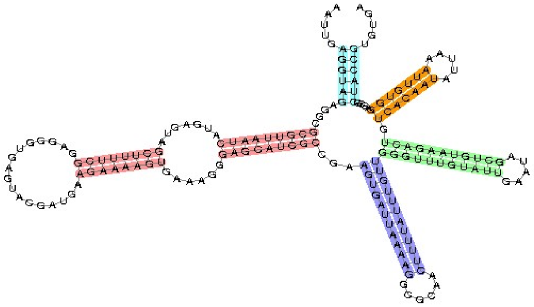


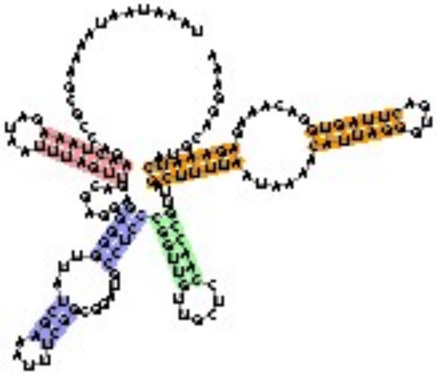
Usually *single* stranded

RNA Secondary Structure:

Not everything,
but important,
easier than 3d



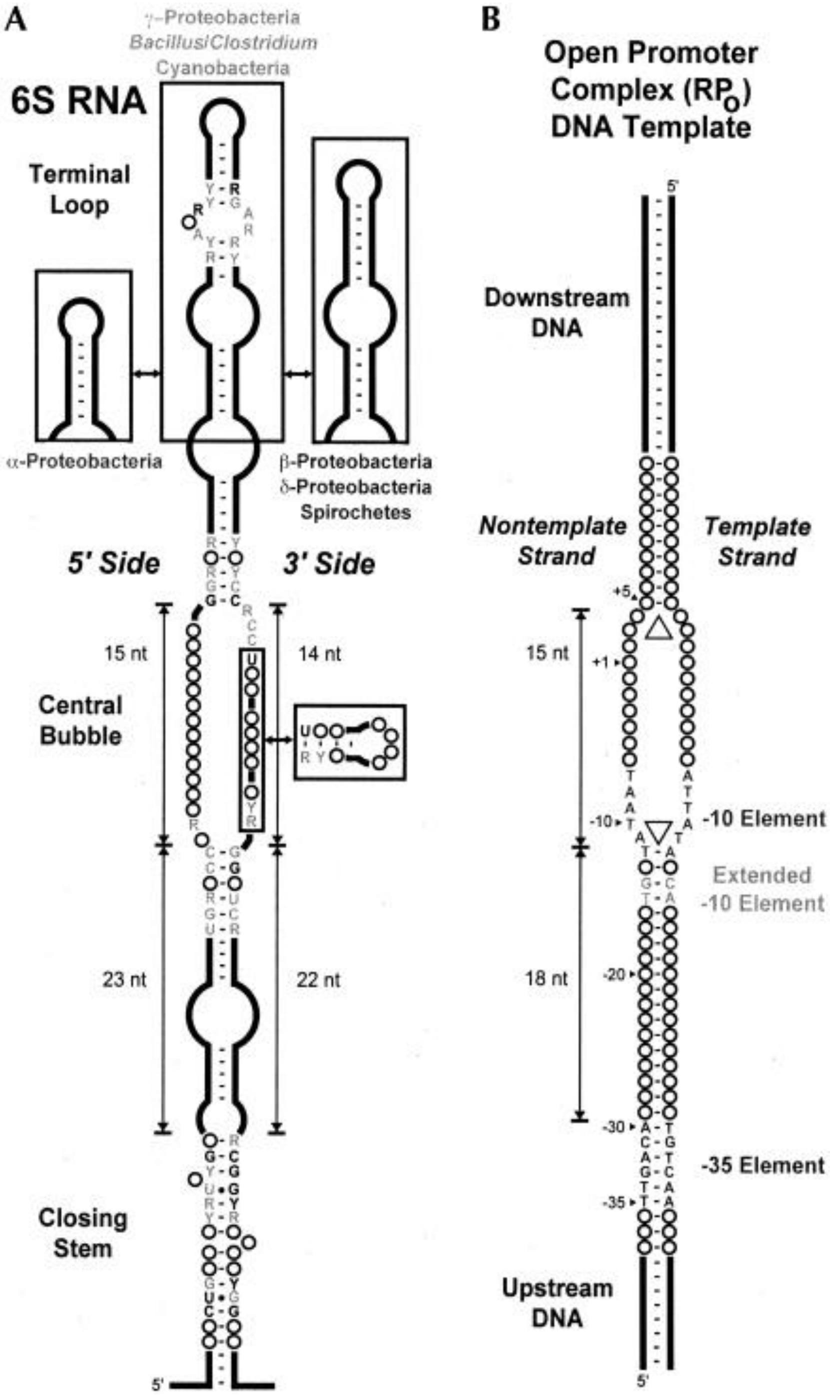


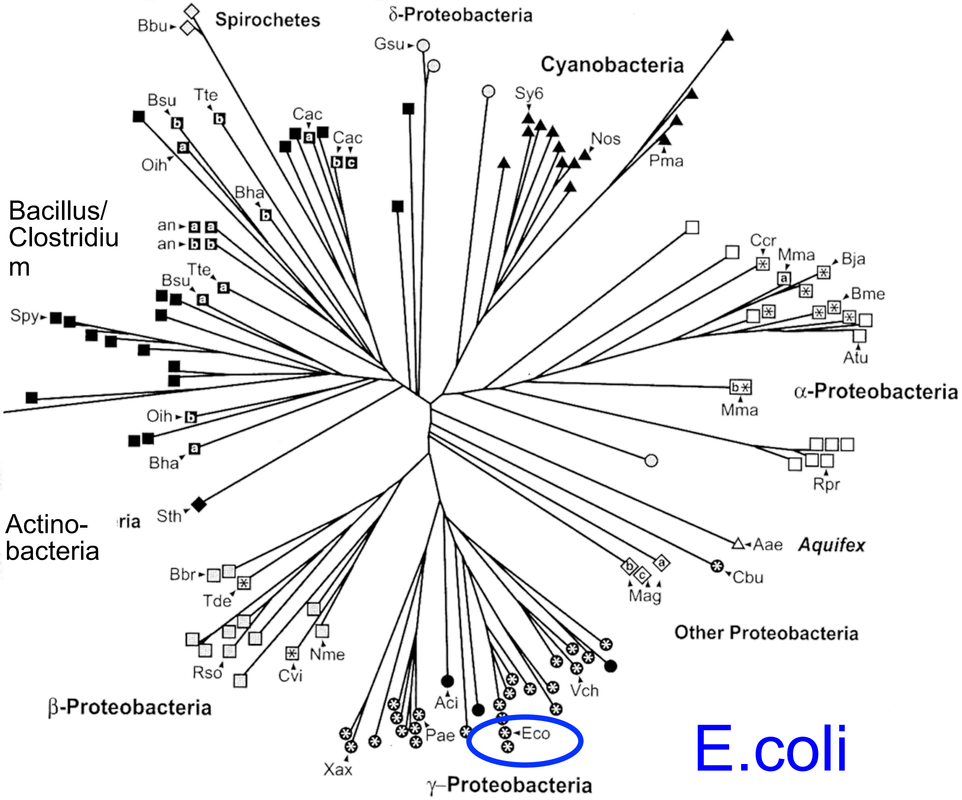


Why is structure important?

- For protein-coding, similarity in sequence is a powerful tool for finding related sequences
  - e.g. “hemoglobin” is easily recognized in all vertebrates
- For non-coding RNA, many different sequences have the same structure, and structure is most important for function.
  - So, using structure plus sequence, can find related sequences at much greater evolutionary distances

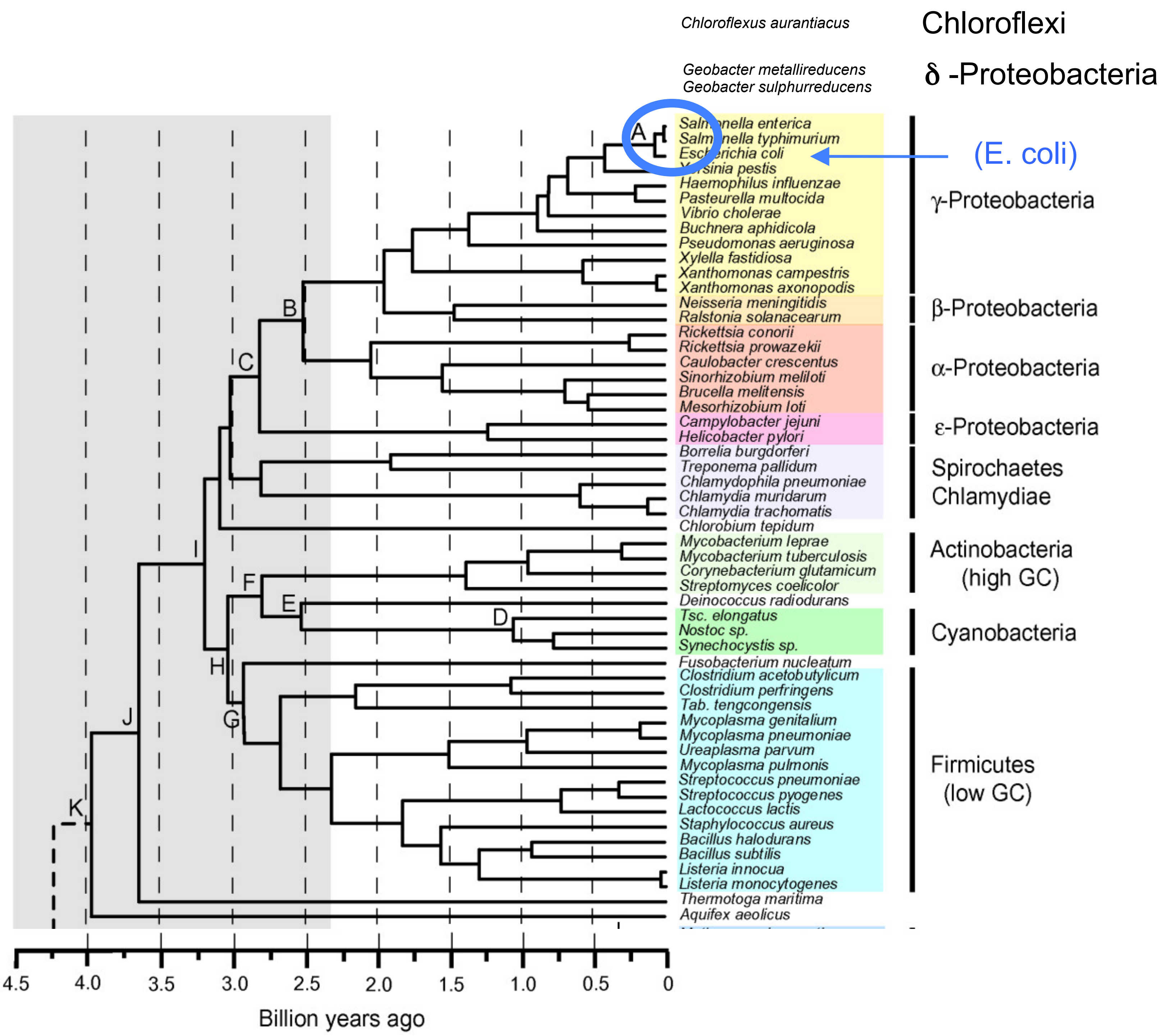
6S mimics an open promoter



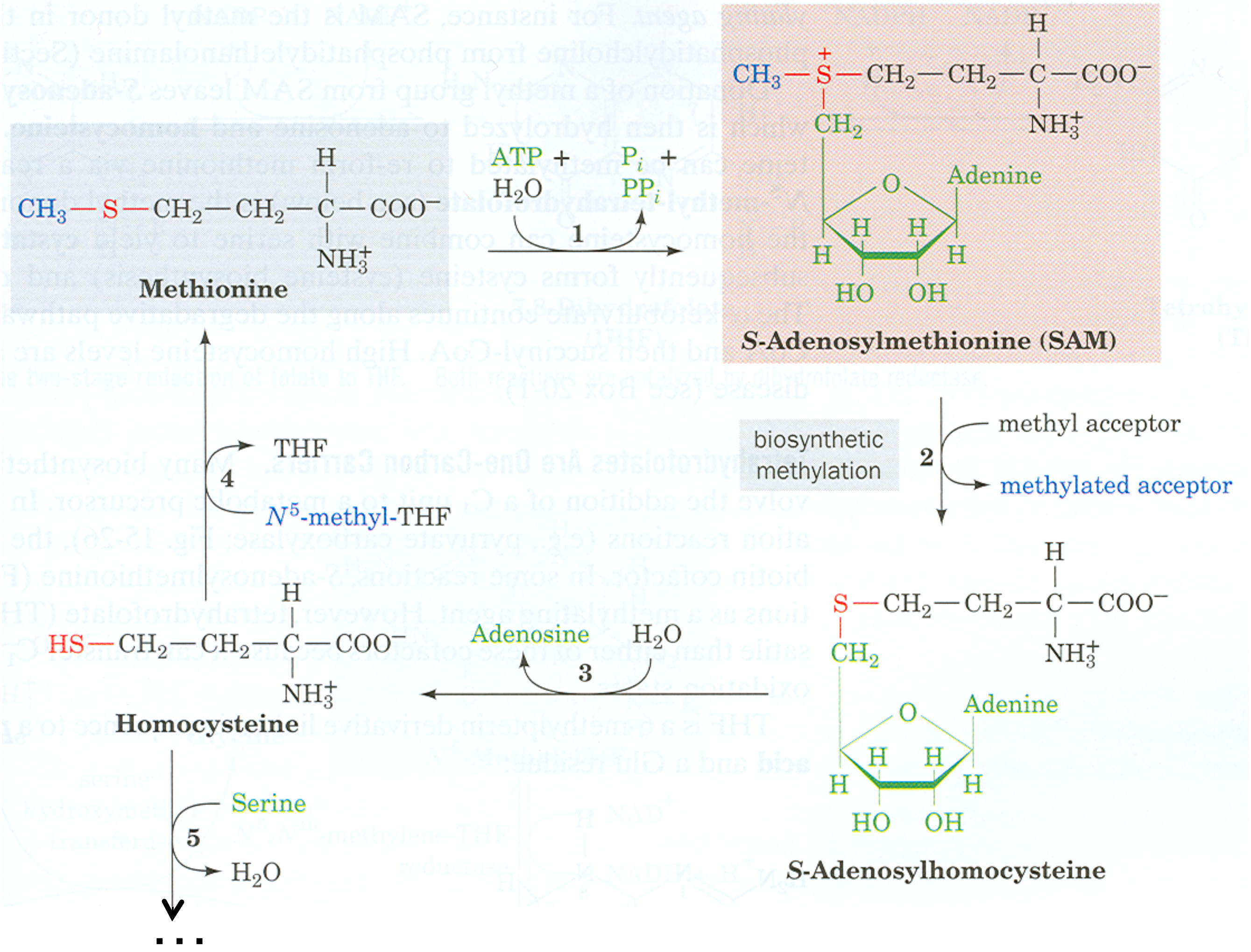


E.coli

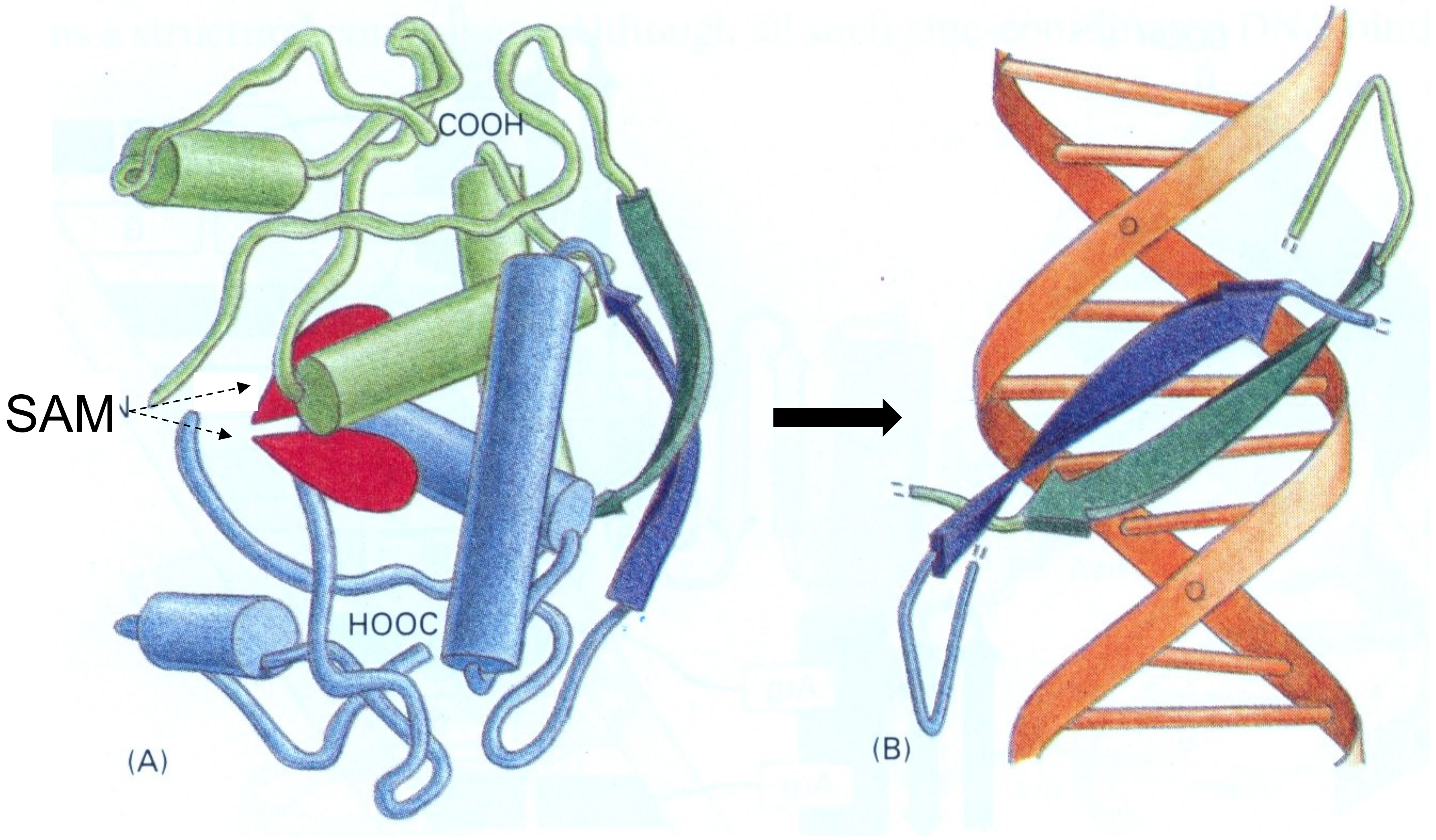
Barrick et al. *RNA* 2005
Trotochaud et al. *NSMB* 2005
Willkomm et al. NAR 2005


Chloroflexus aurantiacus
Chloroflexi
Geobacter metallireducens
Geobacter sulphurreducens
δ -Proteobacteria
Salmonella enterica
Salmonella typhimurium
Escherichia coli
Yersinia pestis
Haemophilus influenzae
Pasteurella multocida
Vibrio cholerae
Buchnera aphidicola
Pseudomonas aeruginosa
Xylella fastidiosa
Xanthomonas campestris
Xanthomonas axonopodis
(E. coli)
γ-Proteobacteria
Neisseria meningitidis
Ralstonia solanacearum
β-Proteobacteria
Rickettsia conorii
Rickettsia prowazekii
Caulobacter crescentus
Sinorhizobium meliloti
Brucella melitensis
Mesorhizobium loti
α-Proteobacteria
Campylobacter jejuni
Helicobacter pylori
ε-Proteobacteria
Borrelia burgdorferi
Treponema pallidum
Chlamydophila pneumoniae
Chlamydia muridarum
Chlamydia trachomatis
Spirochaetes
Chlamydiae
Chlorobium tepidum
Mycobacterium leprae
Mycobacterium tuberculosis
Corynebacterium glutamicum
Streptomyces coelicolor
Actinobacteria
(high GC)
Deinococcus radiodurans
Tsc. elongatus
Nostoc sp.
Synechocystis sp.
Cyanobacteria
Fusobacterium nucleatum
Clostridium acetobutylicum
Clostridium perfringens
Tab. tengcongensis
Mycoplasma genitalium
Mycoplasma pneumoniae
Ureaplasma parvum
Mycoplasma pulmonis
Streptococcus pneumoniae
Streptococcus pyogenes
Lactococcus lactis
Staphylococcus aureus
Bacillus halodurans
Bacillus subtilis
Listeria innocua
Listeria monocytogenes
Firmicutes
(low GC)
Thermotoga maritima
Aquifex aeolicus
A
B
C
D
E
F
G
H
I
J
K
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0
Billion years ago

In Bacteria: A typical biosynthetic cycle around a critical metabolite (“SAM”)



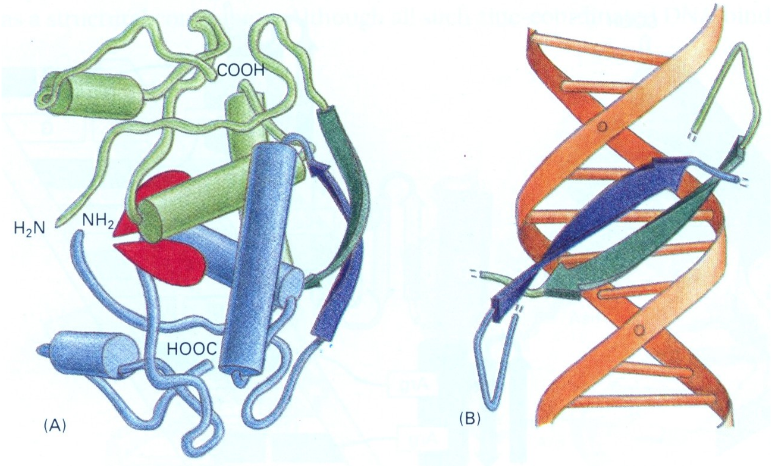
Gene Regulation: The MET Repressor



Protein

Alberts, et al, 3e.

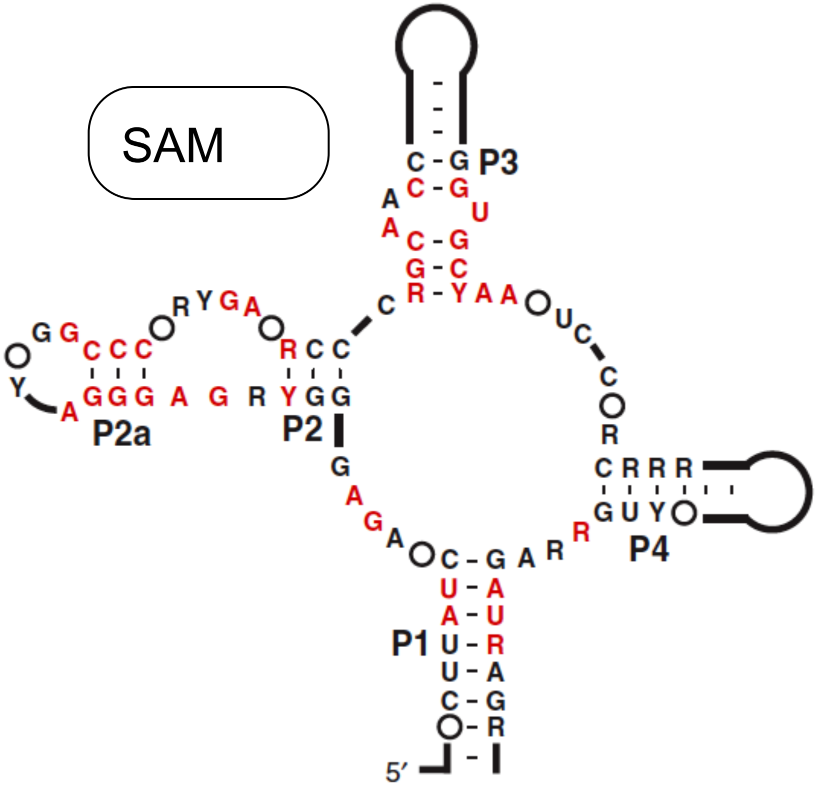
DNA



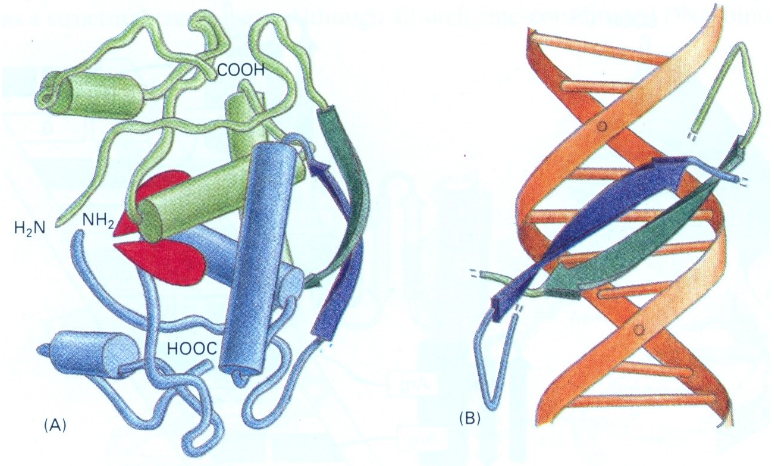
Alberts, et al, 3e.

← The protein way

Riboswitch alternative



Grundy & Henkin, Mol. Microbiol 1998
Epshtein, et al., PNAS 2003
Winkler et al., Nat. Struct. Biol. 2003



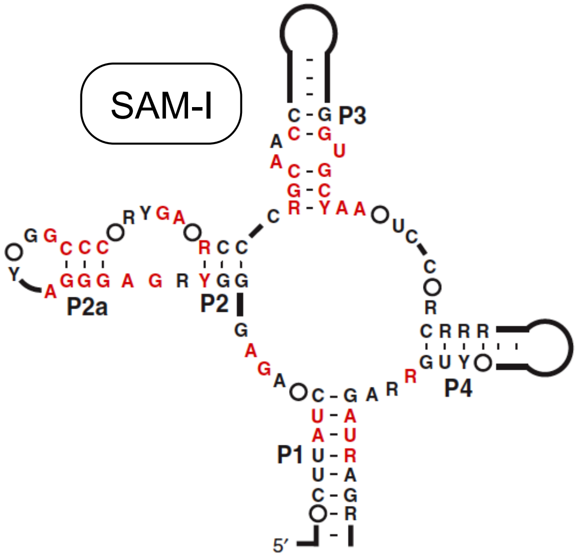
Alberts, et al, 3e.

← The protein way

Riboswitch alternatives

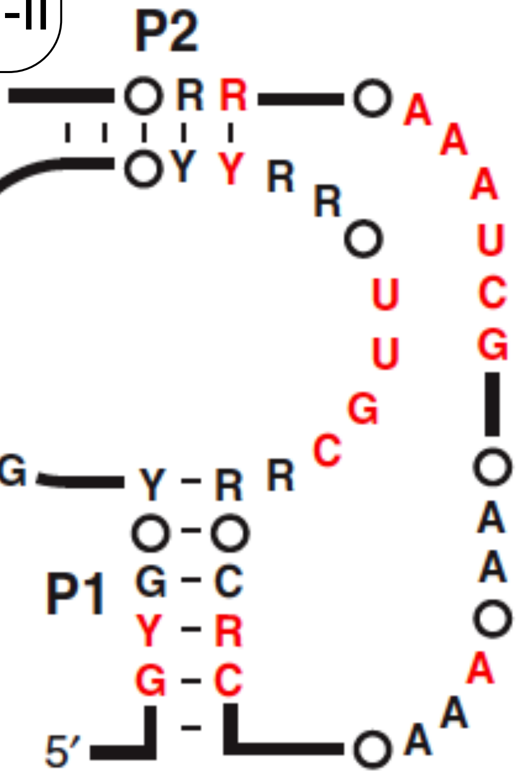
↓

SAM-I



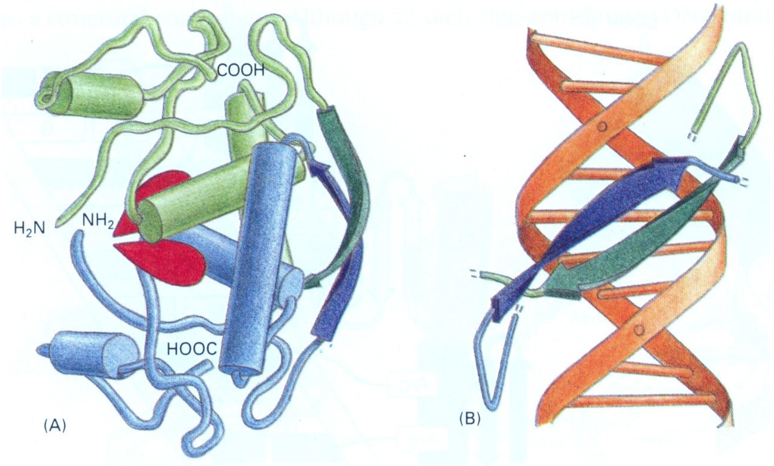
Grundy, Epshtein, Winkler et al., 1998, 2003

SAM-II



Corbino et al., Genome Biol. 2005

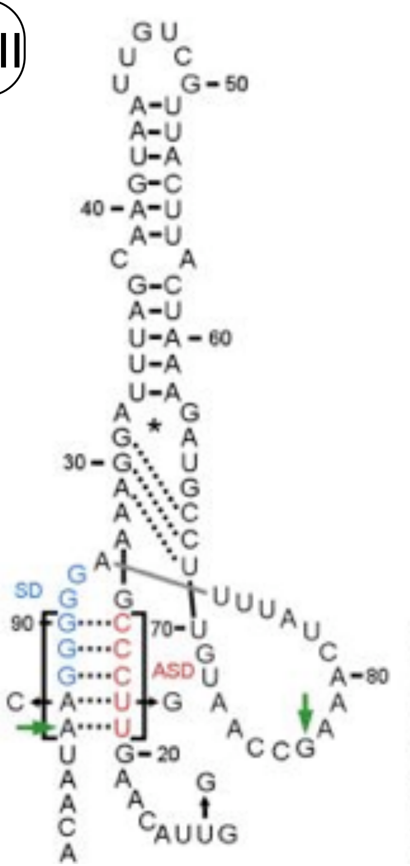
Alberts, et al, 3e.



The protein way

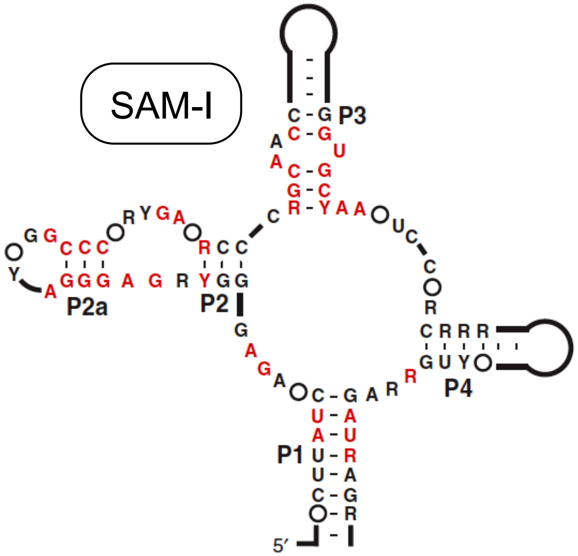
Riboswitch alternatives

SAM-III



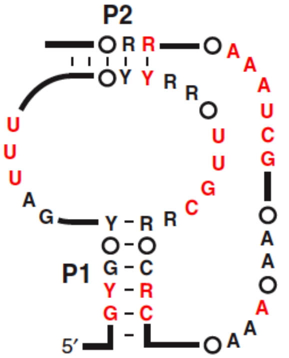
Fuchs et al.,
NSMB 2006

SAM-I



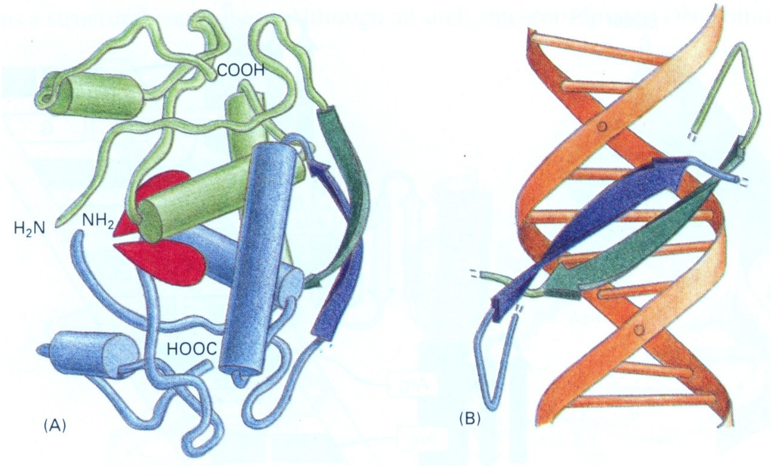
Grundy, Epshtein, Winkler
et al., 1998, 2003

SAM-II



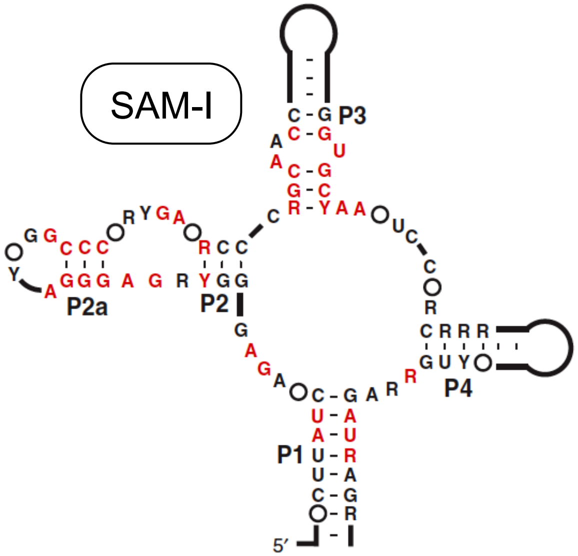
Corbino et al.,
Genome Biol. 2005

Alberts, et al, 3e.



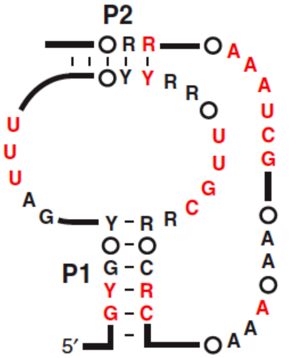
← The protein way

Riboswitch alternatives ↓



SAM-I

Grundy, Epshtein, Winkler et al., 1998, 2003

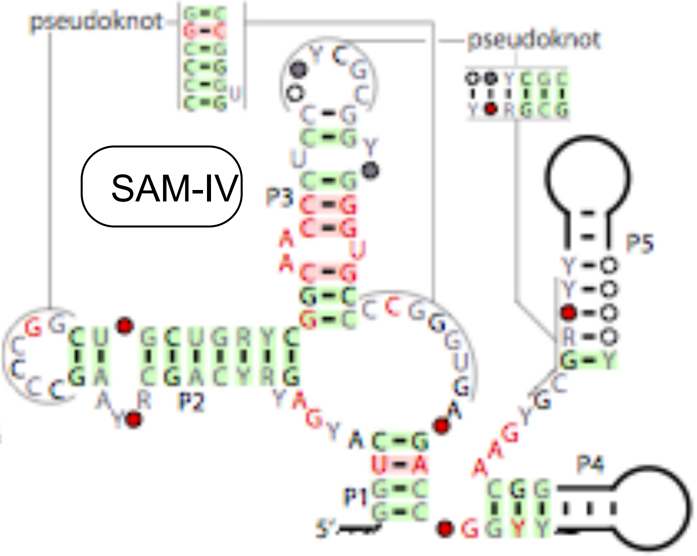


SAM-II

Corbino et al., Genome Biol. 2005

SAM-III

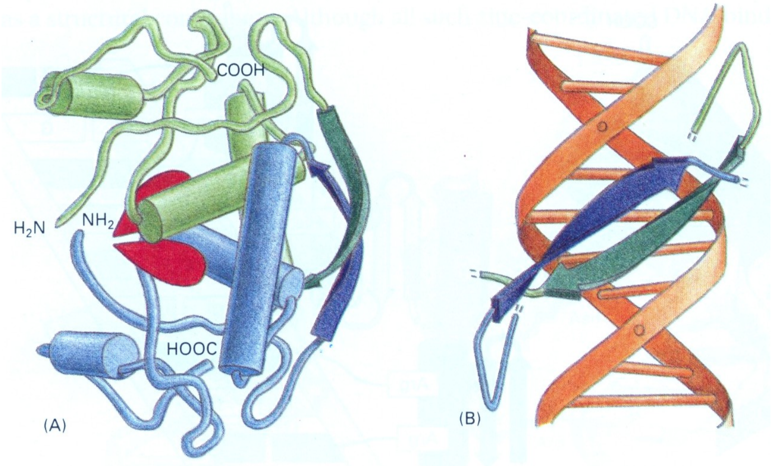
Fuchs et al., NSMB 2006



SAM-IV

Weinberg et al., RNA 2008

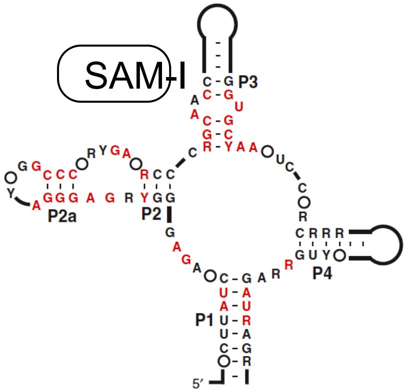
Alberts, et al, 3e.



← The protein way

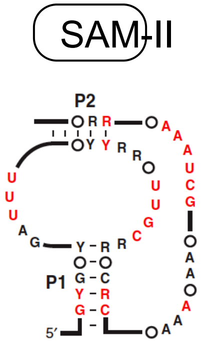
Riboswitch alternatives ↓

SAM-I



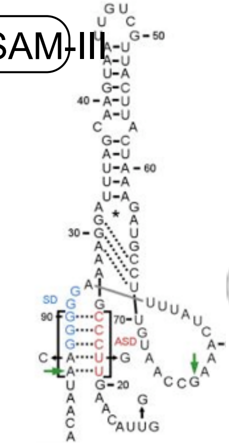
Grundy, Epshtein, Winkler
et al., 1998, 2003

SAM-II



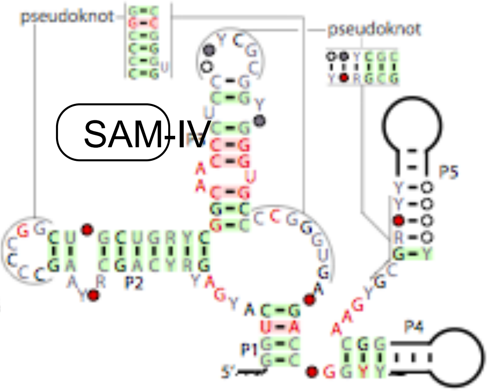
Corbino et al.,
Genome Biol. 2005

SAM-III



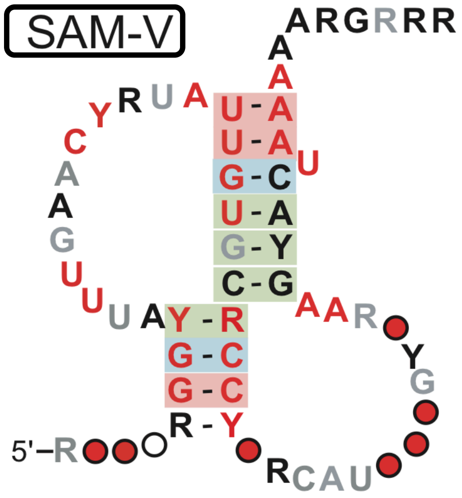
Fuchs
et al.,
NSMB
2006

SAM-IV



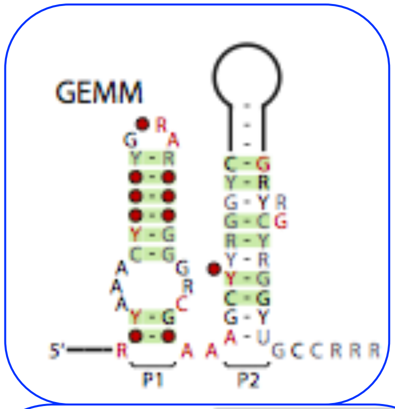
Weinberg
et al.,
RNA 2008

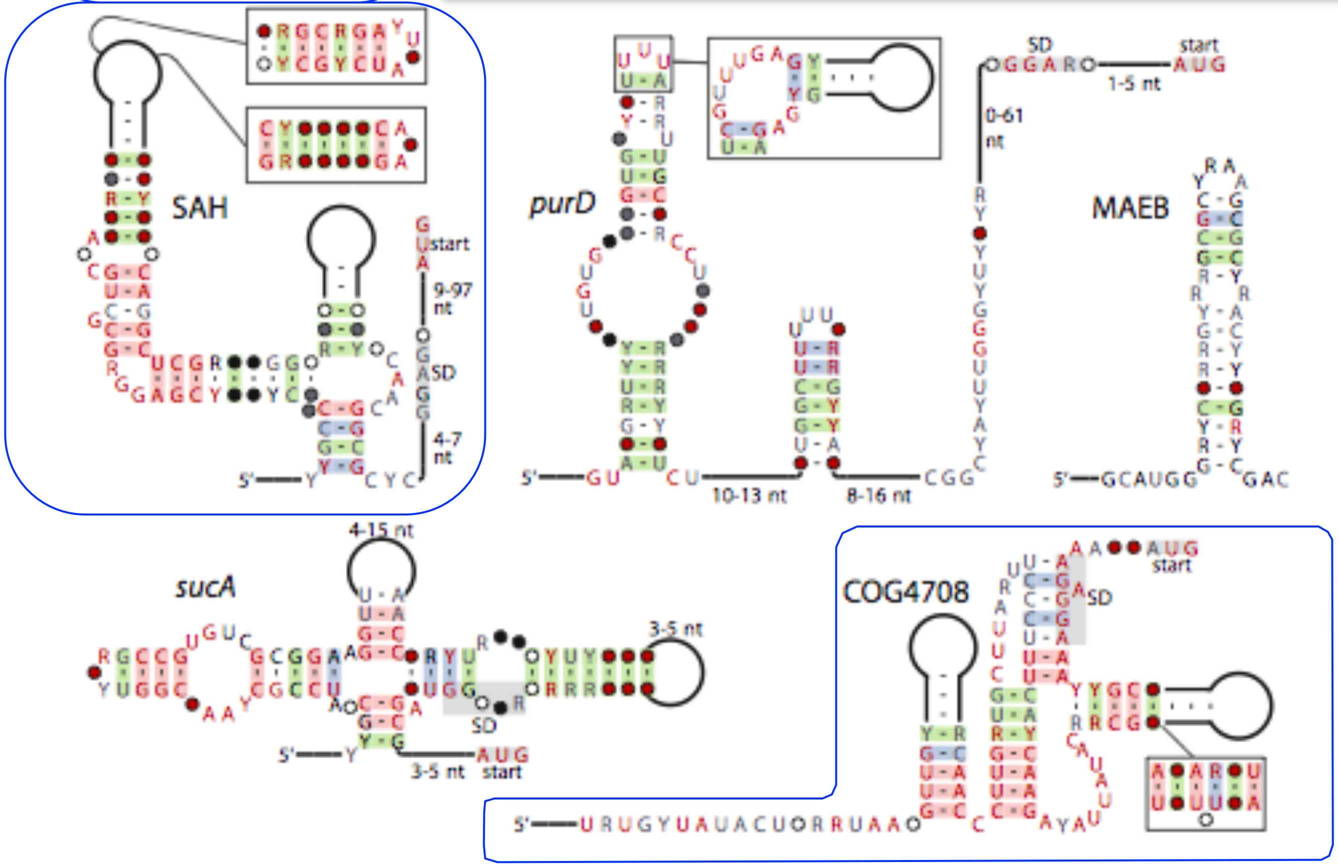
SAM-V



Meyer, etal., BMC
Genomics 2009

And many other examples. Widespread, deeply conserved, structurally sophisticated, functionally diverse, biologically important uses for ncRNA throughout prokaryotic world.

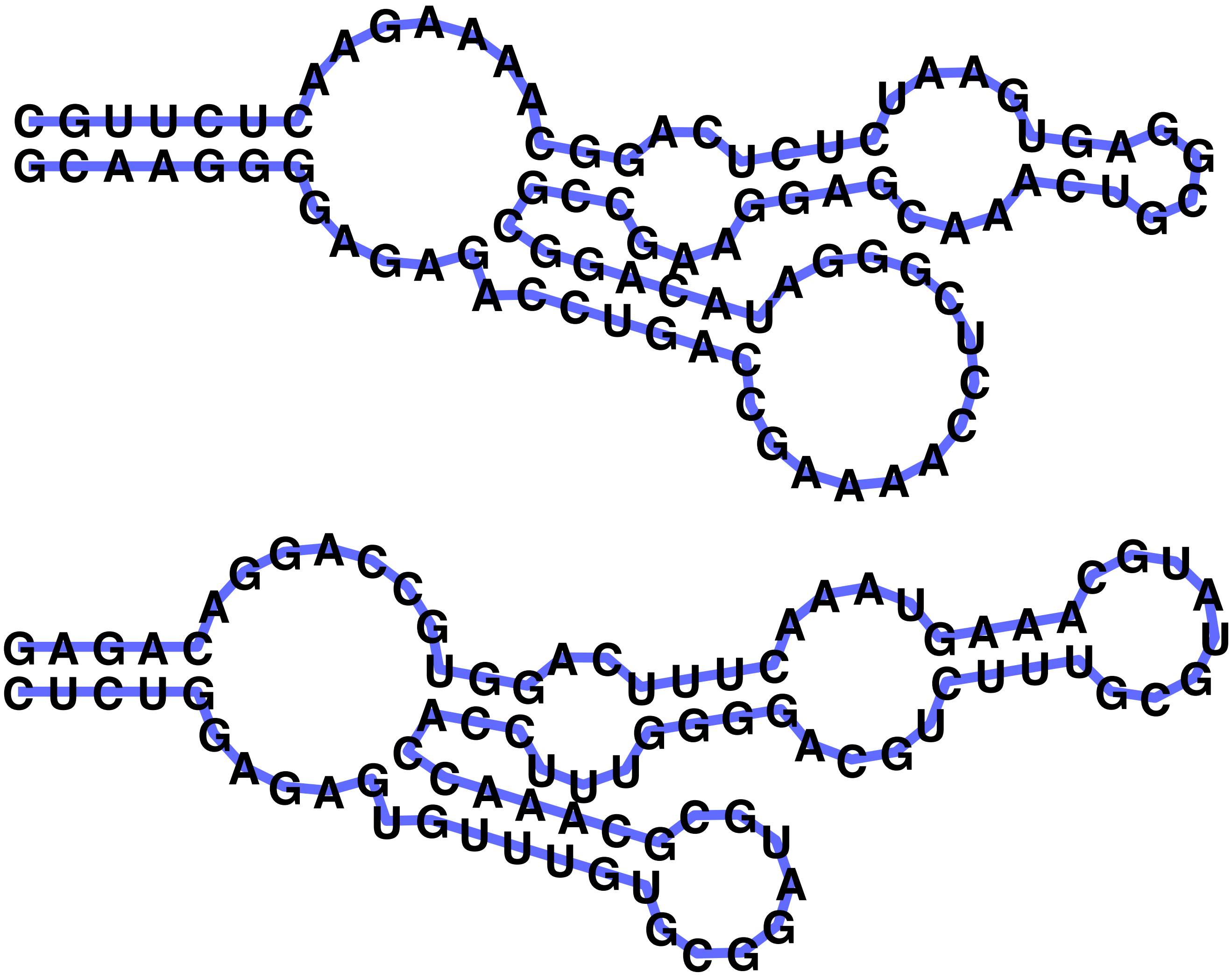




boxed = confirmed riboswitch (+2 more)

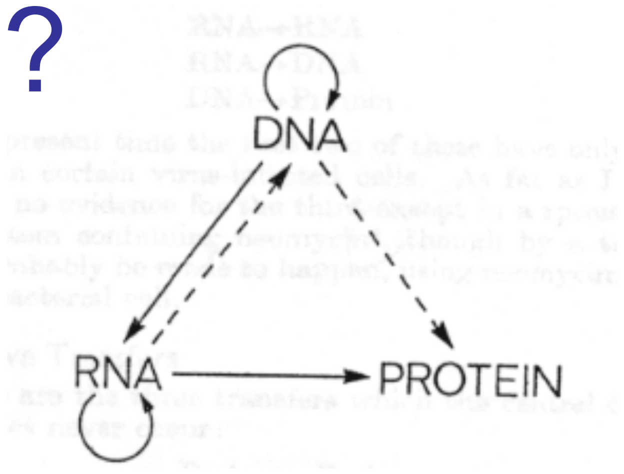
Weinberg, et al. Nucl. Acids Res., July 2007 35: 4809-4819.

Q: What's so hard?



A: Structure often more important than sequence

Origin of Life?



Life needs
- information carrier: DNA
- molecular machines, like enzymes: Protein
- making proteins needs DNA + RNA + proteins
- making (duplicating) DNA needs proteins

Horrible circularities! How could it have arisen in an abiotic environment?

Origin of Life?

RNA can carry information, too

  RNA double helix; RNA-directed RNA polymerase

RNA can form complex structures

RNA enzymes exist (ribozymes)

RNA can control, do logic (riboswitches)

The “RNA world” hypothesis:
1st life was RNA-based

6.5 RNA Secondary Structure

Nussinov's Algorithm – core technology
for RNA structure prediction

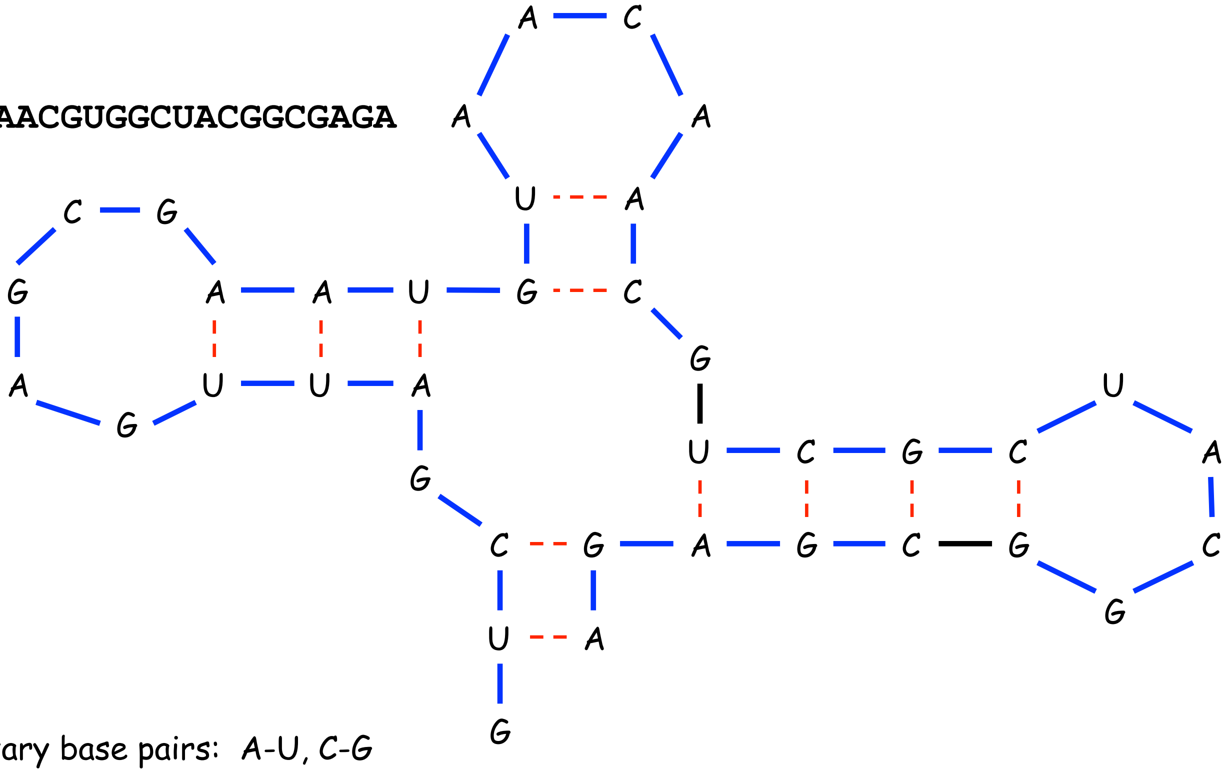
RNA Secondary Structure

RNA. String $B = b_1 b_2 \ldots b_n$ over alphabet { A, C, G, U }.

Secondary structure. RNA is usually single-stranded, and tends to loop back and form base pairs with itself. This structure is essential for understanding behavior of molecule.

Ex: **GUCGAUUGAGCGAAUGUAACAACGUGGCUACGGCGAGA**

— backbone
- - pairing



complementary base pairs: A-U, C-G

RNA Secondary Structure (somewhat oversimplified)

Secondary structure. A set of pairs $S = \{ (b_i, b_j) \}$ that satisfy:

- [Watson-Crick.]
  - S is a *matching* , i.e. each base pairs with at most one other, and
  - each pair in S is a Watson-Crick pair: A-U, U-A, C-G, or G-C.
- [No sharp turns.] The ends of each pair are separated by at least 4 intervening bases. If $(b_i, b_j) \in S$, then $i < j - 4$.
- [Non-crossing.] If (b_i, b_j) and (b_k, b_l) are two pairs in S, then we cannot have $i < k < j < l$. (Violation of this is called a *pseudoknot*.)

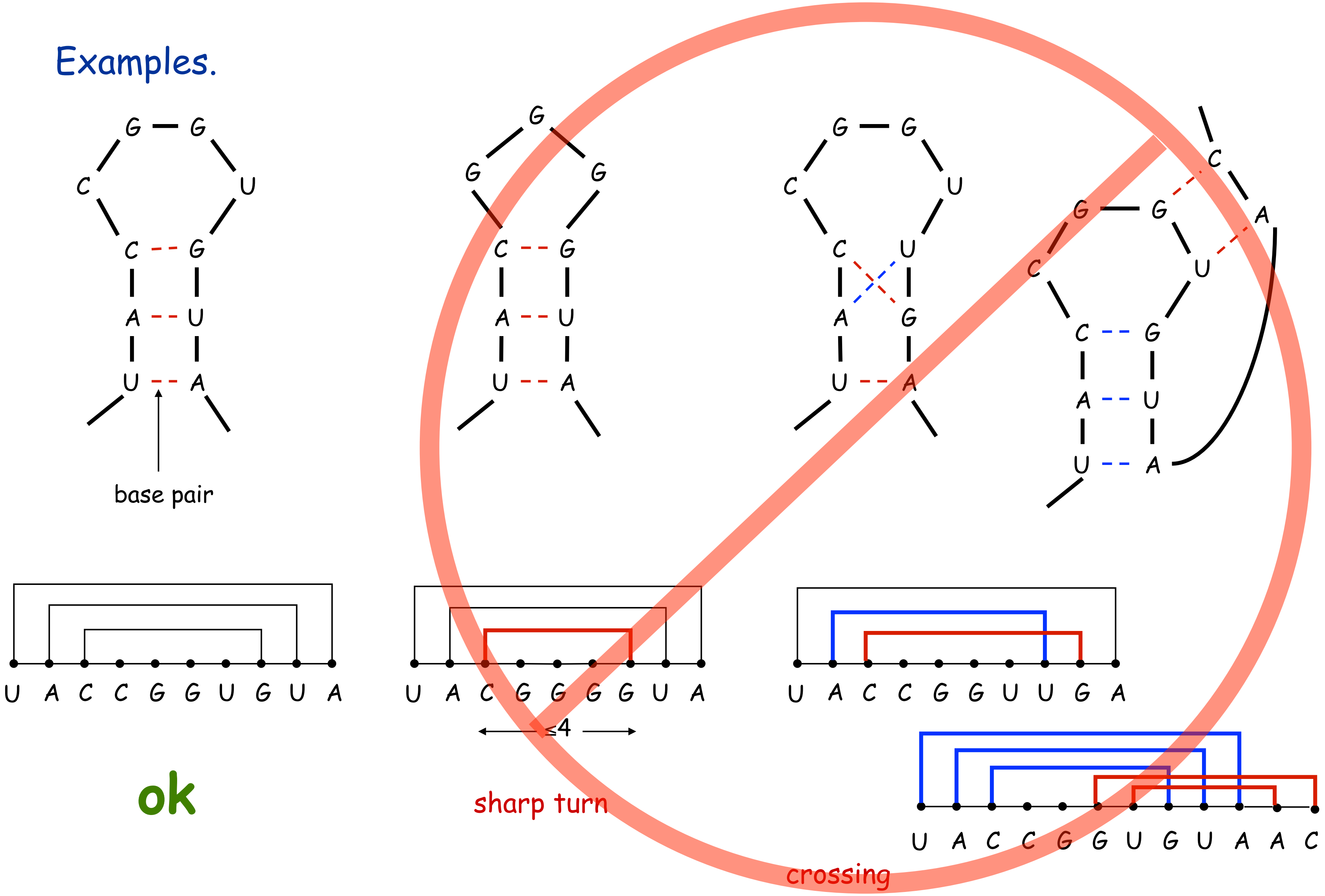
Free energy. Usual hypothesis is that an RNA molecule will form the secondary structure with the optimum total free energy.

↖ approximate by number of base pairs

Goal. Given an RNA molecule $B = b_1 b_2 \ldots b_n$, find a secondary structure S that maximizes the number of base pairs.

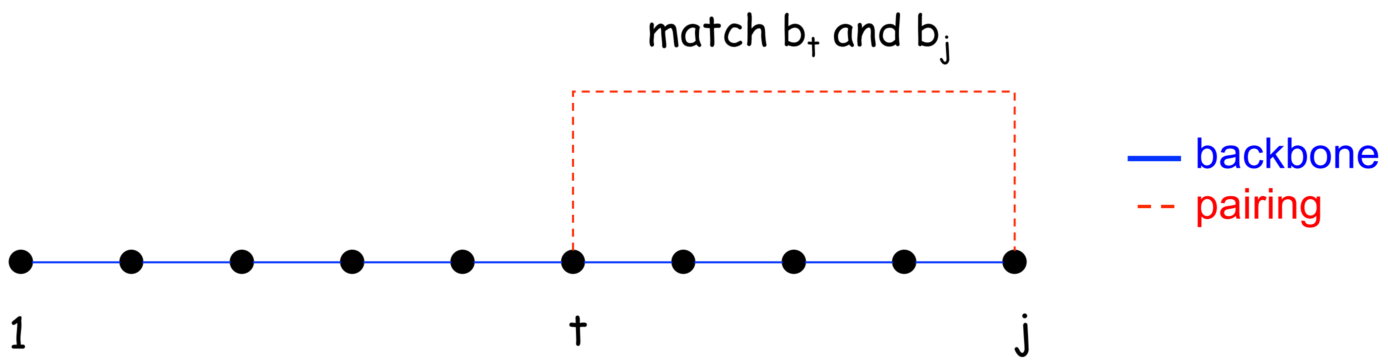
RNA Secondary Structure: Examples

Examples.



RNA Secondary Structure: Subproblems

First attempt. OPT[j] = maximum number of base pairs in a secondary structure of the substring $b_1 b_2 \ldots b_j$.



Difficulty. Results in two sub-problems.

- Finding secondary structure in: $b_1 b_2 \ldots b_{t-1}$. ← OPT(t-1)
- Finding secondary structure in: $b_{t+1} b_{t+2} \ldots b_{j-1}$. ← not OPT of anything; need more sub-problems

Dynamic Programming Over Intervals: (R. Nussinov's algorithm)

Notation. OPT[i, j] = maximum number of base pairs in a secondary structure of the substring $b_i b_{i+1} \dots b_j$.

- Case 1. If $i \geq j - 4$.

  OPT[i, j] = 0 by no-sharp turns condition.

- Case 2. Base b_j is not involved in a pair.

  OPT[i, j] = OPT[i, j-1]

- Case 3. Base b_j pairs with b_t for some $i \leq t < j - 4$.

  non-crossing constraint decouples resulting sub-problems

  $$\mathrm{OPT}[i, j] = 1 + \underset{\uparrow}{\max_t} \{ \mathrm{OPT}[i, t-1] + \mathrm{OPT}[t+1, j-1] \}$$

  take max over t such that $i \leq t < j-4$ and
  b_t and b_j are Watson-Crick complements

Key point:
Either last base is unpaired (case 1,2) or paired (case 3)

Remark. Same core idea in CKY algorithm to parse context-free grammars.

Bottom Up Dynamic Programming Over Intervals

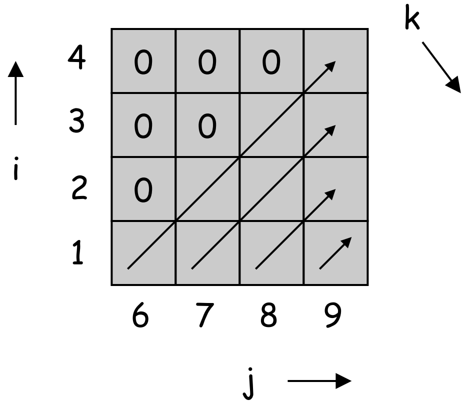
Q. What order to solve the sub-problems?

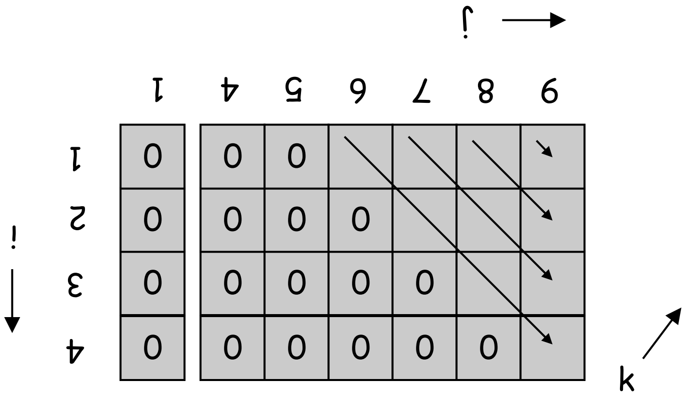
A. Do shortest intervals first.

```
RNA($b_1$,…,$b_n$) {
   for k = 5, 6, …, n-1
      for i = 1, 2, …, n-k
         j = i + k
         Compute OPT[i, j]

   return OPT[1, n]
}
```

using recurrence





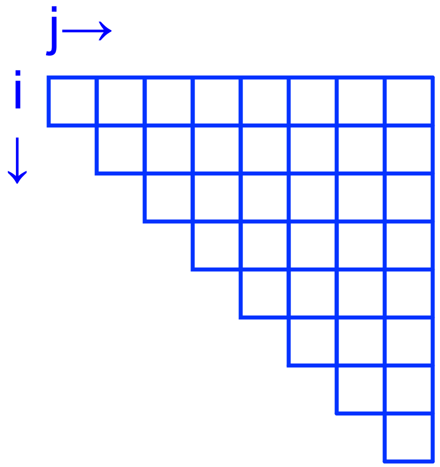
Running time. $O(n^3)$.

Nussinov: Max Pairing

opt(i,j) = # pairs in optimal pairing of $r_i \ldots r_j$

opt(i,j) = 0 for all i, j with $i \geq j-4$; otherwise

opt(i,j) = max of:



$$\left\{ \begin{array}{l} \text{opt}(i,j-1) \\ \max \{ \text{opt}(I,t-1)+1+\text{opt}(t+1,j-1) \mid \\ \qquad\qquad i \leq t < j-4 \text{ and } r_t\text{-}r_j \text{ may pair}\} \end{array} \right.$$

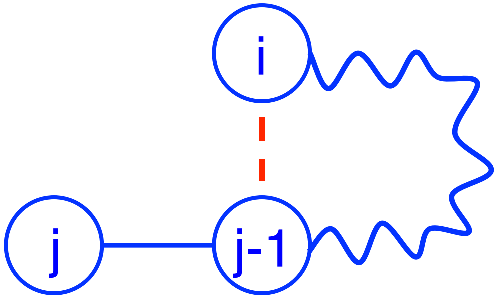
R Nussinov, AB Jacobson, "Fast algorithm for predicting the secondary structure of single-stranded RNA." PNAS 1980.

“Optimal pairing of r_i ... r_j”

Two possibilities

j Unpaired:

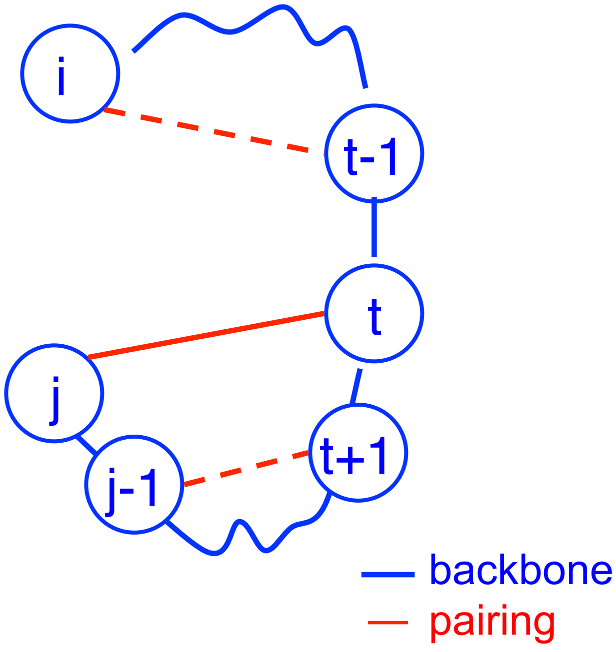
Find best pairing of r_i ... r_{j-1}



j Paired (with some t):

Find best r_i ... r_{t-1} +

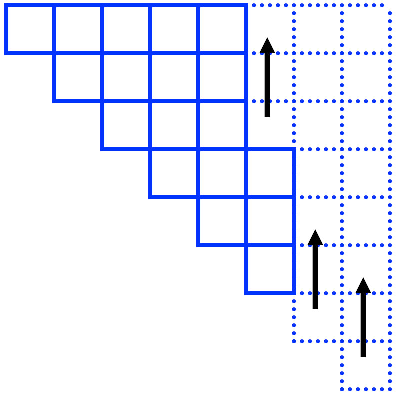
best r_{t+1} ... r_{j-1} plus 1



Why is it slow?

Why do pseudoknots matter?

Nussinov: Computation Order

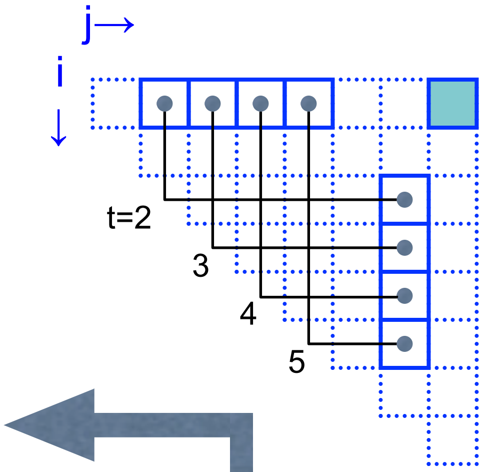


opt(i,j) = # pairs in optimal pairing of $r_i \ldots r_j$

opt(i,j) = 0 for all i, j with $i \geq j-4$; otherwise:

opt(i,j) = max of:

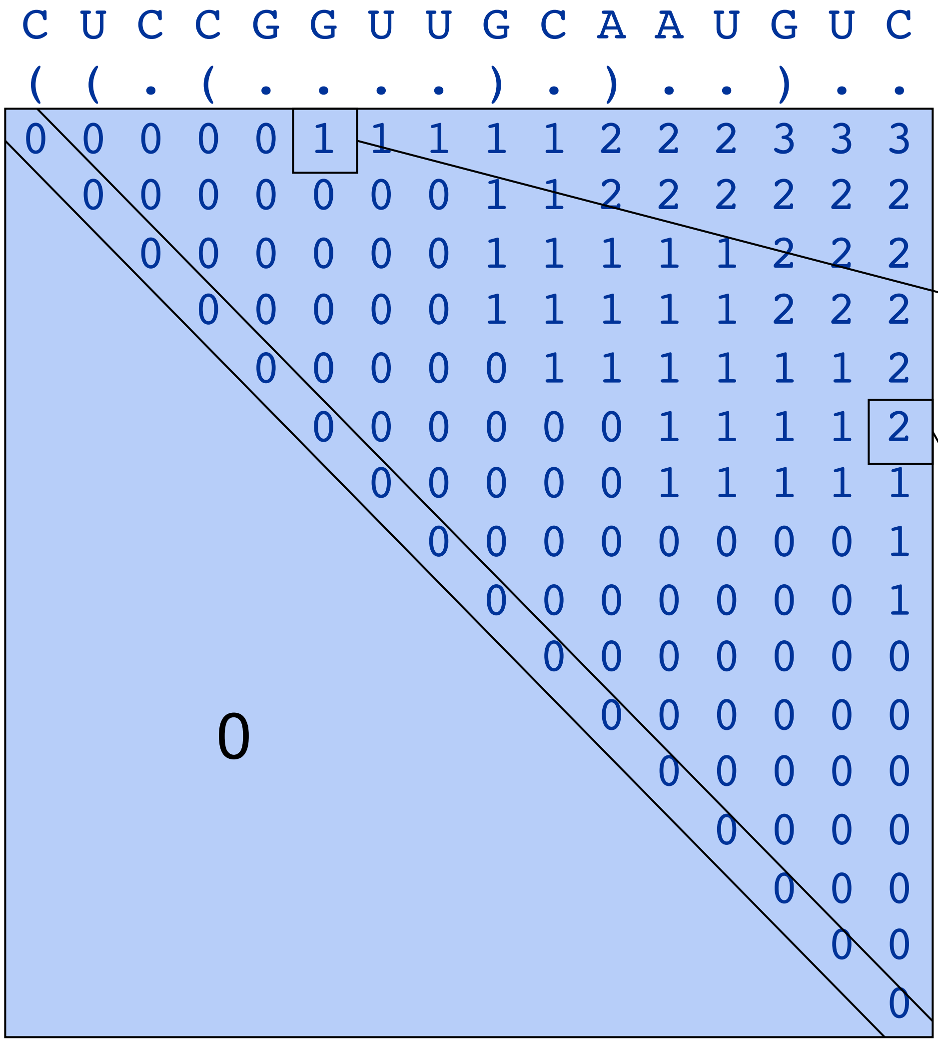
$$\begin{cases} \text{opt}(i,j-1) \\ \max\{\, \text{opt}(I,t-1)+1+\text{opt}(t+1,j-1) \mid i \leq t < j-4 \text{ and } r_t\text{-}r_j \text{ may pair}\} \end{cases}$$



Time: $O(n^3)$

Which Pairs?

Usual dynamic programming “trace-back” tells you *which* base pairs are in the optimal solution, not just how many



C	U	C	C	G	G	U	U	G	C	A	A	U	G	U	C
(	(	.	(	.	.	.	.	)	.	)	.	.	)	.	.
0	0	0	0	0	1	1	1	1	1	2	2	2	3	3	3
	0	0	0	0	0	0	0	1	1	2	2	2	2	2	2
		0	0	0	0	0	0	1	1	1	1	1	2	2	2
			0	0	0	0	0	1	1	1	1	1	2	2	2
				0	0	0	0	0	1	1	1	1	1	1	2
					0	0	0	0	0	0	1	1	1	1	2
						0	0	0	0	0	1	1	1	1	1
							0	0	0	0	0	0	0	0	1
								0	0	0	0	0	0	0	1
									0	0	0	0	0	0	0
			0							0	0	0	0	0	0
											0	0	0	0	0
												0	0	0	0
													0	0	0
														0	0
															0

n = 16

E.g.:
OPT[1,6] = 1:

CUCCGG

(....)

E.g.:
OPT[6,16] = 2:

GUUGCAAUGUC

((....)...)

Computing one cell: OPT[2,18] = ?

n= 20

G	G	G	A	A	A	A	C	C	C	A	A	A	G	G	G	G	U	U	U
(	(	(	.	.	.	.	)	)	)	(	(	(	.	.	.	.	)	)	)
0	0	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3	4	5	6
0	0	0	0	0	0	0	1	2	2	2	2	2	2	3	3	3	4	5	6
0	0	0	0	0	0	0	1	1	1	1	1	1	2	2	3	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	5
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	4	4
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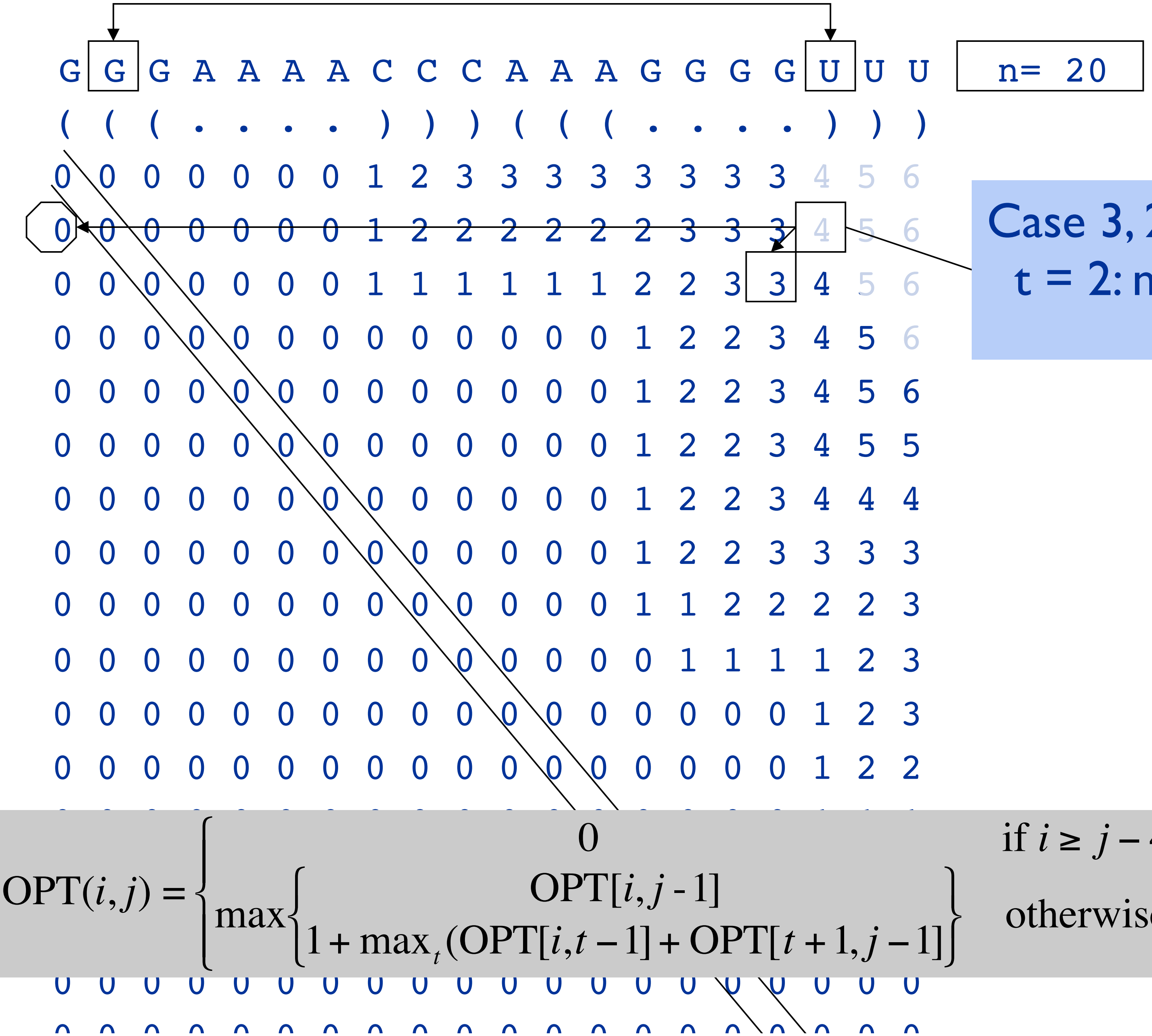
0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Case 1:
2 ≥ 18-4? no.
Case 2:
B_{18} unpaired?
Always a possibility;
then OPT[2,18] ≥ 3

GGAAAACCCAAAGGGGU
((....))(....)...

$$\mathrm{OPT}(i,j) = \begin{cases} 0 & \text{if } i \ge j-4 \\ \max\left\{ \begin{array}{c} \mathrm{OPT}[i,j-1] \\ 1+\max_t(\mathrm{OPT}[i,t-1]+\mathrm{OPT}[t+1,j-1]) \end{array} \right\} & \text{otherwise} \end{cases}$$

Computing one cell: OPT[2,18] = ?



n= 20

G	G	G	A	A	A	A	C	C	C	A	A	A	G	G	G	G	U	U	U
(	(	(	.	.	.	.	)	)	)	(	(	(	.	.	.	.	)	)	)
0	0	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3	4	5	6
0	0	0	0	0	0	0	1	2	2	2	2	2	2	3	3	3	4	5	6
0	0	0	0	0	0	0	1	1	1	1	1	1	2	2	3	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	5
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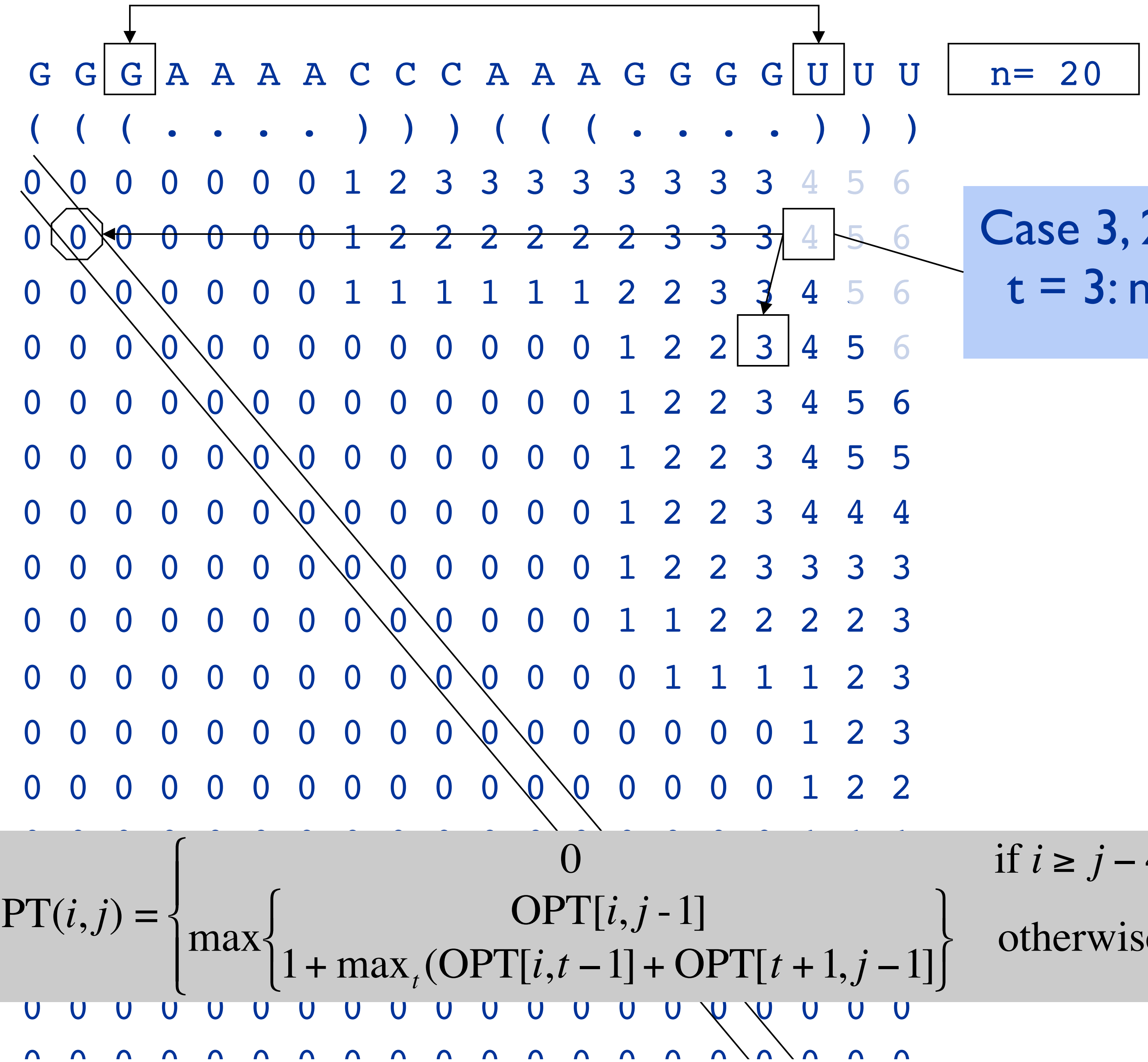
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	3	3	3
0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	2	2	2	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Case 3, 2 ≤ t <18-4:
t = 2: no pair

$$\mathrm{OPT}(i,j) = \begin{cases} 0 & \text{if } i \geq j-4 \\ \max\begin{Bmatrix} \mathrm{OPT}[i,j\text{ - }1] \\ 1+\max_t(\mathrm{OPT}[i,t-1]+\mathrm{OPT}[t+1,j-1]) \end{Bmatrix} & \text{otherwise} \end{cases}$$

Computing one cell: OPT[2,18] = ?



n= 20

G	G	G	A	A	A	A	C	C	C	A	A	A	G	G	G	G	U	U	U
(	(	(	.	.	.	.	)	)	)	(	(	(	.	.	.	.	)	)	)
0	0	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3	4	5	6
0	0	0	0	0	0	0	1	2	2	2	2	2	2	3	3	3	4	5	6
0	0	0	0	0	0	0	1	1	1	1	1	1	2	2	3	3	4	5	6
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0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
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0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	4	4
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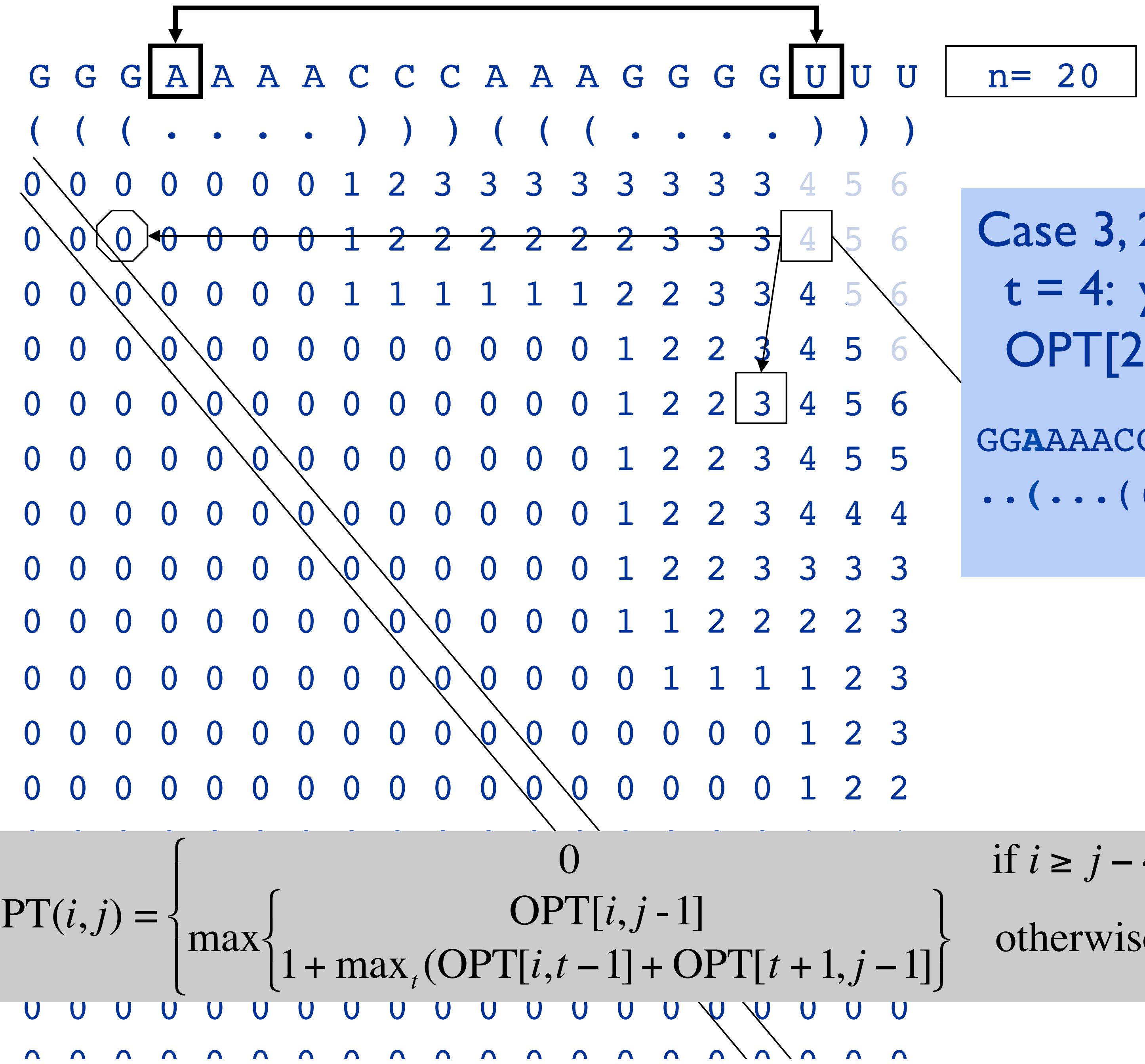
0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	2	2	2	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Case 3, 2 ≤ t <18-4:
t = 3: no pair

$$\mathrm{OPT}(i,j) = \begin{cases} 0 & \text{if } i \geq j-4 \\ \max\begin{Bmatrix} \mathrm{OPT}[i,j-1] \\ 1+\max_t(\mathrm{OPT}[i,t-1]+\mathrm{OPT}[t+1,j-1]) \end{Bmatrix} & \text{otherwise} \end{cases}$$

Computing one cell: OPT[2,18] = ?



n= 20

G	G	G	**A**	A	A	A	C	C	C	A	A	A	G	G	G	G	**U**	U	U
(	(	(	.	.	.	.	)	)	)	(	(	(	.	.	.	.	)	)	)
0	0	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3	4	5	6
0	0	0	0	0	0	0	1	2	2	2	2	2	2	3	3	3	4	5	6
0	0	0	0	0	0	0	1	1	1	1	1	1	2	2	3	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	5
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	4	4
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	3	3	3
0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	2	2	2	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Case 3, 2 ≤ t <18-4:

t = 4: yes pair

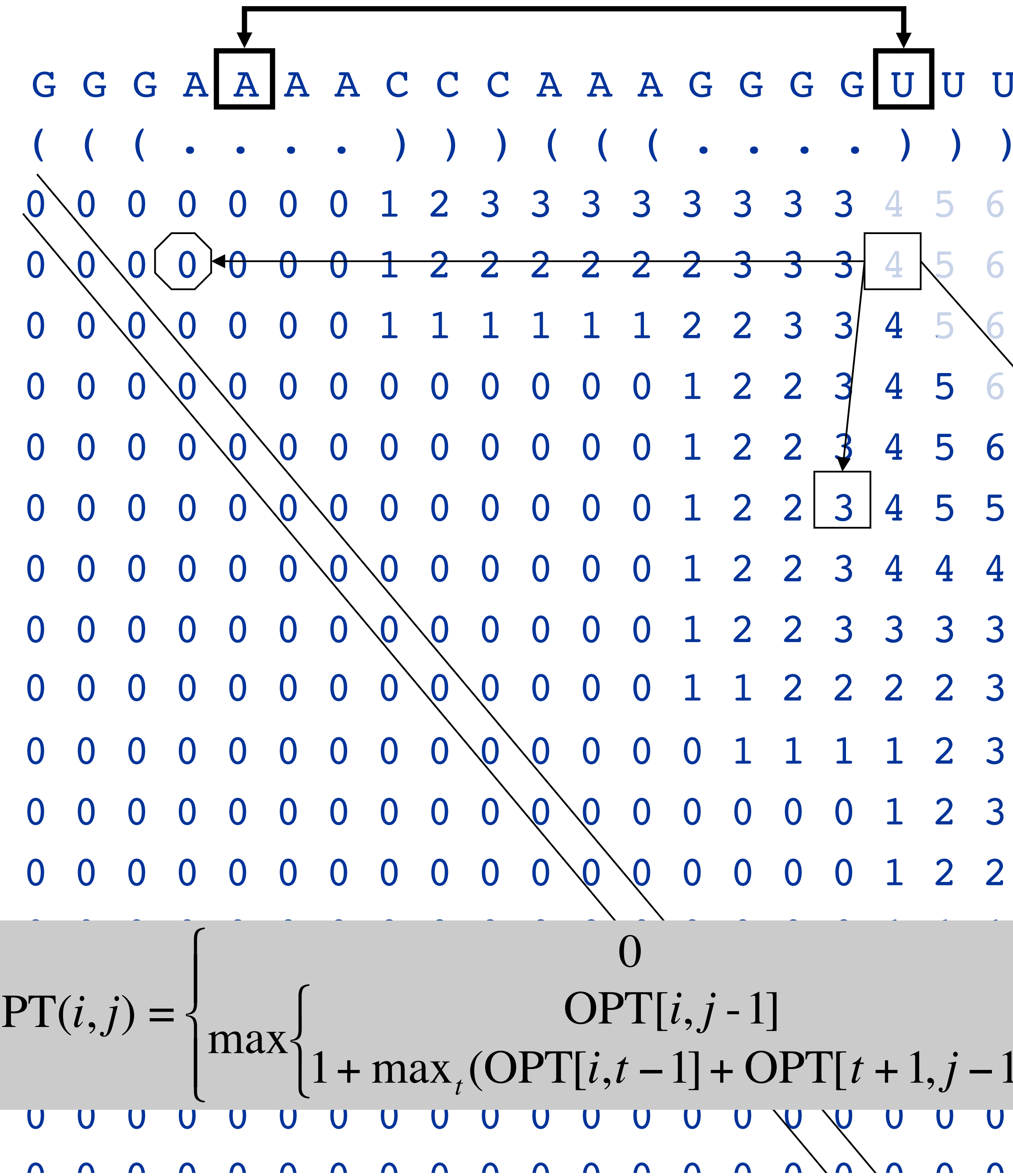
OPT[2,18]≥1+0+3

GG**A**AAACCCAAAGGGG**U**

..(....(((....))))

$$\mathrm{OPT}(i,j) = \begin{cases} 0 & \text{if } i \geq j-4 \\ \max\left\{ \begin{array}{c} \mathrm{OPT}[i,j\text{ - }1] \\ 1+\max_t(\mathrm{OPT}[i,t-1]+\mathrm{OPT}[t+1,j-1]) \end{array} \right\} & \text{otherwise} \end{cases}$$

Computing one cell: OPT[2,18] = ?



n= 20

G	G	G	A	A	A	A	C	C	C	A	A	A	G	G	G	G	U	U	U
(	(	(	.	.	.	.	)	)	)	(	(	(	.	.	.	.	)	)	)
0	0	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3	4	5	6
0	0	0	0	0	0	0	1	2	2	2	2	2	2	3	3	3	4	5	6
0	0	0	0	0	0	0	1	1	1	1	1	1	2	2	3	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	5
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	4	4
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	3	3	3
0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	2	2	2	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Case 3, 2 ≤ t < 18-4:

t = 5: yes pair

OPT[2,18]≥1+0+3

GGA**A**AACCCAAAGGGG**U**

...(..((((....))))

$$\mathrm{OPT}(i,j) = \begin{cases} 0 & \text{if } i \ge j-4 \\ \max\begin{Bmatrix} \mathrm{OPT}[i,j-1] \\ 1+\max_t(\mathrm{OPT}[i,t-1]+\mathrm{OPT}[t+1,j-1]) \end{Bmatrix} & \text{otherwise} \end{cases}$$

Computing one cell: OPT[2,18] = ?

n= 20

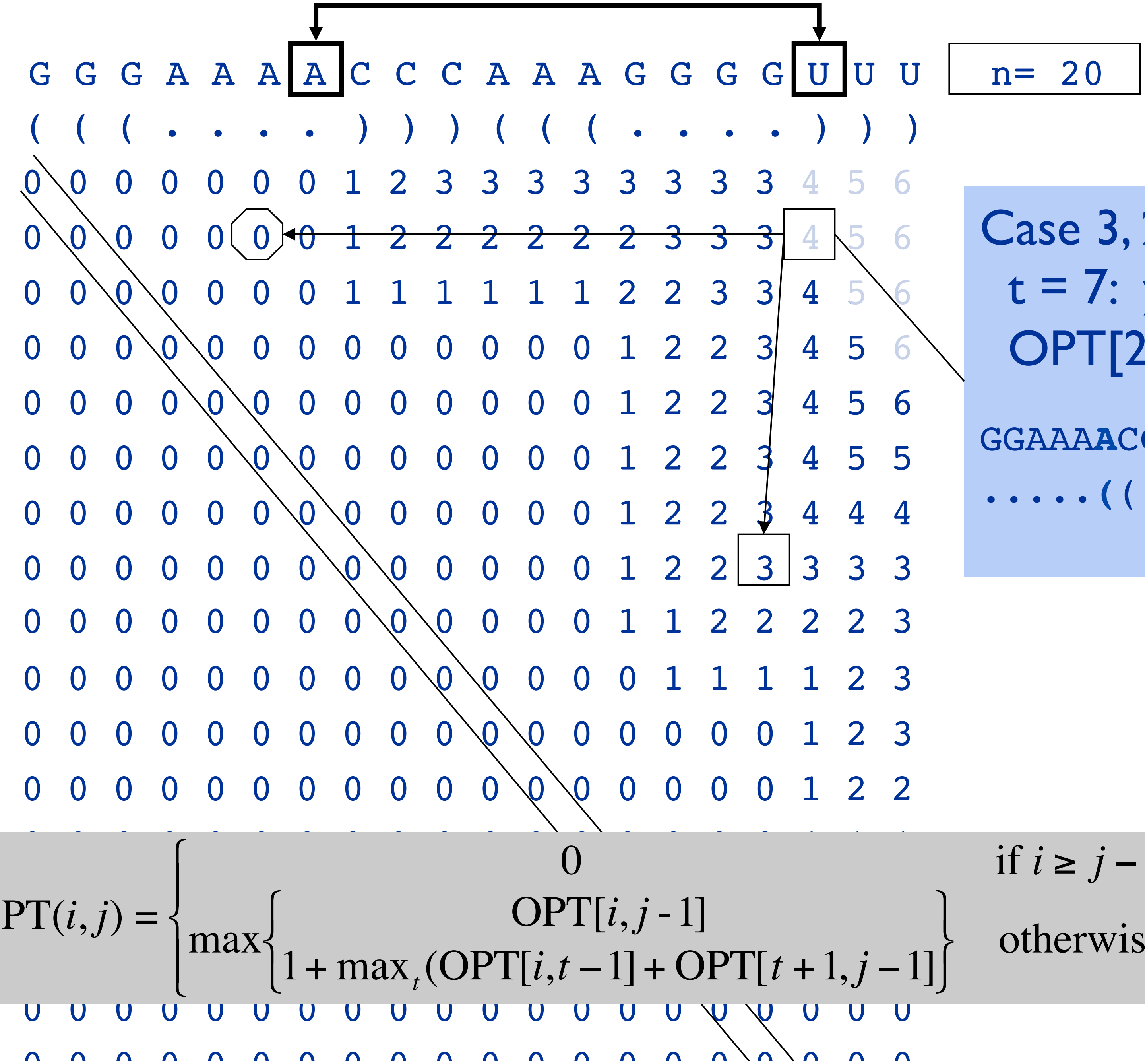
G	G	G	A	A	A	A	C	C	C	A	A	A	G	G	G	G	U	U	U
(	(	(	.	.	.	.	)	)	)	(	(	(	.	.	.	.	)	)	)
0	0	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3	4	5	6
0	0	0	0	0	0	0	1	2	2	2	2	2	2	3	3	3	4	5	6
0	0	0	0	0	0	0	1	1	1	1	1	1	2	2	3	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	5
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	4	4
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	3	3	3
0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	2	2	2	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Case 3, 2 ≤ t <18-4:
t = 6: yes pair
OPT[2,18]≥1+0+3

GGAA**A**ACCCAAAGGGG**U**
....(.(((....))))

$$\mathrm{OPT}(i,j)=\begin{cases}0 & \text{if } i \geq j-4\\ \max\begin{Bmatrix}\mathrm{OPT}[i,j-1]\\ 1+\max_t(\mathrm{OPT}[i,t-1]+\mathrm{OPT}[t+1,j-1])\end{Bmatrix} & \text{otherwise}\end{cases}$$

Computing one cell: OPT[2,18] = ?



n= 20

G	G	G	A	A	A	A	C	C	C	A	A	A	G	G	G	G	U	U	U
(	(	(	.	.	.	.	)	)	)	(	(	(	.	.	.	.	)	)	)
0	0	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3	4	5	6
0	0	0	0	0	0	0	1	2	2	2	2	2	2	3	3	3	4	5	6
0	0	0	0	0	0	0	1	1	1	1	1	1	2	2	3	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	5
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	4	4
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	3	3	3
0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	2	2	2	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Case 3, 2 ≤ t <18-4:

t = 7: yes pair

OPT[2,18]≥1+0+3

GGAAA**A**CCCAAAGGGG**U**

.....((((....))))

$$\mathrm{OPT}(i,j)=\begin{cases}0 & \text{if } i\ge j-4\\ \max\begin{Bmatrix}\mathrm{OPT}[i,j-1]\\ 1+\max_t(\mathrm{OPT}[i,t-1]+\mathrm{OPT}[t+1,j-1])\end{Bmatrix} & \text{otherwise}\end{cases}$$

Computing one cell: OPT[2,18] = ?

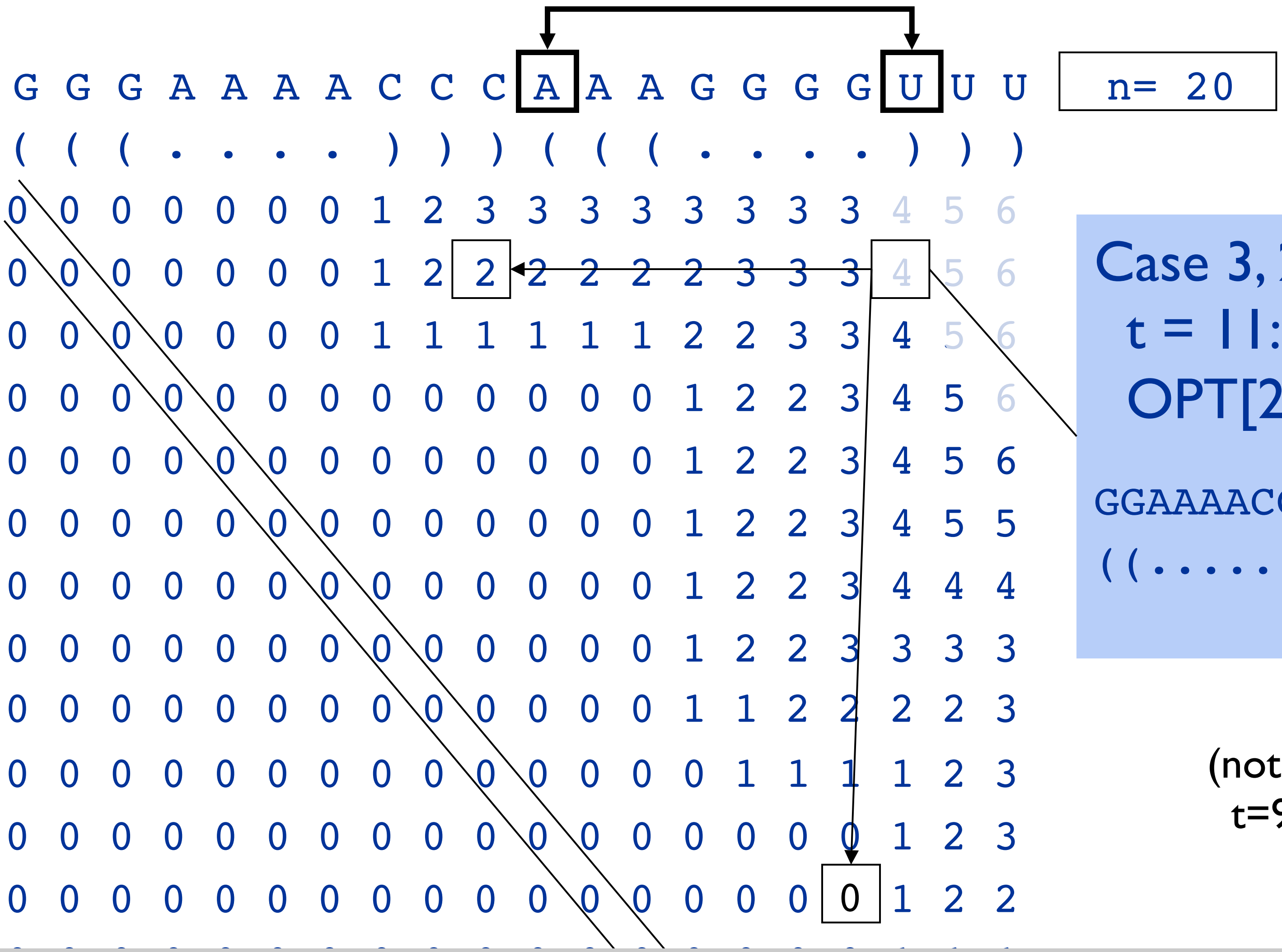
n= 20

G	G	G	A	A	A	A	C	C	C	A	A	A	G	G	G	G	U	U	U
(	(	(	.	.	.	.	)	)	)	(	(	(	.	.	.	.	)	)	)
0	0	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3	4	5	6
0	0	0	0	0	0	0	1	2	2	2	2	2	2	3	3	3	4	5	6
0	0	0	0	0	0	0	1	1	1	1	1	1	2	2	3	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	5
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	4	4
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	3	3	3
0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	2	2	2	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Case 3, 2 ≤ t <18-4:
t = 8: no pair

$$\mathrm{OPT}(i,j) = \begin{cases} 0 & \text{if } i \geq j-4 \\ \max\left\{ \begin{array}{c} \mathrm{OPT}[i,j-1] \\ 1+\max_t(\mathrm{OPT}[i,t-1]+\mathrm{OPT}[t+1,j-1]) \end{array} \right\} & \text{otherwise} \end{cases}$$

Computing one cell: OPT[2,18] = ?



n= 20

G	G	G	A	A	A	A	C	C	C	A	A	A	G	G	G	G	U	U	U
(	(	(	.	.	.	.	)	)	)	(	(	(	.	.	.	.	)	)	)
0	0	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3	4	5	6
0	0	0	0	0	0	0	1	2	2	2	2	2	2	3	3	3	4	5	6
0	0	0	0	0	0	0	1	1	1	1	1	1	2	2	3	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	5
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	4	4
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	3	3	3
0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	2	2	2	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Case 3, 2 ≤ t <18-4:

t = 11: yes pair

OPT[2,18]≥1+2+0

GGAAAACCC**A**AAGGGG**U**

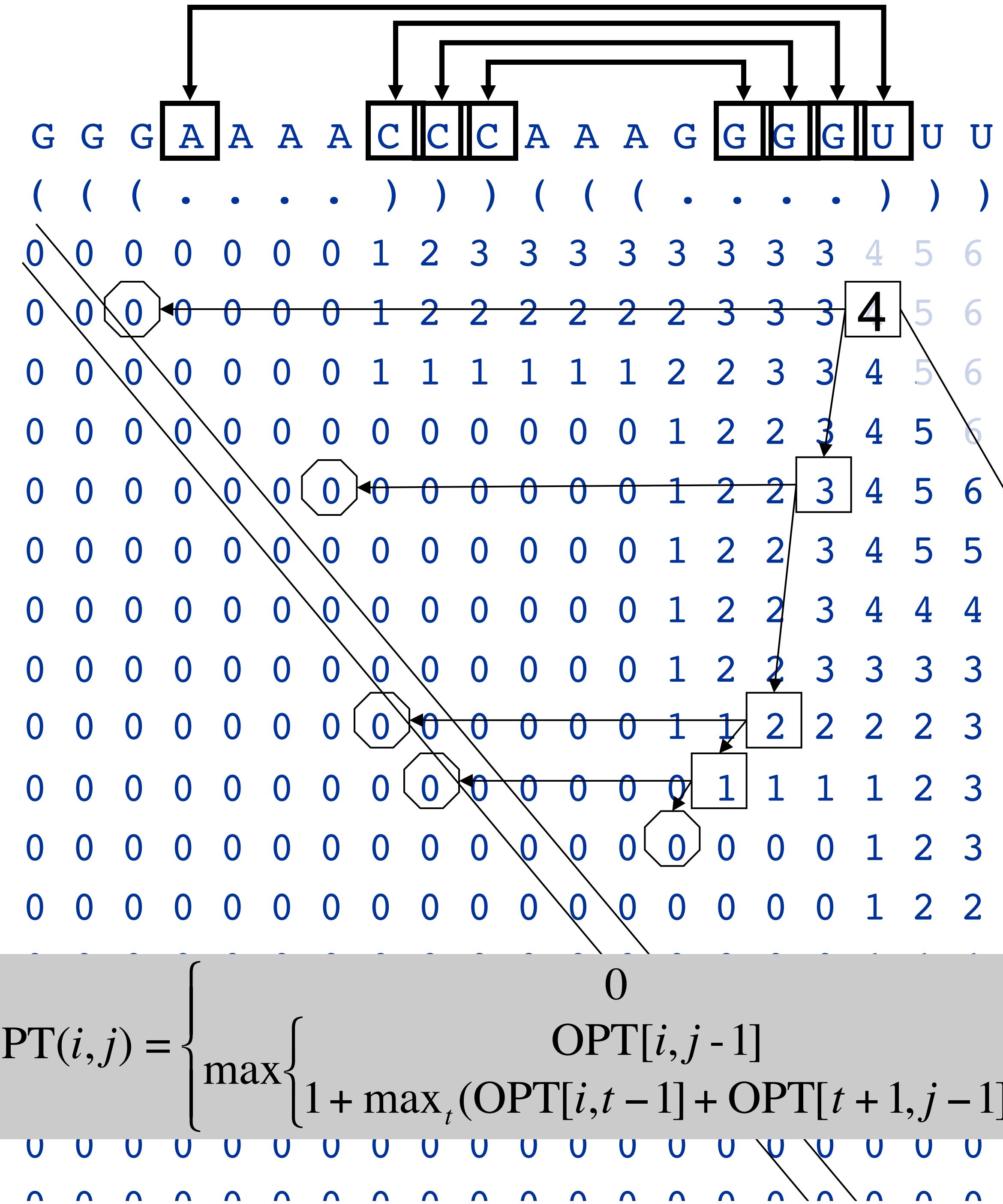
((.....))(......)

(not shown:
t=9,10, 12,13)

$$\mathrm{OPT}(i,j) = \begin{cases} 0 & \text{if } i \geq j-4 \\ \max\begin{Bmatrix} \mathrm{OPT}[i,j-1] \\ 1+\max_t(\mathrm{OPT}[i,t-1]+\mathrm{OPT}[t+1,j-1]) \end{Bmatrix} & \text{otherwise} \end{cases}$$

Computing one cell: OPT[2,18] = 4

n= 20



G	G	G	A	A	A	A	C	C	C	A	A	A	G	G	G	G	U	U	U
(	(	(	.	.	.	.	)	)	)	(	(	(	.	.	.	.	)	)	)
0	0	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3	4	5	6
0	0	0	0	0	0	0	1	2	2	2	2	2	2	3	3	3	4	5	6
0	0	0	0	0	0	0	1	1	1	1	1	1	2	2	3	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	6
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	5	5
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	4	4	4
0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2	3	3	3	3
0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	2	2	2	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	3
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	2	2
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Overall, Max = 4
several ways, e.g.:

GGAAAACCCAAAGGGGU

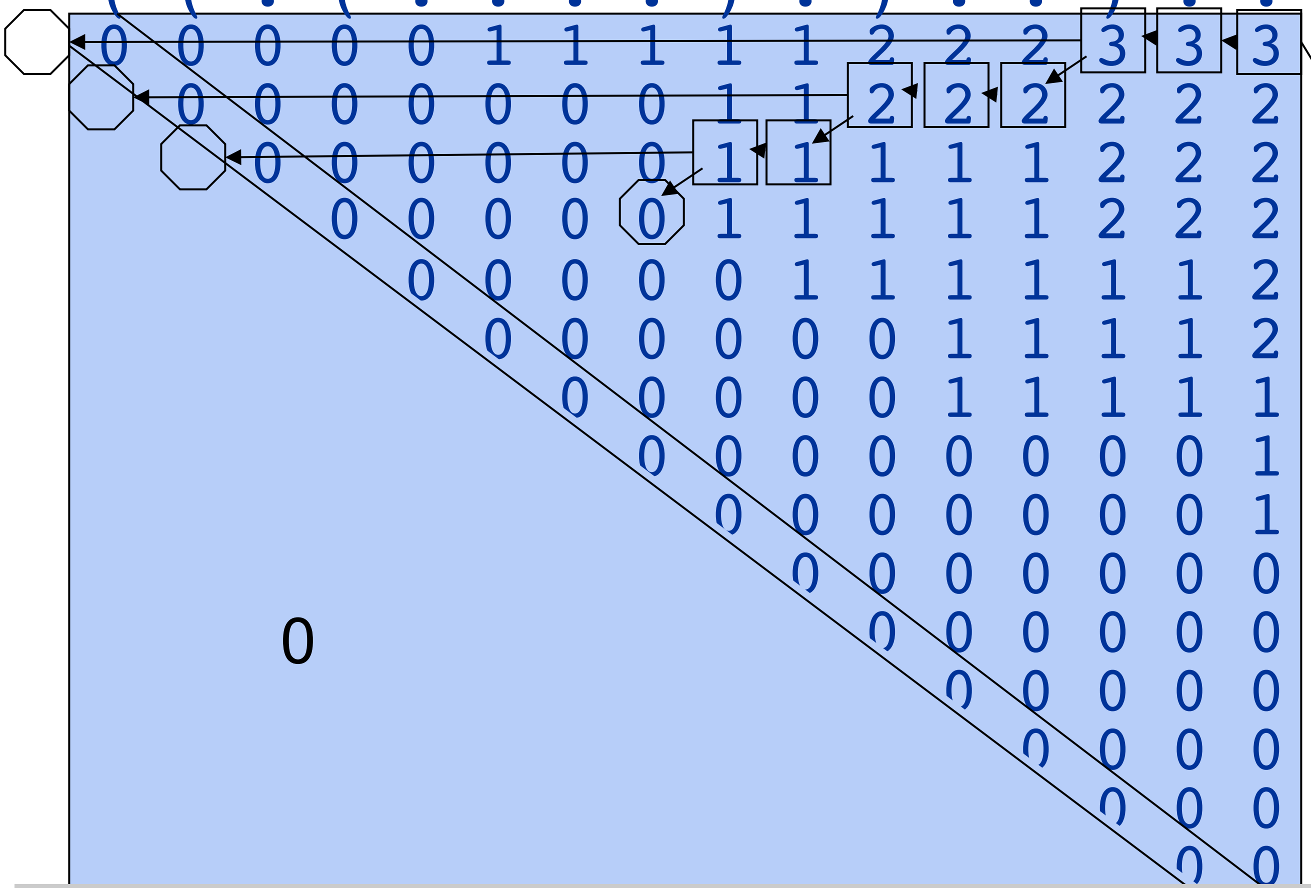
..(...(((....))))

tree shows trace back:
square = case 3
octagon = case 1

$$\text{OPT}(i,j)=\begin{cases}0 & \text{if } i\ge j-4\\ \max\begin{cases}\text{OPT}[i,j-1]\\ 1+\max_t(\text{OPT}[i,t-1]+\text{OPT}[t+1,j-1])\end{cases} & \text{otherwise}\end{cases}$$

Another Trace Back Example

n = 16



C	U	C	C	G	G	U	U	G	C	A	A	U	G	U	C
(	(	.	(	.	.	.	.	)	.	)	.	.	)	.	.
0	0	0	0	0	1	1	1	1	1	2	2	2	3	3	3
	0	0	0	0	0	0	0	1	1	2	2	2	2	2	2
		0	0	0	0	0	0	1	1	1	1	1	2	2	2
			0	0	0	0	0	1	1	1	1	1	2	2	2
				0	0	0	0	0	1	1	1	1	1	1	2
					0	0	0	0	0	0	1	1	1	1	2
						0	0	0	0	0	1	1	1	1	1
							0	0	0	0	0	0	0	0	1
								0	0	0	0	0	0	0	1
									0	0	0	0	0	0	0
										0	0	0	0	0	0
											0	0	0	0	0
												0	0	0	0
													0	0	0
														0	0
															0

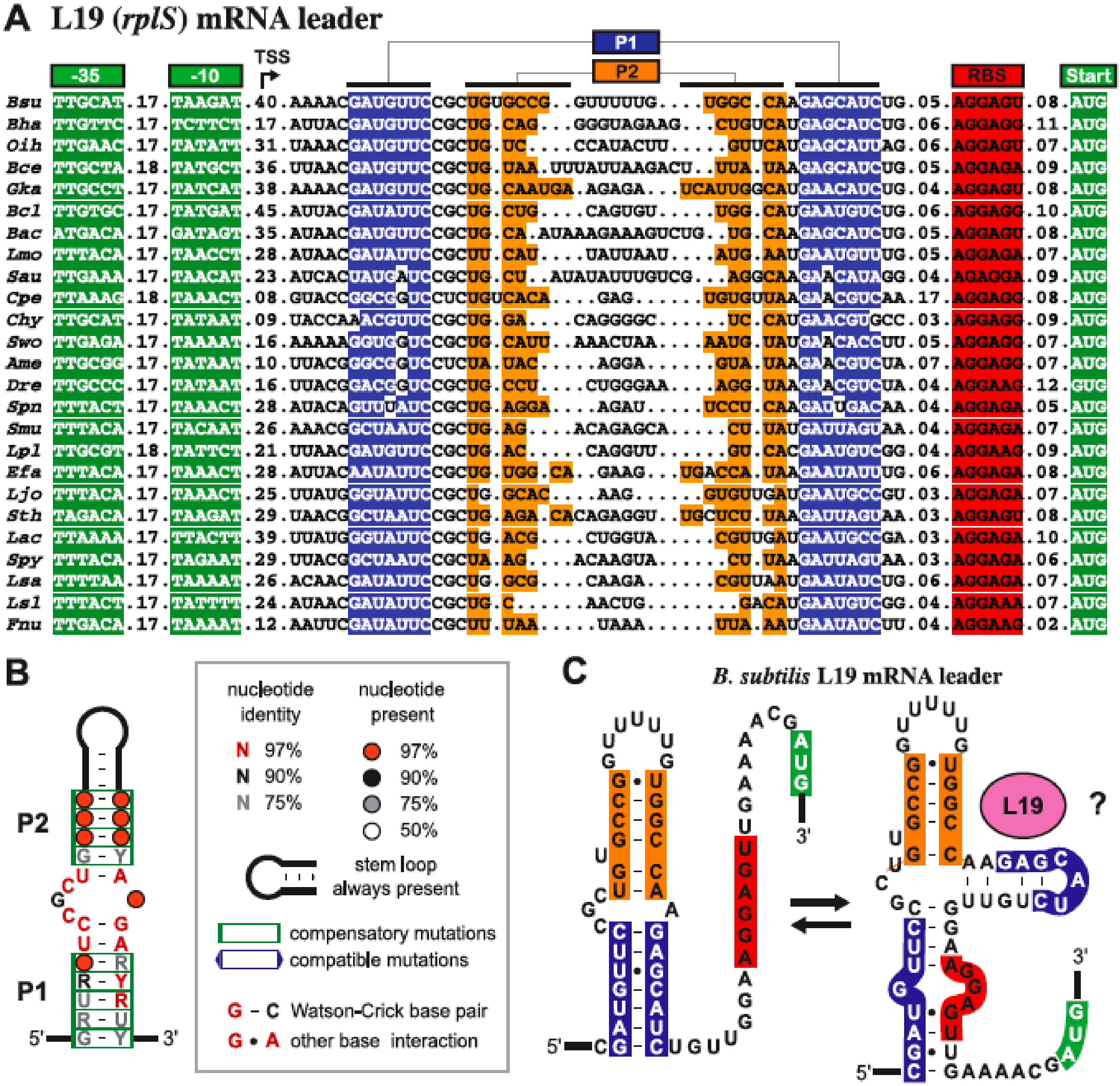
0

E.g.:
OPT[1,16] = 3:

`CUCCGGUUGCAAUGUC`

`((.(....).)..)..`

$$\mathrm{OPT}(i,j)=\begin{cases}0 & \text{if } i\ge j-4\\ \max\left\{\begin{matrix}\mathrm{OPT}[i,j-1]\\ 1+\max_t(\mathrm{OPT}[i,t-1]+\mathrm{OPT}[t+1,j-1])\end{matrix}\right\} & \text{otherwise}\end{cases}$$



A L19 (*rplS*) mRNA leader

```
    -35       -10     TSS                      P1   P2                           RBS       Start
Bsu TTGCAT.17.TAAGAT.40.AAAACGAUGUUCCGCUGUGCCG..GUUUUUG....UGGC.CAAGAGCAUCUG.05.AGGAGU.08.AUG
Bha TTGTTC.17.TCTTCT.17.AUUACGAUGUUCCGCUG.CAG...GGGUAGAAG...CUGUCAUGAGCAUCUG.06.AGGAGG.11.AUG
Oih TTGAAC.17.TATATT.31.UAAACGAUGUUCCGCUG.UC....CCAUACUU.....GUUCAUGAGCAUUAG.06.AGGAGU.07.AUG
Bce TTGCTA.18.TATGCT.36.UUAACGAUGUUCCGCUG.UAA.UUUAUUAAGACU..UUA.UAAGAGCAUCUG.05.AGGAGA.09.AUG
Gka TTGCCT.17.TATCAT.38.AAAACGAUGUUCCGCUG.CAAUGA.AGAGA...UCAUUGGCAUGAACAUCUG.04.AGGAGU.08.AUG
Bcl TTGTGC.17.TATGAT.45.AUUACGAUAUUCCGCUG.CUG....CAGUGU.....UGG.CAUGAAUGUCUG.06.AGGAGG.10.AUG
Bac ATGACA.17.GATAGT.35.AUAACGAUGUUCCGCUG.CA.AUAAAGAAAGUCUG..UG.CAAGAGCAUCUG.05.AGGAGU.08.AUG
Lmo TTTACA.17.TAACCT.28.AUAACGAUAUUCCGCUU.CAU....UAUUAAU....AUG.AAUGAAUGUUUG.05.AGGAGA.07.AUG
Sau TTGAAA.17.TAACAT.23.AUCACUAUGAUCCGCUG.CU..AUAUAUUUGUCG...AGGCAAGAACAUAGG.04.AGAGGA.09.AUG
Cpe TTAAAG.18.TAAACT.08.GUACCGGCCGUCCUCUGUCACA....GAG......UGUGUUAAGAACGUCAA.17.AGGAGG.08.AUG
Chy TTGCAT.17.TATAAT.09.UACCAAACGUUCCGCUG.GA....CAGGGGC......UC.CAUGAACGUGCC.03.AGGAGG.09.AUG
Swo TTGAGA.17.TAAAAT.16.AAAAAGGUCGUCCGCUG.CAUU..AAACUAA....AAUG.UAUGAACACCUU.05.AGGAGG.07.AUG
Ame TTGCGG.17.TATAAT.10.UUACGGGCCGUCCUCUA.UAC.....AGGA......GUA.UAAGAACGUCUA.07.AGGAGG.07.AUG
Dre TTGCCC.17.TATAAT.16.UUACGGACGGUCCGCUG.CCU....CUGGGAA....AGG.UAAGAACGUCUA.04.AGGAAG.12.GUG
Spn TTTACT.17.TAAACT.28.AUACAGUUUAUCCGCUG.AGGA....AGAU.....UCCU.CAAGAUUGACAA.04.AGGAGA.05.AUG
Smu TTTACA.17.TACAAT.26.AAACGGCUAAUCCGCUG.AG....ACAGAGCA.....CU.UAUGAUUAGUAA.04.AGGAGA.07.AUG
Lpl TTGCGT.18.TATTCT.21.UUAACGAUGUUCCGCUG.AC.....CAGGUU......GU.CACGAAUGUCGG.04.AGGAAG.09.AUG
Efa TTTACA.17.TAAACT.28.AUUACAAUAUUCCGCUG.UGG.CA..GAAG...UGACCA.UAAGAAUAUUUG.06.AGGAGA.08.AUG
Ljo TTTACA.17.TAAACT.25.UUAUGGGUAUUCCGCUG.GCAC....AAG......GUGUUGAUGAAUGCCGU.03.AGGAGA.07.AUG
Sth TAGACA.17.TAAGAT.29.UAACGGCUAAUCCGCUG.AGA.CACAGAGGU..UGCUCU.UAAGAUUAGUAA.03.AGGAGU.08.AUG
Lac TTAAAA.17.TTACTT.39.UUAUGGGUAUUCCGCUG.ACG....CUGGUA.....CGUUGAUGAAUGCCGA.03.AGGAGA.10.AUG
Spy TTTACA.17.TAGAAT.29.UUACGGCUAAUCCGCUA.AG....ACAAGUA......CU.UAAGAUUAGUAA.03.AGGAGA.06.AUG
Lsa TTTTAA.17.TAAAAT.26.ACAACGAUAUUCCGCUG.GCG....CAAGA......CGUUAAUGAAUAUCUG.06.AGGAGA.07.AUG
Lsl TTTACT.17.TATTTT.24.AUAACGAUAUUCCGCUG.C.......AACUG.........GACAUGAAUGUCGG.04.AGGAAA.07.AUG
Fnu TTGACA.17.TAAAAT.12.AAUUCGAUAUUCCGCUU.UAA.....UAAA.........UUA.AAUGAAUAUCUU.04.AGGAAG.02.AUG
```

B

nucleotide identity
N 97%
N 90%
N 75%

nucleotide present
● 97%
● 90%
● 75%
○ 50%

stem loop always present

compensatory mutations

compatible mutations

G - C Watson-Crick base pair

G • A other base interaction

C *B. subtilis* L19 mRNA leader

Figure 3. Putative Autoregulatory Structure in L19 mRNA Leaders