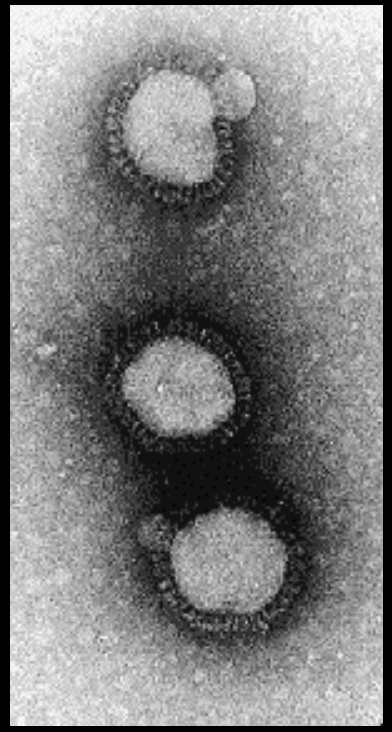
第9回ウイルス学キャンプ in 湯河原
2012年7月10−11日

ウイルス研究が育んだ現代生命科学

筑波大学医学医療系・永田恭介

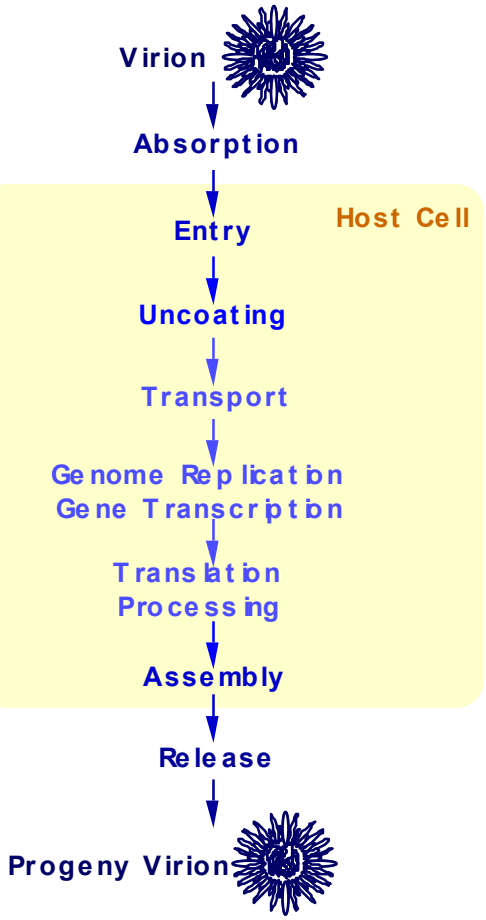
ウイルス学／ウイルス研究とは？

- ウイルスの増殖と病原性の機構を理解して、
  ウイルスによる疾病を制御する
- ウイルスを探針として、
  生物の原理を理解する
- ウイルスを改変して、
  新たなバイオテクノロジーを開発する



インフルエンザウイルス

ウイルスというものは、
ゲノムとしてDNAかRNAを持つ有機化合物の複合体



従って、ウイルスの複製には、
ウイルスのみならず宿主細胞由来の因子が必須

ウイルス学研究から生まれた主なノーベル賞

2008 **Harald zur Hausen,** for his discovery of human papilloma viruses causing cervical cancer (The Nobel Prize in Physiology or Medicine)
Françoise Barré-Sinoussi, Luc Montagnier, for their discovery of human immunodeficiency virus (The Nobel Prize in Physiology or Medicine)

2006 **Andrew Z. Fire, Craig C. Mello**, for their discovery of RNA interference - gene silencing by double-stranded RNA (The Nobel Prize in Physiology or Medicine)

1999 **Günter Blobel**, for the discovery that proteins have intrinsic signals that govern their transport and localization in the cell (The Nobel Prize in Physiology or Medicine)

1997 **Stanley B. Prusiner**, for his discovery of Prions - a new biological principle of infection (The Nobel Prize in Physiology or Medicine)

1996 **Peter C. Doherty, Rolf M. Zinkernagel**, for their discoveries concerning the specificity of the cell mediated immune defence (The Nobel Prize in Physiology or Medicine)

1993 **Richard J. Roberts, Phillip A. Sharp**, for their discoveries of split genes (The Nobel Prize in Physiology or Medicine)

1989 **J. Michael Bishop, Harold E. Varmus**, for their discovery of the cellular origin of retroviral oncogenes (The Nobel Prize in Physiology or Medicine)

1989 **Baruj Benacerraf, Jean Dausset, George D. Snell**, for their discoveries concerning genetically determined structures on the cell surface that regulate immunological reactions (The Nobel Prize in Physiology or Medicine)
Paul Berg, for his fundamental studies of the biochemistry of nucleic acids, with particular regard to recombinant-DNA (The Nobel Prize in Chemistry)
Walter Gilbert, Frederick Sanger, for their contributions concerning the determination of base sequences in nucleic acids (The Nobel Prize in Chemistry)

1978 **Werner Arber, Daniel Nathans, Hamilton O. Smith**, for the discovery of restriction enzymes and their application to problems of molecular genetics (The Nobel Prize in Physiology or Medicine)

1976 **Baruch S. Blumberg, D. Carleton Gajdusek**, for their discoveries concerning new mechanisms for the origin and dissemination of infectious diseases (The Nobel Prize in Physiology or Medicine)

1975 **David Baltimore, Renato Dulbecco, Howard Martin Temin**, for their discoveries concerning the interaction between tumour viruses and the genetic material of the cell (The Nobel Prize in Physiology or Medicine)

1969 **Max Delbru"ck, Alfred D. Hershey, Salvador E. Luria**, for their discoveries concerning the replication mechanism and the genetic structure of viruses (The Nobel Prize in Physiology or Medicine)

1966 **Peyton Rous**, for his discovery of tumour-inducing viruses (The Nobel Prize in Physiology or Medicine)

1965 **Franc,ois Jacob, Andre' Lwoff, Jacques Monod**, for their discoveries concerning genetic control of enzyme and virus synthesis (The Nobel Prize in Physiology or Medicine)

1962 **Francis Harry Compton Crick, James Dewey Watson, Maurice Hugh Frederick Wilkins**, for their discoveries concerning the molecular structure of nucleic acids and its significance for information transfer in living materia (Nobel Prize in Physiology or Medicine)

1958 **Joshua Lederberg**, for his discoveries concerning genetic recombination and the organization of the genetic material of bacteria (Nobel Prize in Physiology or Medicine)
George Wells Beadle, Edward Lawrie Tatum, for their discovery that genes act by regulating definite chemical events (Nobel Prize in Physiology or Medicine)

1954 **John Franklin Enders, Thomas Huckle Weller, Frederick Chapman Robbins**, for their discovery of the ability of poliomyelitis viruses to grow in cultures of various types of tissue (Nobel Prize in Physiology or Medicine)

1951 **Max Theiler**, for his discoveries concerning yellow fever and how to combat it (Nobel Prize in Physiology or Medicine)

1946 **John Howard Northrop, Wendell Meredith Stanley**, for their preparation of enzymes and virus proteins in a pure form (The Nobel Prize in Chemistry)

Methods from Virus Research

Brakke (1951, 1953)
sucrose density gradient for virus isolation

Watson & Crick (+Franklin & Wilkins) (1953)
DNA double helix

Harris & Watkins (1965)
virus -mediated cell fusion to produce heterokaryon

Graham & van der Eb (1973)
DNA transfection

Holmes & Klug (1977)
3D structure of TMV

Sanger (1977)
dideoxy sequencing method

Maxam & Gilbert (1977)
chemical sequencing method

DNA transfection

A question was whether adenovirus DNA itself is infectious.

A new technique for assaying infectivity of adenovirus 5 DNA has been developed. Viral DNA was diluted in isotonic saline containing phosphate at a low concentration, and calcium chloride was added, resulting in the formation of a calcium phosphate precipitate. DNA coprecipitated with the calcium phosphate and, when the resulting suspension was added to human KB cell monolayers, became adsorbed to the cells. Following adsorption, uptake of DNA into the cells occurred during an incubation in liquid medium at 37 °C in the continued presence of extra calcium chloride. For adenovirus 5 DNA the assay resulted in up to 100-fold more plaques than could be obtained using DEAE-dextran. Furthermore a reproducible relationship between amounts of DNA inoculated per culture and numbers of plaques produced was demonstrated. The assay was most efficient at high DNA concentrations (10–30 μg/ml); below this range the addition of carrier DNA was necessary for optimum results.

In addition to adenovirus 5 DNA, the technique has been used successfully to assay infectivity of DNA from adenovirus 1 and simian virus 40.

A new technique for the assay of infectivity of human adenovirus 5 DNA
Graham F L, van der Eb A J. Virology, 52 :456-467, 1973.

Maxam & Gilbert method -chemical sequencing method-

DNA binding of the lac repressor.
Riggs A D, Bourgeois S, Newby R F, Cohn M.
J. Mol. Biol., 34: 365-368, 1968.

Catabolite repression of the lac operon. Effect of mutations in the lac promoter.
Yudkin M D.
Biochem J., 118: 741-746, 1970.

A question was what is the lac operator.

Proc. Nat. Acad. Sci. USA
Vol. 70, No. 12, Part I, pp. 3581–3584, December 1973

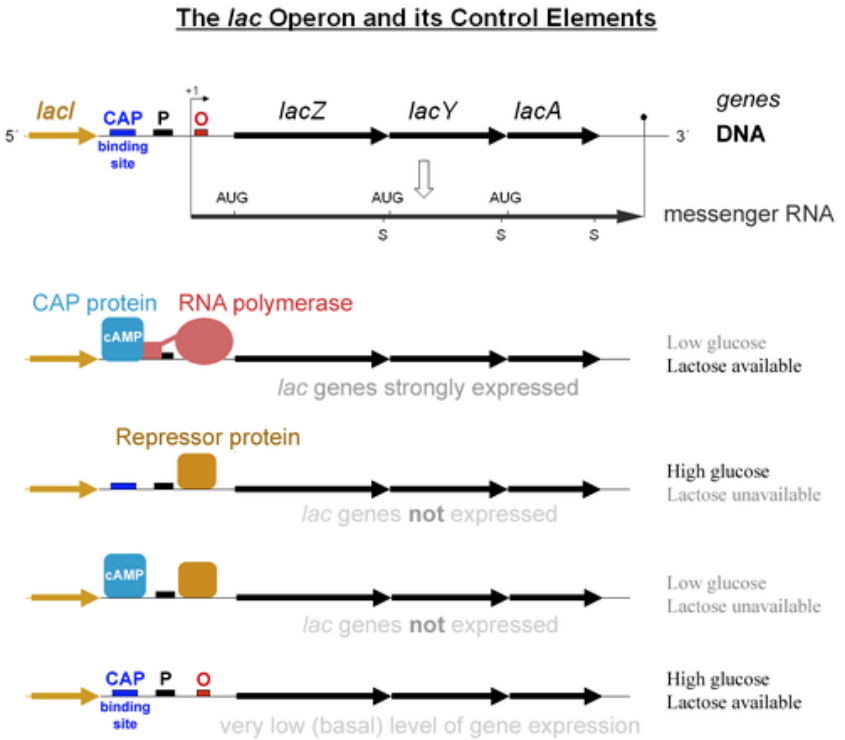
The Nucleotide Sequence of the *lac* Operator

(regulation/protein–nucleic acid interaction/DNA–RNA sequencing/oligonucleotide priming)

WALTER GILBERT AND ALLAN MAXAM

Department of Biochemistry and Molecular Biology, Harvard University, Cambridge, Massachusetts 02138

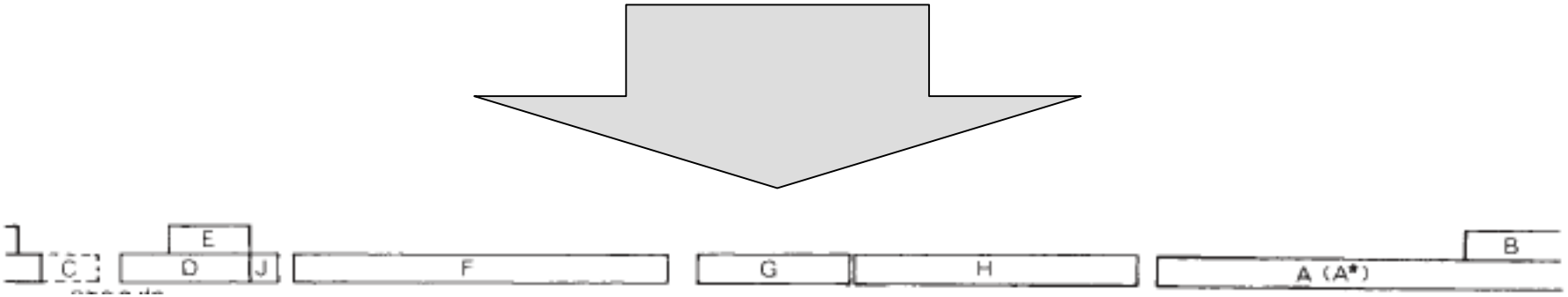
Communicated by J. D. Watson, August 9, 1973



Sanger method -dideoxy sequencing method-

A question was why nucleotide numbers do not correspond to amino acid numbers in ϕX174.

A rapid method for determining sequences in DNA by primed synthesis with DNA polymerase.
Sanger F, Coulson A R. J. Mol. Biol., 94: 441-448, 1975.



Nature Vol. 265 February 24 1977

articles

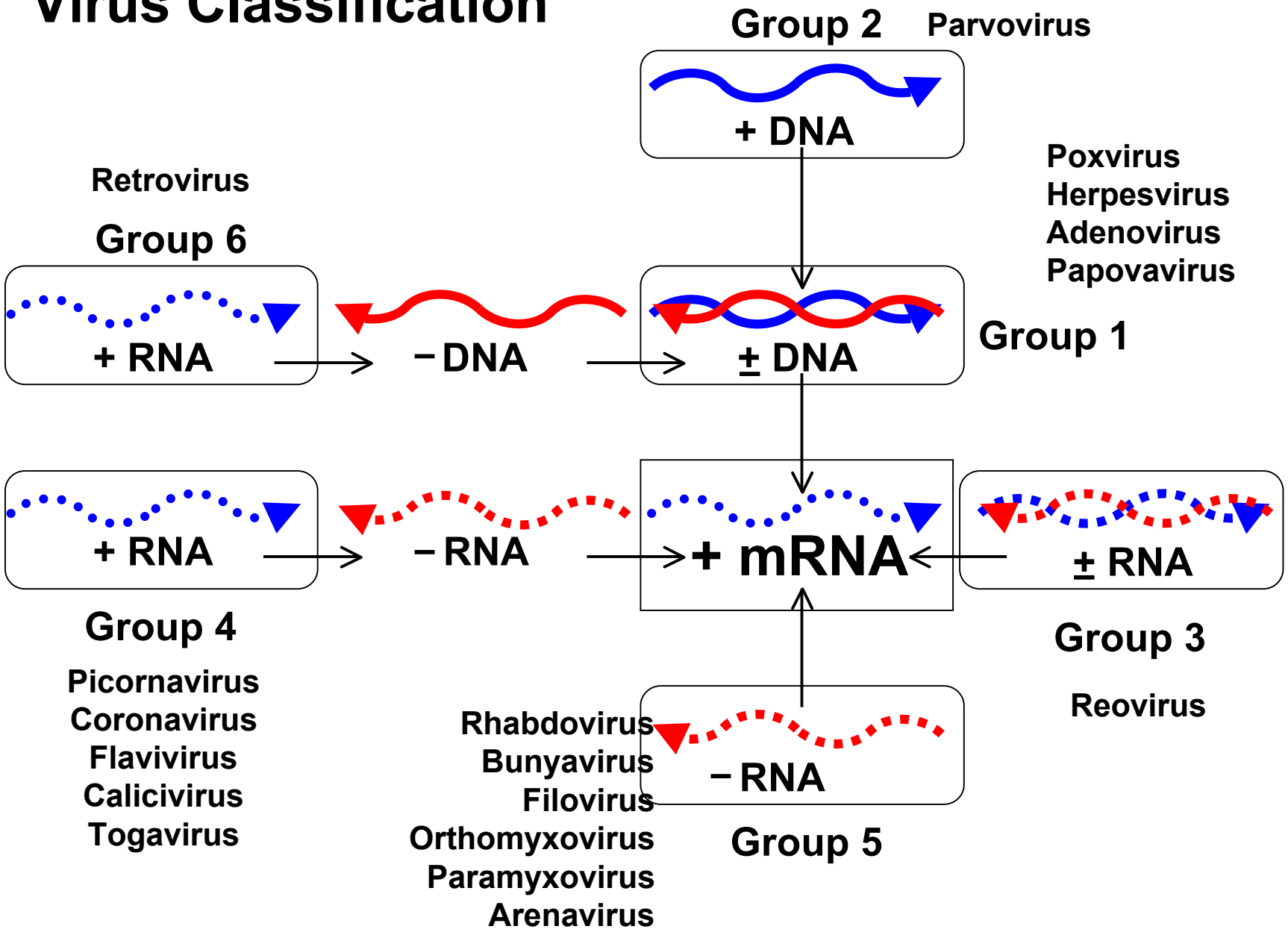
Nucleotide sequence of bacteriophage ΦX174 DNA

F. Sanger, G. M. Air*, B. G. Barrell, N. L. Brown†, A. R. Coulson, J. C. Fiddes, C. A. Hutchison III‡, P. M. Slocombe§ & M. Smith‖

MRC Laboratory of Molecular Biology, Hills Road, Cambridge CB2 2QH, UK

A DNA sequence for the genome of bacteriophage ΦX174 of approximately 5,375 nucleotides has been determined using the rapid and simple 'plus and minus' method. The sequence identifies many of the features responsible for the production of the proteins of the nine known genes of the organism, including initiation and termination sites for the proteins and RNAs. Two pairs of genes are coded by the same region of DNA using different reading frames.

Virus Classification



David Baltimoreのポリメレース遍歴

Poliovirus-induced RNA polymerase and the effects of virus-specific inhibitors on its production.
Baltimore D, Eggers H J, Franklin R M, Tamm I.
Proc. Natl. Acad. Sci. U S A., 49: 843-849, 1963.

In vitro synthesis of viral RNA by the poliovirus RNA polymerase.
Baltimore D.
Proc. Natl. Acad. Sci. U S A., 51: 450-456, 1964.

Ribonucleic acid synthesis of vesicular stomatitis virus. I. Species of ribonucleic acid found in Chinese hamster ovary cells infected with plaque-forming and defective particles.
Stampfer M, Baltimore D, Huang A S.
J. Virol., : 154-161, 1969.

Ribonucleic acid synthesis of vesicular stomatitis virus, II. An RNA polymerase in the virion.
Baltimore D, Huang A S, Stampfer M.
Proc. Natl. Acad. Sci. U S A., 66: 572-576, 1970.

NATURE VOL. 226 JUNE 27 1970

Viral RNA-dependent DNA Polymerase

Two independent groups of investigators have found evidence of an enzyme in virions of RNA tumour viruses which synthesizes DNA from an RNA template. This discovery, if upheld, will have important implications not only for carcinogenesis by RNA viruses but also for the general understanding of genetic transcription: apparently the classical process of information transfer from DNA to RNA can be inverted.

Table 3. EFFECT OF RIBONUCLEASE ON THE DNA POLYMERASE ACTIVITY OF RAUSCHER MOUSE LEUKAEMIA VIRUS

Conditions	pmoles ^{3}H-TMP incorporation
No preincubation	2·50
Preincubated with no addition	2·20
Preincubated with 20 μg/ml. ribonuclease	0·69
Preincubated with 50 μg/ml. ribonuclease	0·31
Preincubated with 200 μg/ml. ribonuclease	0·08
Preincubated with no addition	3·09
Preincubated with 50 μg/ml. ribonuclease	0·52
Preincubated with 50 μg/ml. lysozyme	3·67
Preincubated with 50 μg/ml. cytochrome *c*	3·97

In experiment 1, for the preincubation, 15 μg of viral protein in 5 μl. of solution was added to 45 μl. of water at 4° C containing the indicated amounts of enzyme. After incubation for 30 min at 22° C, the samples were chilled and 50 μl. of a 2-fold concentrated standard reaction mixture was added. The samples were then incubated at 37° C for 45 min and acid-insoluble radioactivity was measured. In experiment 2, the same procedure was followed, except that the preincubation was for 20 min at 22° C and the 37° C incubation was for 60 min.

日本人も貢献、ポリメレースの歴史

Autocatalytic synthesis of a viral RNA in vitro.
Haruna I, Spiegelman S.
Science, 150: 884-886, 1965.

A search for an intermediate involving a complement during synchronous synthesis by a purified RNA replicase.
Haruna I, Spiegelman S.
Proc. Natl. Acad. Sci. U S A., 55: 1256-1263, 1966

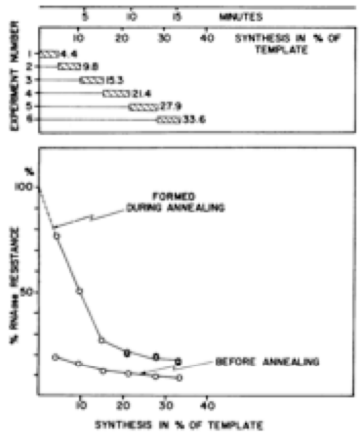
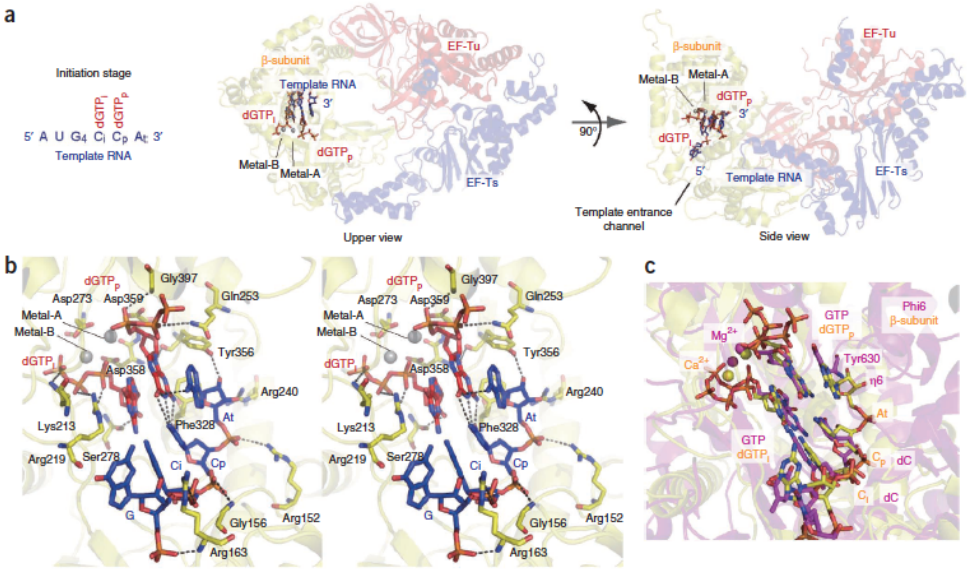


FIG. 4.—Complementarity of product to template at various periods of synthesis. Complete numerical and experimental details are given in Table 2, its legend, and in *Methods*. The amount of resistance due to annealing is obtained by subtracting the resistance observed prior to annealing from that measured after the annealing. This is necessary since the initial RNase resistance of early product was shown to be insensitive to heat denaturation and due principally to its high content of adenine. The upper portion gives the time plan of the six parallel syntheses, the *hatched areas* indicating the interval of labeling with UTP^{32} and the numbers representing synthesis finally achieved in % of template. The last three samples were challenged with 40 γ (*open circles*) and 20 γ (*closed circles*) of unlabeled Qβ-RNA.



Molecular basis for RNA polymerization by Qβ replicase.
Takeshita D, Tomita K.
Nat. Struct. Mol. Biol., 19: 229-237, 2012.

To Understand the Central Dogma

DNA Replication
- **adenovirus**
  - Kelly (1979) first *in vitro* eukaryotic DNA replication system
  - Hurwitz (1979, 1980), Stillman (1981)
- **SV40**
  - Kelly (1984) *in vitro* cellular type DNA replication system
  - Hurwitz (1985), Stillman (1985)

Transcription
- **adenovirus**
  - Roeder (1979) first *in vitro* eukaryotic transcription system
  - Matsui (1980), Chambon (1980), Sharp (1982)

Reverse Transcriptase
- **mouse leukemia virus, Rous sarcoma virus**
  - Baltimore, Temin (1970) finding of RT

Splicing
- **adenovirus**
  - Sharp, Roberts, Chambon (1977) finding of splicing

Cap Structure
- **cytoplasmic polyhedrosis virus**
  - Miura (1975) finding of cap structure

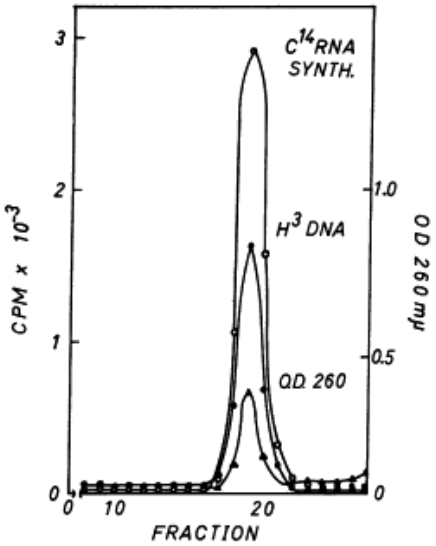
IRES
- **poliovirus, encephalomyocarditis virus**
  - Sonenberg, Wimmer (1988) cap-independent translation

ちなみに、　DNA-dependent RNA polymerases

Poxvirus DNA-dependent RNA polymerase.
Kates J R, McAuslan B R.
Proc. Natl. Acad. Sci. U S A., 58: 134-141, 1967.

RNA polymerase activity in purified infectious vaccinia virus.
Munyon W, Paoletti E, Grace J T Jr.
Proc. Natl. Acad. Sci. U S A., 58: 2280-2287, 1967.

FIG. 1.—Cosedimentation of RP cores and of the capacity to synthesize RNA *in vitro*. RP cores were purified as described in *Materials and Methods* using H^3-thymidine-labeled virus. The procedure employed for sucrose density gradient centrifugation is described in the section on *Materials and Methods*. Fractions of 1 ml were collected from a pin hole at the bottom of the tube and the optical density at 260 mμ was determined. Aliquots of 0.3 ml from each fraction were tested for their ability to synthesize RNA *in vitro* (see *Materials and Methods*). C^{14}-GTP incorporated into RNA *in vitro* (○); H^3-thymine in RP core DNA (●); optical density at 260 mμ (▲).

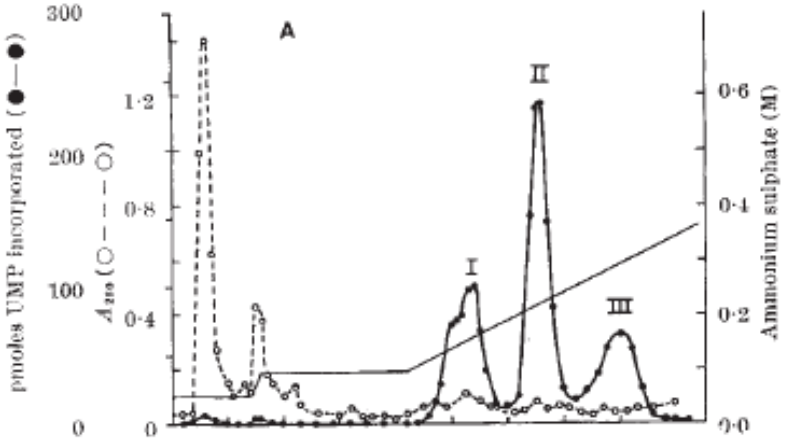


Multiple forms of DNA-dependent RNA polymerase in eukaryotic organisms.
Roeder R G, Rutter W J.
Nature, 224: 234-237, 1969.

Multiple Forms of DNA-dependent RNA Polymerase in Eukaryotic Organisms

by
ROBERT G. ROEDER*
WILLIAM J. RUTTER†
Department of Biochemistry, University of Washington, Seattle 98105

Three distinct RNA polymerase activities have been isolated from developing sea urchin embryos. In rat liver nuclei there are two RNA polymerase activities. One polymerase (I) is probably localized in the nucleolus and one (II) in the nucleoplasm.



翻訳では・・

Cap structure

A blocked structure at the 5' terminus of mRNA from cytoplasmic polyhedrosis virus.
Furuichi Y, Miura K.
Nature, 253: 374-375, 1975.

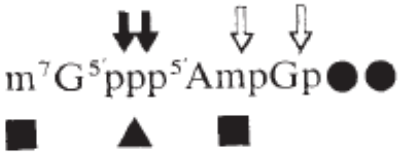
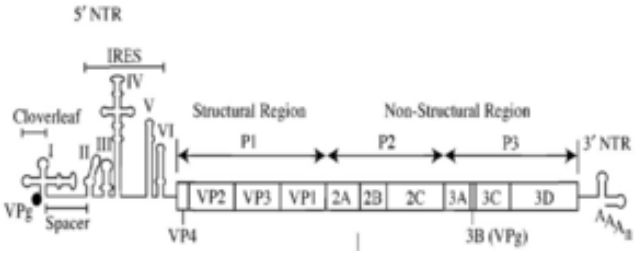


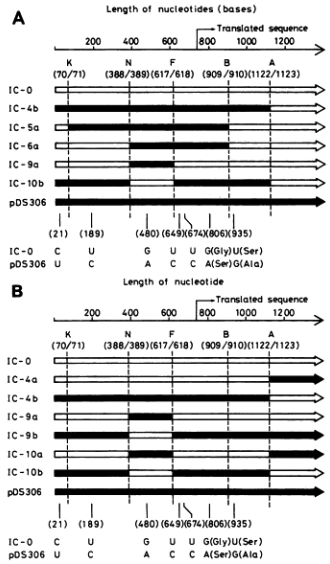
Fig. 3 A proposed structure of the 5′ terminal part of CPV mRNA and the enzyme-attacking sites. Radioactive components: (■, ^{3}H-methyl; ▲, ^{32}P). White arrow, attacking of *Penicillium* nuclease; black arrow, attacking of venom phosphodiesterase (after *Penicillium* nuclease digestion).

IRES

A segment of the 5' nontranslated region of encephalomyocarditis virus RNA directs internal entry of ribosomes during in vitro translation.
Jang S K, Kräusslich H G, Nicklin M J, Duke G M, Palmenberg A C, Wimmer E.
J. Virol., 62: 2636-2643, 1988.



Determinants in the 5' noncoding region of poliovirus Sabin 1 RNA that influence the attenuation phenotype.
Kawamura N, Kohara M, Abe S, Komatsu T, Tago K, Arita M, Nomoto A.
J. Virol., 63: 1302-1309, 1989.

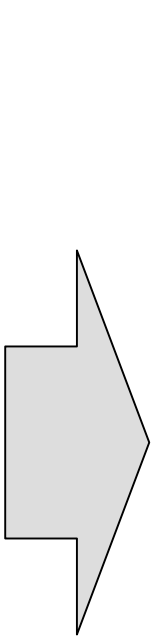


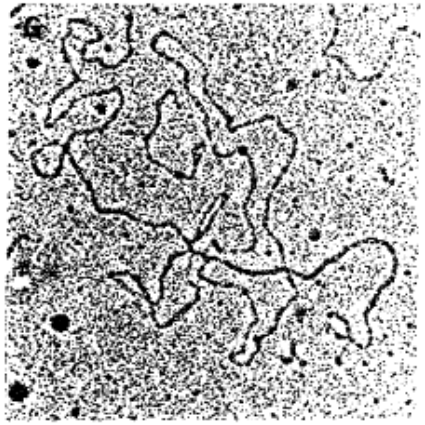
お手軽ノーベル賞（１）

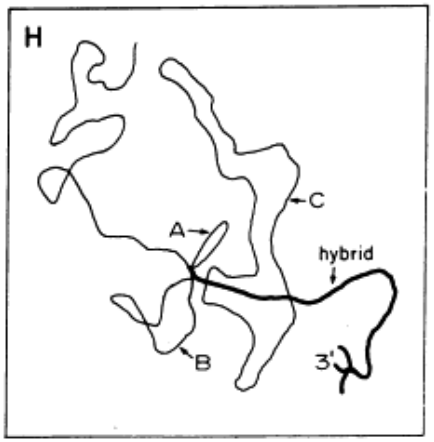
Splicing

Spliced segments at the 5' terminus of adenovirus 2 late mRNA.
Berget S M, Moore C, Sharp P A.
Proc. Natl. Acad. Sci. U S A., 74: 3171-3175, 1977.

Northern blottingで短かったので、・・・







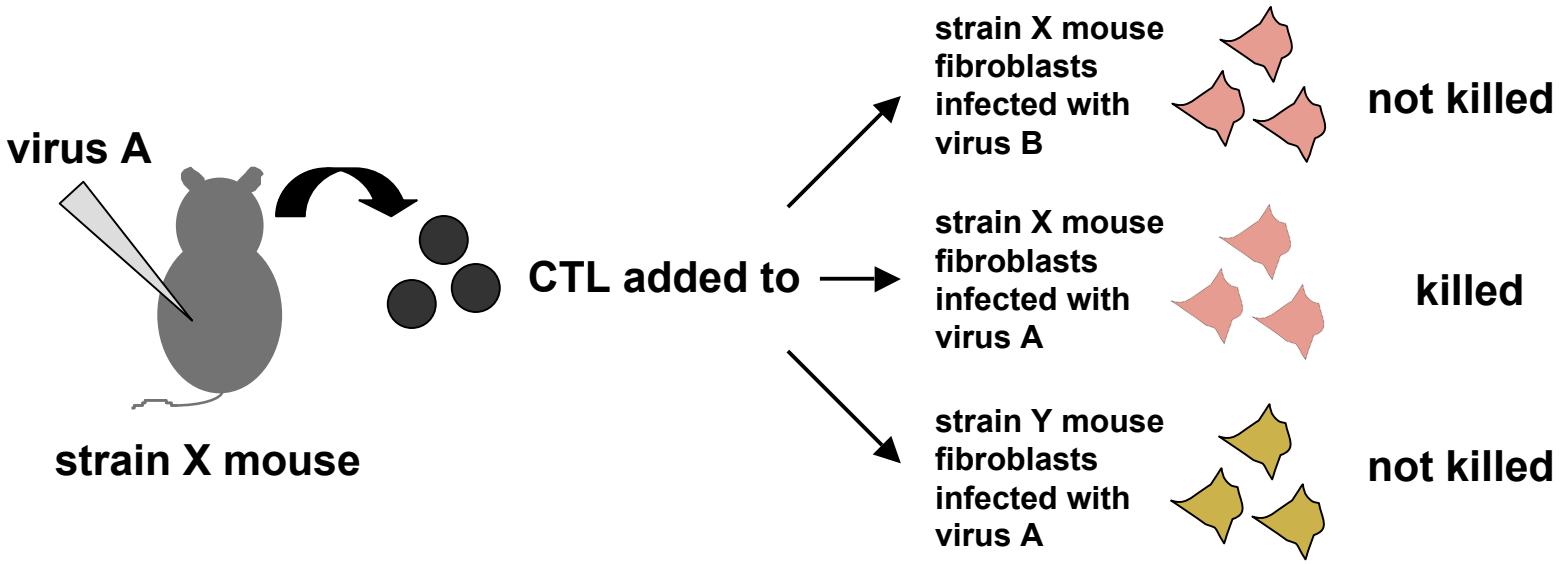
Immune System

Interferon

Nagano & Sawai, Kojima (1954)
Isaacs & Lindenmann (1957)

MHC restriction

Zinkernagel & Doherty (1974) using lymphoctyic choriomeningitis virus



Antigen presentation on MHC
Townsend, Wraitch (1987) using influenza virus
X-ray crystallography of class I MHC
Bjorkman (1987)

お手軽ノーベル賞（2）

MHC

Restriction of in vitro T cell-mediated cytotoxicity in lymphocytic choriomeningitis within a syngeneic or semiallogeneic system.

Zinkernagel R M, Doherty P C.

Nature, 248: 701-702, 1974.

TABLE 1 Cytotoxic activity of spleen cells from various strains of mice injected i.c. 7 d previously with 300 LD_{50}* of WE3 LCM virus for monolayers of LCM-infected or normal C3H (H-2^k) mouse L cells.

Experiment	Mouse strain	H-2 type	% ^{51}Cr release† Infected	Normal
1	CBA/H	*k*	65.1 ± 3.3	17.2 ± 0.7
	Balb/C	*d*	17.9 ± 0.9	17.2 ± 0.6
	C57Bl	*b*	22.7 ± 1.4	19.8 ± 0.9
	CBA/H × C57Bl	*k/b*	56.1 ± 0.5	16.7 ± 0.3
	C57Bl × Balb/C	*b/d*	24.8 ± 2.4	19.8 ± 0.9
	nu/+ or +/+		42.8 ± 2.0	21.9 ± 0.7
	nu/nu		23.3 ± 0.6	20.0 ± 1.4
2	CBA/H	*k*	85.5 ± 3.1	20.9 ± 1.2
	AKR	*k*	71.2 ± 1.6	18.6 ± 1.2
	DBA/2	*d*	24.5 ± 1.2	21.7 ± 1.7
3	CBA/H	*k*	77.9 ± 2.7	25.7 ± 1.3
	C3H/HeJ	*k*	77.8 ± 0.8	24.5 ± 1.5

* Other mice were injected with 2 × 10^6 LD_{50}, but levels of specific release were invariably lower due to the high dose immune paralysis[8,20] associated with viscerotropic (WE3) LCM virus.

† % ^{51}Cr release by normal spleen cells on infected targets ranged from: (experiment 1) 17.1 ± 0.3 to 20.0 ± 0.7; (experiment 2) 20.0 ± 1.4 to 25.3 ± 0.7; (experiment 3) 27.2 ± 2.0.

TABLE 2 % ^{51}Cr release* from infected C3H L cells overlaid with spleen cells from mice sampled at 7, 10 and 13 d after intravenous inoculation with 2,000 LD_{50} of WE3 LCM virus.

Mouse strain	Days after inoculation 7	10	13
CBA/H	72.0 ± 2.0	66.4 ± 1.4	27.5 ± 0.5
Balb/C	26.1 ± 0.7	28.0 ± 1.6	22.7 ± 1.8
C57Bl	27.3 ± 1.1	24.3 ± 1.8	24.0 ± 0.4

* Levels of ^{51}Cr release due to overlaying normal L cells with immune spleen cells, infected L cells with control spleen cells or with medium alone ranged from 17.1 ± 0.4 to 24.0 ± 1.4. Other mice were injected with 2 × 10^6 LD_{50}, but levels of specific release were invariably lower.

TABLE 3 % ^{51}Cr release from normal and infected peritoneal macrophages by spleen cells from control mice and from mice injected i.c. with 300 LD_{50} of WE3 LCM virus 7 d previously.

Spleen cells		Macrophage source	% ^{51}Cr release from macrophages Experiment 1 Infected	Normal	Experiment 2 Infected	Normal
Balb/C	Immune	Balb/C	61.8 ± 4.2^c	27.6 ± 1.9^e	77.5 ± 4.2^d	47.0 ± 3.5^d
	Anti-θ*		ND	ND	40.6 ± 2.5^e	ND
	N ascitic*		ND	ND	90.0 ± 2.7	ND
	Control		42.0 ± 4.8^a	40.5 ± 5.2^a	49.6 ± 2.5	43.5 ± 1.6
CBA/H	Immune		42.7 ± 6.7^a	33.7 ± 5.4^a	32.9 ± 3.0^a	48.6 ± 3.9^a
	Control		28.0 ± 4.1	40.5 ± 5.2	46.5 ± 3.7	39.7 ± 4.3
CBA/H	Immune	CBA/H	69.1 ± 2.8^e	30.9 ± 3.4^e	72.5 ± 5.2^d	40.0 ± 2.9^d
	Anti-θ		ND	ND	44.0 ± 2.5^d	ND
	N ascitic		ND	ND	74.3 ± 8.4	ND
	Control		34.2 ± 1.1	35.1 ± 3.7	46.5 ± 3.6	44.4 ± 6.2
Balb/C	Immune		46.2 ± 3.3^a	30.4 ± 3.8^b	44.0 ± 2.9^a	41.0 ± 2.4^a
	Control		34.9 ± 5.7	33.7 ± 5.6	40.5 ± 2.5	41.0 ± 2.4

* Treated with AKR anti-θ (C3H) ascitic fluid and guinea pig complement, or normal AKR ascitic fluid and guinea pig complement.

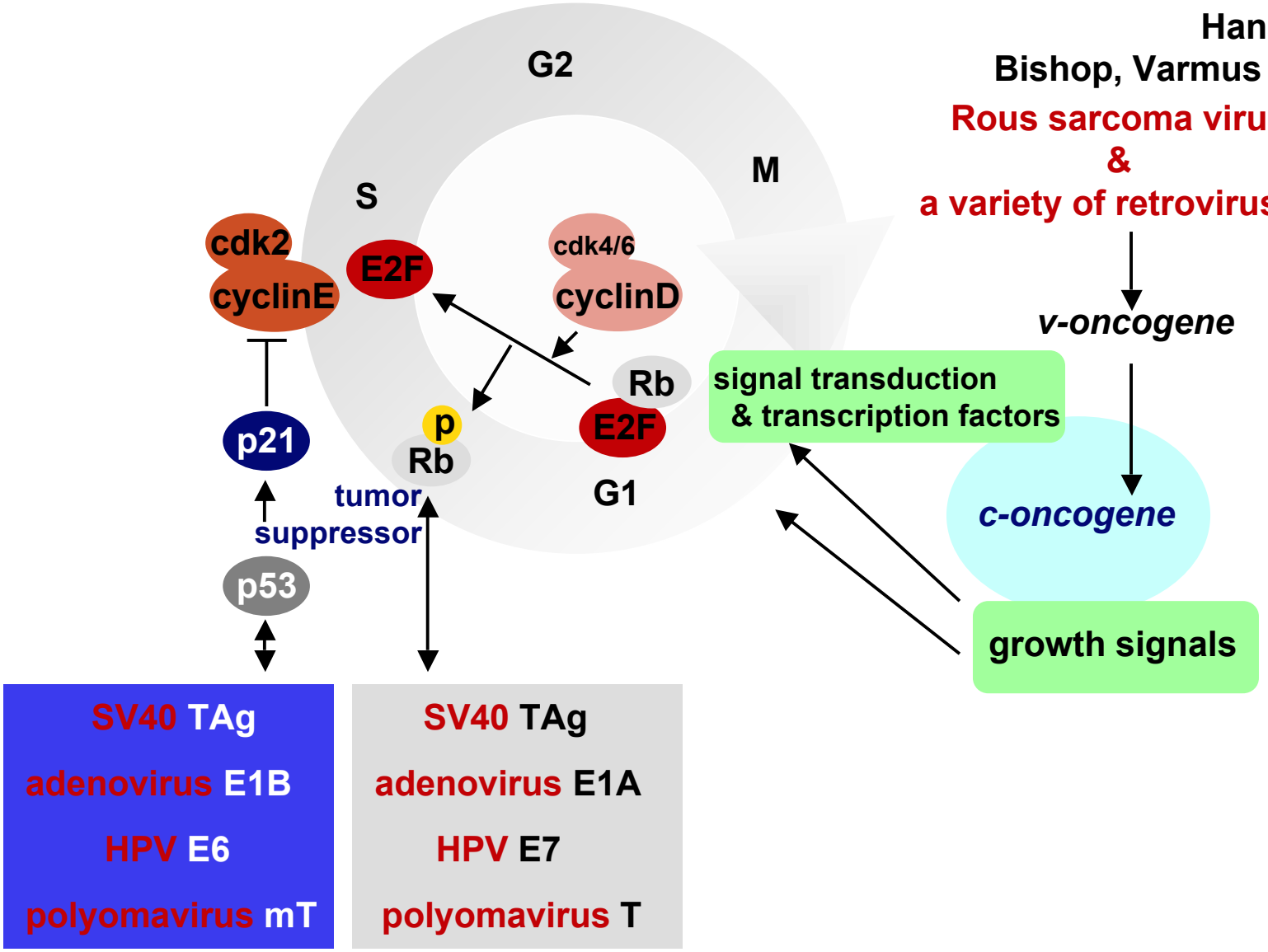
a, *b*, *c*, *d*, *e*, Differences by Student's *t* test between values for immune spleen cells treated with anti-θ ascitic fluid or normal ascitic fluid, immune and control spleen cells overlaid on infected macrophages (infected column), or immune spleen cells overlaid on infected and normal macrophages (normal column). *a*, $P > 0.05$; *b*, $P < 0.05$; *c*, $P < 0.02$; *d*, $P < 0.01$; *e*, $P < 0.001$.

ND, Not done.

for Cancer Research/Cell Cycle Control

Rous (1911)
Hanafusa (1971)
Bishop, Varmus (1976, 1984)

Rous sarcoma virus
&
a variety of retroviruses



お手軽ノーベル賞（3）

Oncogene in normal cells

DNA related to the transforming gene(s) of avian sarcoma viruses is present in normal avian DNA.

Stehelin D, Yarmus H E, Bishop J M, Vogt P K.

Nature, 260: 170-173, 1976.

Table 1 Homology between $cDNA_{sarc}$ and normal DNAs

Assay	Hybridisation conditions		Extent of reaction between $cDNA_{sarc}$ and DNA from						
	$[Na^+]$	Temperature	Chicken	Quail	Turkey	Duck	Emu	Mouse	Calf
S1	0.9 M	68°	52%	46%	48%	45%	24%	<2%	<2%
HAP	0.9 M	68°					36%		<5%
HAP	1.5 M	59°					54%		

DNA was extracted from 10–11-d-old embryos of chickens, ducks and quails, 3-d-old mice (strain RIII), livers of adult turkeys, liver and heart of a 22-d-old emu, and calf thymus. Reaction mixtures contining denatured DNA (8 mg ml^{-1}) and ^3H-$cDNA_{sarc}$ (0.32 ng ml^{-1}, 7,000 c.p.m. ml^{-1}) in a final volume of 0.3 ml were incubated at either 59 or 68 °C for 48 h. Samples incubated at 59 °C were in 1.5 M NaCl (final C_0t = 40,000), those incubated at 68 °C were in 0.9 M NaCl (final C_0t = 32,000); all reactions also continued 0.001 M EDTA–0.02 M Tris-HCl, *p*H 7.4. Duplex formation was measured by either hydrolysis with S1 nuclease[28] (in 0.3 M NaCl at 50 °C) or fractionation on hydroxyapatite (HAP) (samples adsorbed in 0.14 M sodium phosphate at 50 °C).

Molecular Virology

To understand the molecular mechanism of multiplication and pathogenesis

⬇

To control and prevent virus diseases
To develop novel technology

Contribution from Japan

quoted in "Virology, 4th edition (edited by Fields)"

Virus Isolation, Cloning and Genome Analysis

± DNA

genome analysis

- VZV — *Kinoshita H*, 1988, JV
- HHV-6A,B — *Isegawa, Y*, 1999, JV; *Inoue N*, 1994, JV
- EB — *Ogura M*, 1987, JV
- CMV — *Tamashiro JC*, 1986, JV
- Cowpox virus — *Funahashi S*, 1988, JGV
- Polyomavirus — *Soeda E*, 1980, Nature
- Vaccinia virus — *Shida H*, 1986, Virology

+ RNA (Retrovirus)

genome analysis

- Moloney murine leukemia virus — *Tsukiyama T*, 1990, Virology

isolation

- HTLV-1 — *Uchiyama T*, 1977, Blood; *Yoshida M*, 1982, PNAS

Isolation ± RNA

- Rotavirus — *Sato K*, 1981, Arch Virol; *Hasegawa A*, 1982, J Clin Microbiol

genome analysis

- Reovirus — *Munemitsu SM*, 1986, BBRC; *Imai M*, 1983, PNAS; *Miura K*, 1974, PNAS
- Rotavirus — *Matsui SM*, 1990, JV; *Nagagomi O*, 1990, JV
- Bluetongue virus — *Sugiyama K*, 1981, Virology

+ RNA

genome analysis

- Japan encephalitis virus — *Takegami T*, 1986, Virology; *Sumiyoshi H*, 1987, Virology; *Hashimoto H*, 1988, Virus Gene
- HCV — *Sakaoka H*, 1994, JGV; *Fukushi S*, 1994, BBRC; *Ito T*, 1997, JV; *Ogata N*, 1991, PNAS; *Kato N*, 1990, PNAS; *Okamoto H*, 1991, JGV; *Tanaka T*, 1995, BBRC
- Poliovirus — *Kitamura N*, 1981, Nature
- Peritonitis virus — *Motokawa K*, 1995, Arch Virol

− RNA

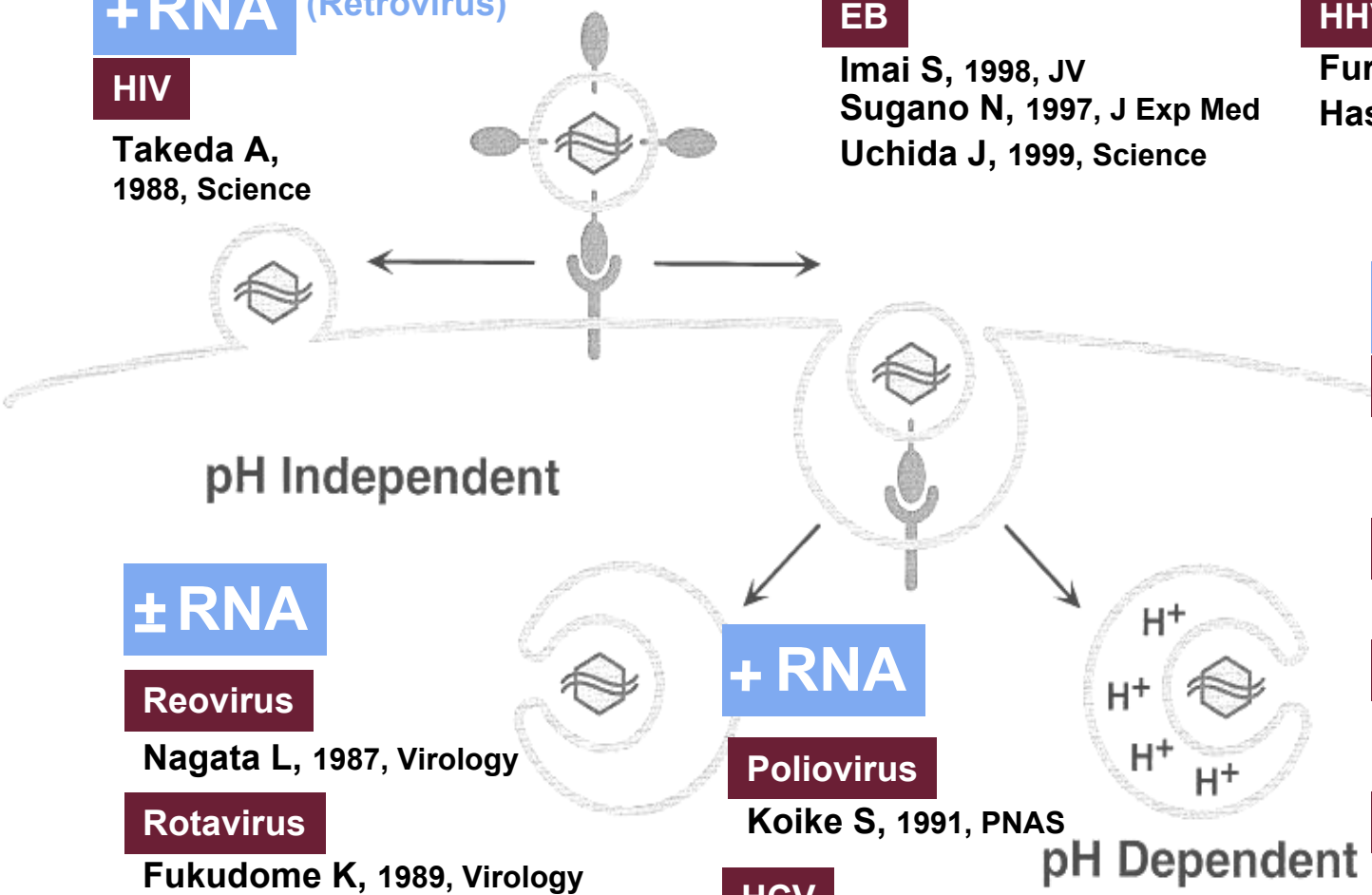
Isolation

- Mumps virus — *Saito H*, 1996, Microbiol Immunol
- Sendai virus — *Kuroda M, Ishida N*, 1953, Yokohama Med Bull
- Influenza virus — *Nakajima K*, ature
- Akabane virus — *Kurogi H*, 1987, Vet Microbiol
- HDV — *Sakaguchi T*, 1994, Arch Virol

genome analysis

- Sendai virus — Shioda T, 1983, 1986, NAR
- Influenza virus — *Enami M*, 1990, PNAS
- Parainfluenza virus — *Bando H*, 1990, Virology; *Suzu S*, 1987, NAR; *Sakai Y*, 1987, NAR; *Tsurudome M*, 1989, Virology; *Yuasa T*, 1990, Virology; *Kawano M*, 1990, Virology
- Measles virus — *Takeda M*, 1999, Virology
- Mumps virus — *Tanabayashi K*, 1990, Virology; *Takeuchi K*, 1988, JGV; *Okazaki K*, 1992, Virology
- Punta Toro phlebovirus — *Ihara T*, 1984, Virology
- HDV — *Makino S*, 1987, Nature

Receptor and Virus-Cell Interaction



+ RNA (Retrovirus)

HIV

Takeda A, 1988, Science

± DNA

EB

Imai S, 1998, JV
Sugano N, 1997, J Exp Med
Uchida J, 1999, Science

HHV-6

Furukawa M, 1994, J Immunol
Hasegawa H, 1994, JV

± RNA

Reovirus

Nagata L, 1987, Virology

Rotavirus

Fukudome K, 1989, Virology
Suzuki H, 1985, Arch Virol

Hantaan virus

Arikawa J, 1989, JGV

+ RNA

Poliovirus

Koike S, 1991, PNAS

HCV

Kimura T, 1994, Arch Virol

Mouse hepatitis virus

Yokomori K, 1992, JV

− RNA

Measles virus

Tatsuo H, 2000, Nature
Iwata K, 1995, JBC

Sendai virus

Ito Y, 1983, Arch Virol

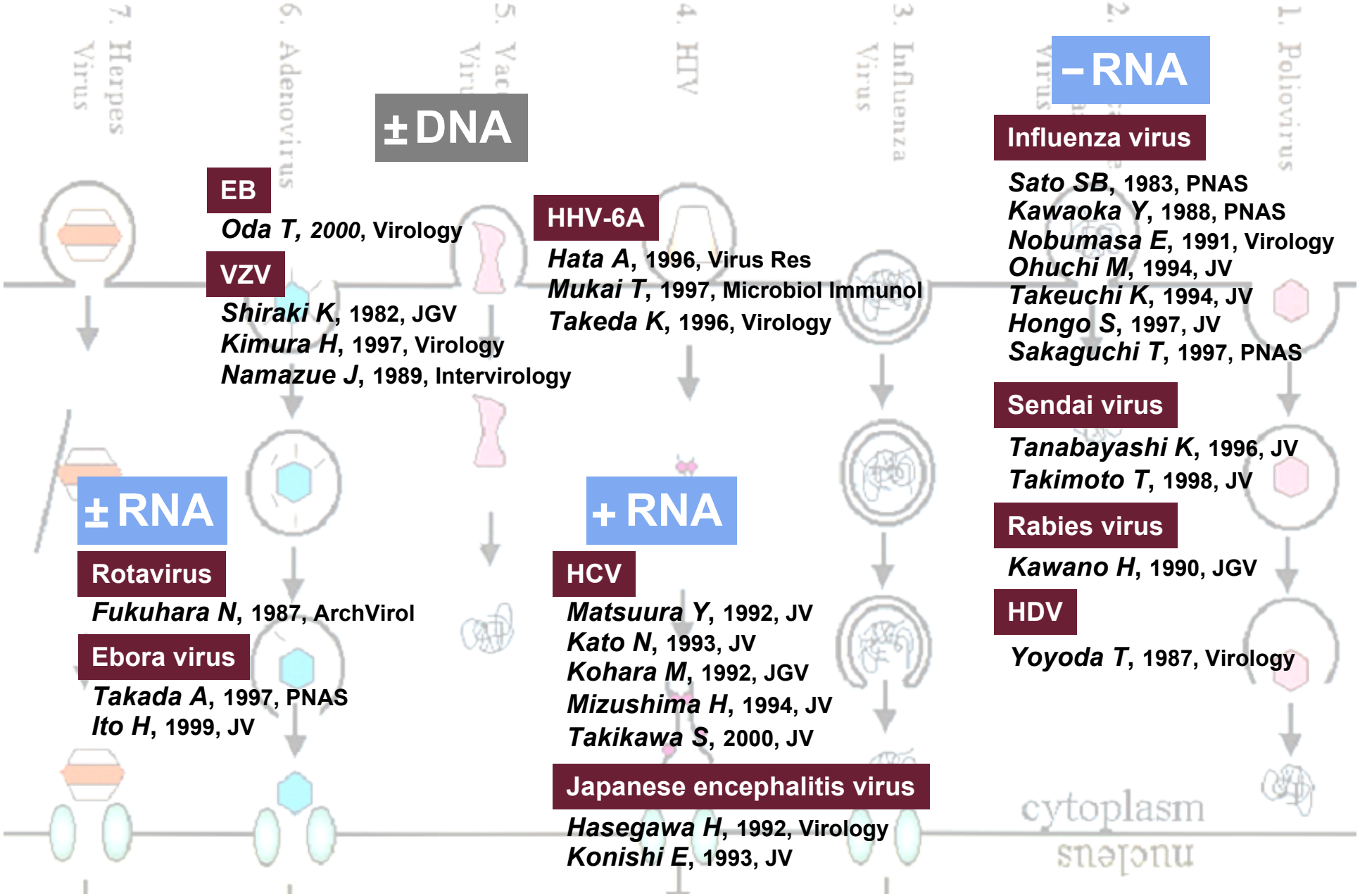
HBV

Komai K, 1988, Virology
Kuroki K, 1994, JV

Coronavirus

Kubo H, 1994, JV
Suzuki H, 1996, JV
Taguchi F, 1993, JV
Yokomori K, 1989, Virology

Entry and Uncoating



± DNA

EB

Oda T, 2000, Virology

VZV

Shiraki K, 1982, JGV
Kimura H, 1997, Virology
Namazue J, 1989, Intervirology

HHV-6A

Hata A, 1996, Virus Res
Mukai T, 1997, Microbiol Immunol
Takeda K, 1996, Virology

± RNA

Rotavirus

Fukuhara N, 1987, ArchVirol

Ebora virus

Takada A, 1997, PNAS
Ito H, 1999, JV

+ RNA

HCV

Matsuura Y, 1992, JV
Kato N, 1993, JV
Kohara M, 1992, JGV
Mizushima H, 1994, JV
Takikawa S, 2000, JV

Japanese encephalitis virus

Hasegawa H, 1992, Virology
Konishi E, 1993, JV

– RNA

Influenza virus

Sato SB, 1983, PNAS
Kawaoka Y, 1988, PNAS
Nobumasa E, 1991, Virology
Ohuchi M, 1994, JV
Takeuchi K, 1994, JV
Hongo S, 1997, JV
Sakaguchi T, 1997, PNAS

Sendai virus

Tanabayashi K, 1996, JV
Takimoto T, 1998, JV

Rabies virus

Kawano H, 1990, JGV

HDV

Yoyoda T, 1987, Virology

Replication

±DNA

SV40

Murakami Y, 1986, PNAS
Waga S, 1994, JBC
Tsurimoto T, 1990, Nature
Ishimi Y, 1991, JBC

HBV

Yokosuka O, 1985, PNAS
Tsurimoto T, 1987, PNAS
Araki K, 1989, PNAS
Tokino T, 1991, JV
Nagaya T, 1987, Genes Dev
Tagawa M, 1987, JV

Adenovirus

Nagata K, 1983, PNAS
Miyamoto NG, 1985, EMBO
Horikoshi M, 1991, PNAS
Tamanoi F, 1982, PNAS
Seto E, 1991, Nature

Vaccinia virus

Koji I, 2001, JV

Polyomavirus

Ito Y, 1977, PNAS

HSV

Umene K, 1996, JGV

+RNA (Retrovirus)

EB

Takaki K, 1984, PNAS
Takada K, 1986, JV
Kawanishi M, 1993, JV
Tsurumi T, 1993, JV
Takada K, 1986, JV
Tsujimoto Y, 1989, PNAS
Matsuo T, 1984, Science
Ohno S, 1977, PNAS
Ooka T, 1984, Virology

Retrovirus

Hagino-Yamagishi K, 1987, JV
Fujisawa T, 1988, Cell
Masuda T, 1998, JV
Sakai H, 1993, JV

CMV

Takekoshi M, 1991, Gene
Watanabe S, 1993, Virology
Hayashi ML, 2000, PNAS

Murine leukaemia virus

Inada T, 1991, JGV

Moloney murine leukemia virus

Tanese N, 1988, PNAS

±RNA

Rotavirus

Kitaoka S, 1986, Antiviral Res

Reovirus

Sakuma S, 1971, JV

−RNA

HDV

Hamaguchi M, 1983, Virology

Coronavirus

Makino S, 1987, PNAS

Sendai virus

Horikami SM, 1996, Virology
Kato A, 1996, Genes Cells

Borna disease virus

Mizutani T, 1998, Arch Virol

Transcription

±DNA

VZV

Kato T, *1989, Microbiol Immunol*

Moriuchi H, *1995, JV*

EB

Shimizu N, 1989, Virology

Harada S, 1997, JV

HSV

HSVNishioka Y, 1977, PNAS

HBV

Kaneko S, 1988, JV

Murakami S, 1994, Virology

Seto E, 1990, Nature

Fukai K, 1997, Virology

Nakatake H, 1993, Virology

Yaginuma K, 1989, JV

Adenovirus

Yoshinaga S, 1986, EMBO

±RNA

Reovirus

Furuichi Y, 1975, PNAS

Watanabe Y, 1968, JMB

Yamakawa, 1981, JBC

Nonoyama M, 1974, NAR

Rotavirus

Imai M, 1983, JV

+RNA

Poliovirus

Nomono A, 1976, PNAS

HCV

Kanai A, 1995, FEBS Lett

Ago H, 1999, Structure Fold Des

Yanagi M, 1997, PNAS

−RNA

Influenza virus

Honda A, 1987, J Biochem

Sendai virus

Mizumoto K, 1987, Prog Nucl Acid Res Mol Biol

HDV

Hamaguchi M, *1985, Virology*

Measles virus

Horikami SM, *1994, JV*

Takeda M, 1998, JV

Punta Toro phlebovirus

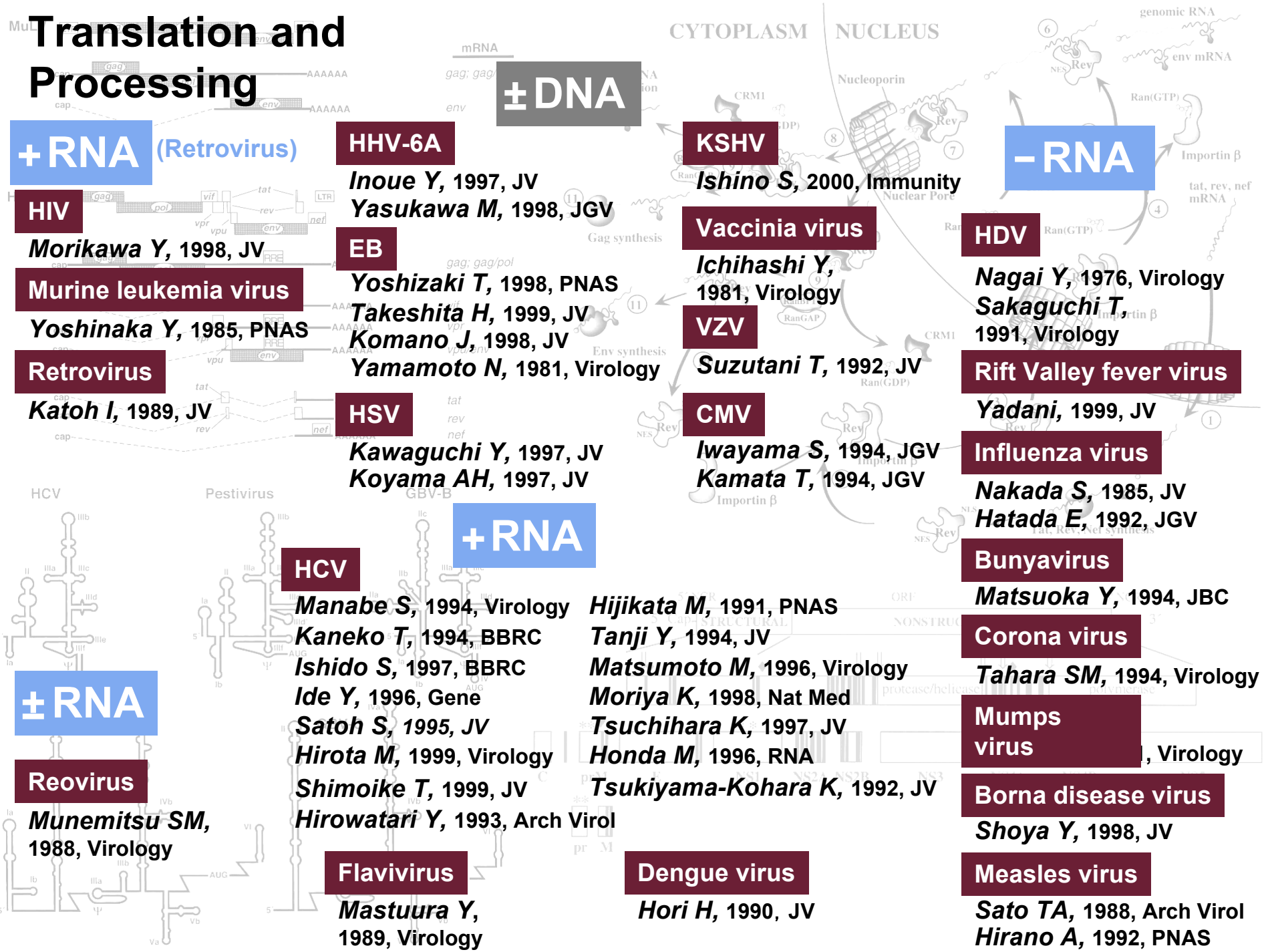
Ihara T, 1985, Virology

Coronavirus

Maeda A, 1998, Virus Res

Makino S, 1993, JV

Translation and Processing



+RNA (Retrovirus)

HIV
Morikawa Y, 1998, JV

Murine leukemia virus
Yoshinaka Y, 1985, PNAS

Retrovirus
Katoh I, 1989, JV

±DNA

HHV-6A
Inoue Y, 1997, JV
Yasukawa M, 1998, JGV

EB
Yoshizaki T, 1998, PNAS
Takeshita H, 1999, JV
Komano J, 1998, JV
Yamamoto N, 1981, Virology

HSV
Kawaguchi Y, 1997, JV
Koyama AH, 1997, JV

KSHV
Ishino S, 2000, Immunity

Vaccinia virus
Ichihashi Y, 1981, Virology

VZV
Suzutani T, 1992, JV

CMV
Iwayama S, 1994, JGV
Kamata T, 1994, JGV

−RNA

HDV
Nagai Y, 1976, Virology
Sakaguchi T, 1991, Virology

Rift Valley fever virus
Yadani, 1999, JV

Influenza virus
Nakada S, 1985, JV
Hatada E, 1992, JGV

Bunyavirus
Matsuoka Y, 1994, JBC

Corona virus
Tahara SM, 1994, Virology

Mumps virus
, Virology

Borna disease virus
Shoya Y, 1998, JV

Measles virus
Sato TA, 1988, Arch Virol
Hirano A, 1992, PNAS

+RNA

HCV
Manabe S, 1994, Virology
Kaneko T, 1994, BBRC
Ishido S, 1997, BBRC
Ide Y, 1996, Gene
Satoh S, 1995, JV
Hirota M, 1999, Virology
Shimoike T, 1999, JV
Hirowatari Y, 1993, Arch Virol
Hijikata M, 1991, PNAS
Tanji Y, 1994, JV
Matsumoto M, 1996, Virology
Moriya K, 1998, Nat Med
Tsuchihara K, 1997, JV
Honda M, 1996, RNA
Tsukiyama-Kohara K, 1992, JV

Flavivirus
Mastuura Y, 1989, Virology

Dengue virus
Hori H, 1990, JV

±RNA

Reovirus
Munemitsu SM, 1988, Virology

Assembly

+ RNA (Retrovirus)

HIV

Ono A, 1999, JV
Kasahara N,
1994, Science

± RNA

Reovirus

Nonoyama M, 1970, JV

Bluetongue virus

Tanaka S, 1994, JV

+ RNA

Flavivirus

Ohyama, 1977,
Microbiol Immunol
Hase T, 1987, Arch virol

HCV

Koike K, 1995, JGV
Takahashi K,
1992, Virology

Sindbis virus

Ohno K,
1997, Nat Biotechol

± DNA

HBV

Yaginuma K, 1987, PNAS
Machida A, 1991, JV
Ueda K, 1991, JV

HSV

Igarashi K, 1993, JV

EB

Hinuma Y, 1967 , JV

– RNA

Sendai virus

Horikami SM, 1992, JV

Corona virus

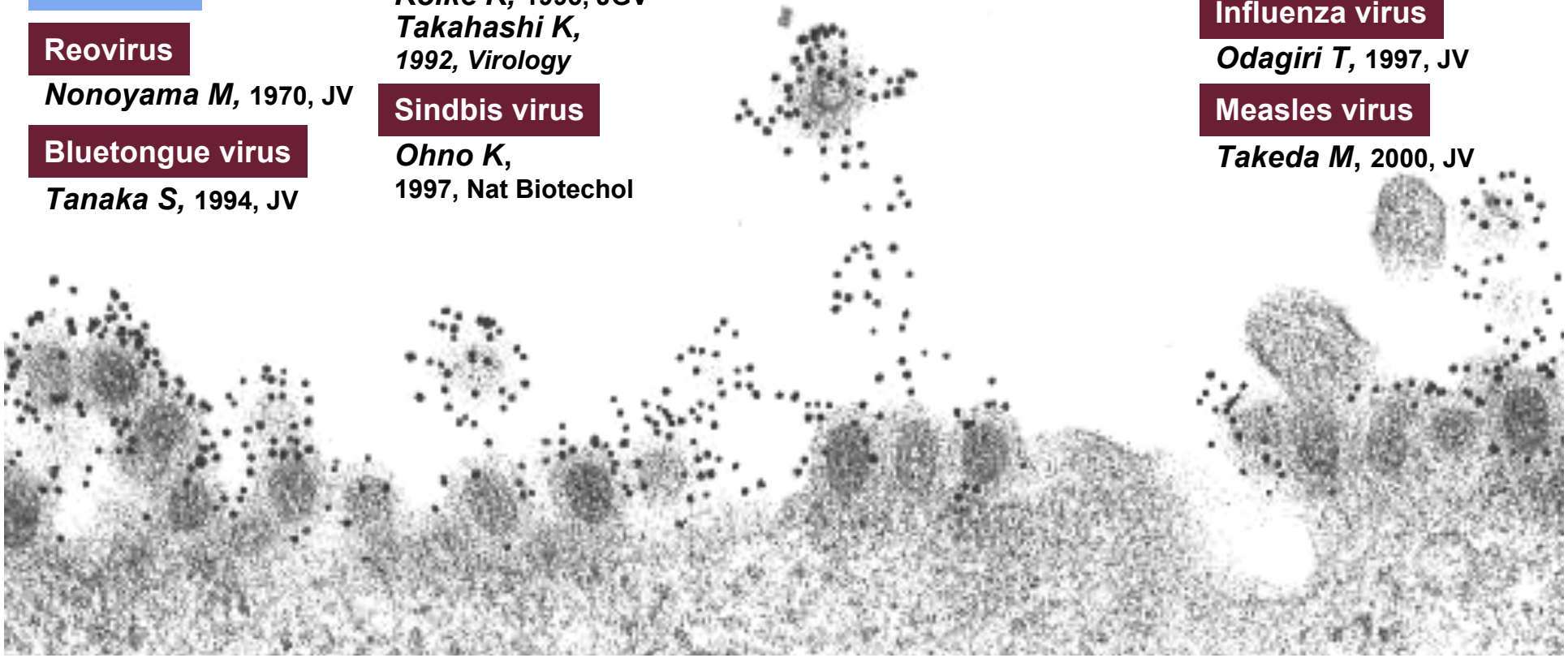
Furuya T, 1993, Virology

Influenza virus

Odagiri T, 1997, JV

Measles virus

Takeda M, 2000, JV



Pathogenicity

±DNA

HBV
Miyaki M, 1986, JGV
Okamoto H, 1987, JV
Ishikawa T, 1995, PNAS
Moriyama T, 1990, Science
Nakamoto Y, 1997, J Immunol
Ohashi K, 2000, Nat Med
Nagaya T,
1987, Genes and Dev

CMV
Kondo K, 1994, PNAS
Yamashita Y,
1993, Virology

EB
Izumi K, 1994, JV
Yoshiyama H, 1997, JV

VZV
Nagashima K,
1975, Acta Neuropathol

HSV
Yoshino K,
1982, Microbiol Immunol

HHV
Suga S, 1992, J Med Virol
Saito Y, 1995, J Neurovirol
Katano H, 2000, JV
Okuno T,
1989, J Clin Microbiol

Prion
Miura T, 1996, FEBS Lett
Kaneko K, 1997, PNAS
Sakaguchi, 1996, Nature
Muramoto T, 1997, Nat Med

+RNA (Retrovirus)

Murine AIDS virus
Kubo Y, 1994, JV

HTLV
Baba E, 1995, J immunol

+RNA

HCV
Saito I, 1990, PNAS
Sakamuro D, 1995, JV
Shimizu Y, 1994, JV
Shimotohno K,
1993, Semin Virol

Dengue virus
Kurane I, 1991, JV

±RNA

Rotavirus
Konno T, 1978, J Med Virol
Hoshino Y, 1985, PNAS
Hashiro G, 1977, Arch Virol
Hoshino Y, 1985, PNAS
Matsui SM, 1989, Adv Vir Res
Urasawa T, 1981, Jpn J Med Sci Biol

−RNA

Coronavirus
Yokomori K, 1991, JV

Hantavirus
Mori F, 1991,
Zentralbl Veterinarmed

Influenza
Tashiro M, 1987, Virology
Horimoto T, 1994, JV
Ito T, 1998, JV
Goto H, 1998, PNAS
Takeda A, 1999, JV

Japanese encephalitis virus
Konishi E, 2000, Virology
Ogata A, 1991, JV

Renal syndrome virus
Yamanishi K, 1988, Vaccine

Sendai virus
Tashiro M, 1988, Virology
Kato A, 1997, JV
Ito M, 1988, JGV
Kurotani A,
1998, Genes Cells

Mumps virus
Yamada A, 1990, Vaccine

Parainfluenza virus
Komada H,
1991, Arch Virol

Sindbis virus
Kimura T,
1993, JV

HDV
Morimoto K,
1998, PNAS

Biology suports Biology and Virology