Poxviruses

- Virion
- Genome
- Genes and proteins
- Viruses and hosts
- Diseases
- Distinctive characteristics

Viruses and hosts

- Poxviridae from English *pocks* (pox), referring to blistering skin lesions
- Two subfamilies:
  - *Chordopoxvirinae:* infects vertebrates
    - Humans: variola, vaccinia (vaccine strain), molluscum contagiosum
    - Viruses that infect a variety of birds and mammals: monkeys, cattle, etc
  - *Entomopoxvirinae*: infect insects (beetles, butterflies, flies, etc.)

Table 26.1 Some poxviruses and their hosts

Virus	Reservoir host	Other hosts
Variola (smallpox)	Humans	None
Molluscum contagiosum	Humans	None
Vaccinia	Unknown	Humans, cows, buffaloes
Monkeypox	Squirrels, rodents	Humans, monkeys
Cowpox	Rodents	Humans, cows, cats, zoo animals
Orf	Sheep	Humans, various ruminants
Fowlpox	Birds	Humans (as vaccine vector)
Myxoma	Rabbits	None
Entomopox	Insects	None

Diseases

- Molluscum contagiosum: the only existing natural poxvirus infection of humans; relatively rare but more common in immunocompromised patients
- Monkeypox, cowpox, tanapox are animal poxviruses that occasionally infect humans
- Smallpox was a debilitating and fatal worldwide disease, but now eradicated
  - Caused lesions (pocks)
  - Records suggest that virus was endemic in Egypt and India by 1st century
  - Spread throughout Asia, Europe and North Africa by the tenth century
  - Colonization brought the virus to the Americas, southern Africa and Australia by the 16-18th centuries
  - Smallpox killed over 500 million people in the 20th century!

    ; Compared to: 320 million deaths caused by wars, the Spanish flu and AIDS combined

Diseases

- **Variolation** led to vaccination, which has eradicated smallpox worldwide
  - Infection with variola virus by an unnatural route results in milder disease
  - Edward Jenner (1796) developed a procedure of **vaccination** for smallpox using the material from cowpox lesions

- Poxviruses remain a subject of intense research interest
  - Vaccinia virus is used for smallpox vaccination and in research laboratories

Virion

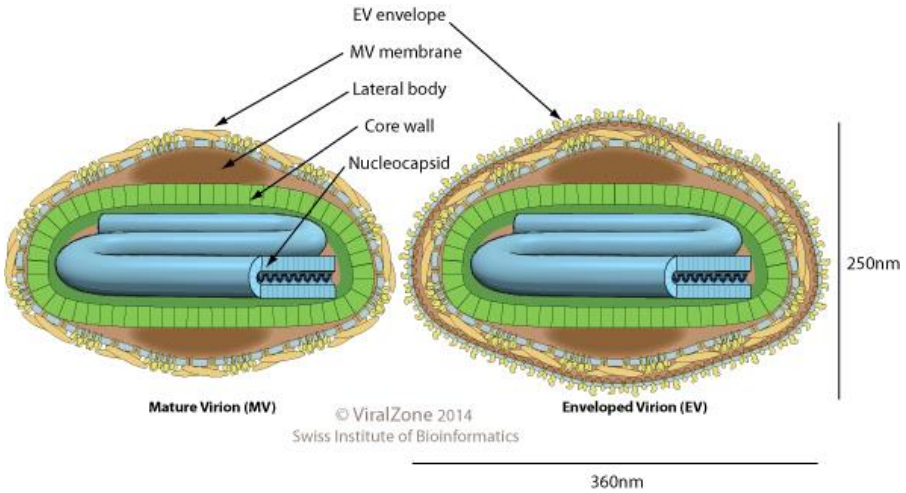
- Complex, ovoid or brick shaped particles, 310 x 240 x 140 nm
- Surface ridges or “tubules”
- No typical symmetry elements
- Internal core and lateral bodies
- Virions exist in two infectious forms:
  - Mature virus (MV): one lipid membrane
  - Extracellular virus (EV): two lipid membranes

Genome, Genes and Proteins

- Linear double-stranded DNA, 150-250 kb (vaccinia virus: 200 kb)
- Covalently closed hairpin ends: no free 3′ and 5′ ends
- 10 kb inverted terminal repeats
- 150-250 genes (vaccinia virus: 200)
- Each gene has its own transcriptional promoter
- No spliced mRNAs (no introns)
- Genes are distributed on both DNA strands
- Genes are expressed in three temporal classes with distinct functions:
  - Early: dissolve core, direct DNA replication and intermediate gene expression, combat host defenses
  - Intermediate: direct late gene expression, combat host defenses
  - Late: virion structural proteins, virion transcription enzymes

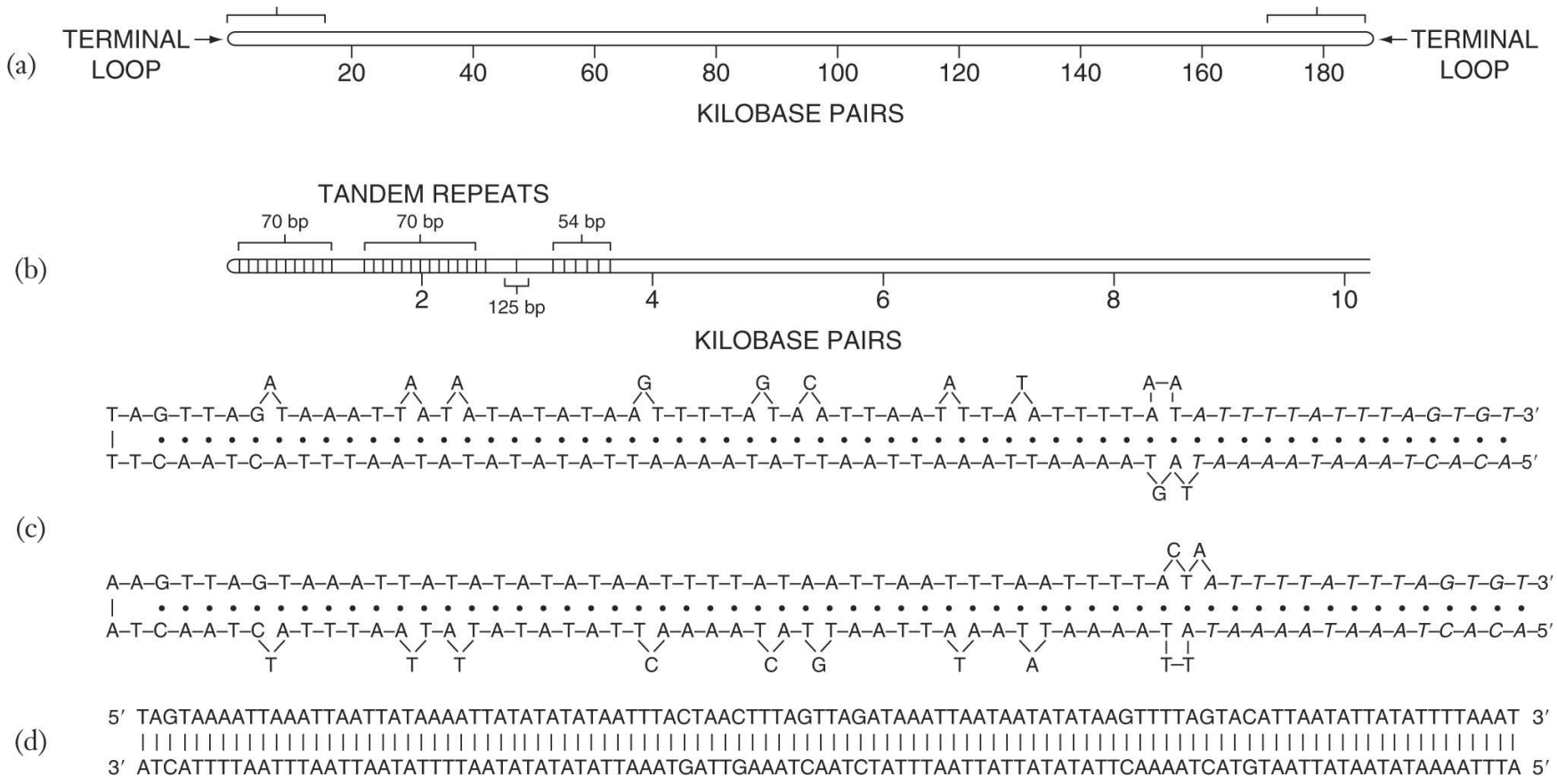
Virion

- Largest of all animal viruses; brick-shaped or ovoid particles
- Two forms of vaccinia virions have different roles in spreading infection
  - Intracellular mature virus (IMV or MV):
    - has an outer envelope derived from ER, released only upon cell lysis
    - Attaches to cellular **gycosaminoglycans** & enters by fusion or endocytosis
    - Is stable and responsible for person to person transmission
  - Extracellular enveloped virus (IEV or EV):
    - is a mature virus wrapped in an additional lipid bilayer (Golgi)
    - Phagocytosis, or the outer membrane ruptures releasing a mature virus
    - Is fragile and responsible for cell to cell spread



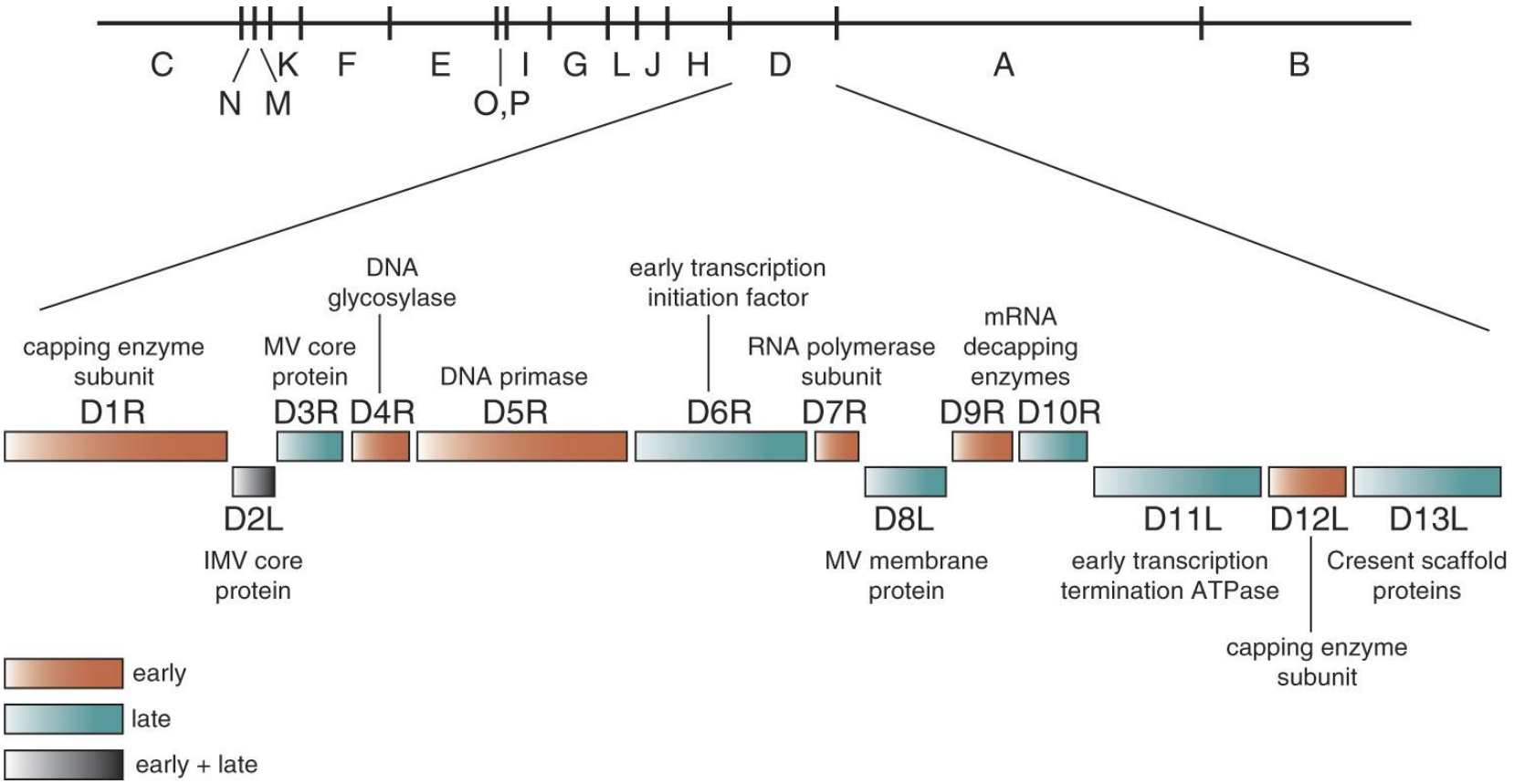
Genome

- Linear vaccinia virus genomes have covalently sealed hairpin ends and lack introns



Genome

- Functional organization of the vaccinia virus genome

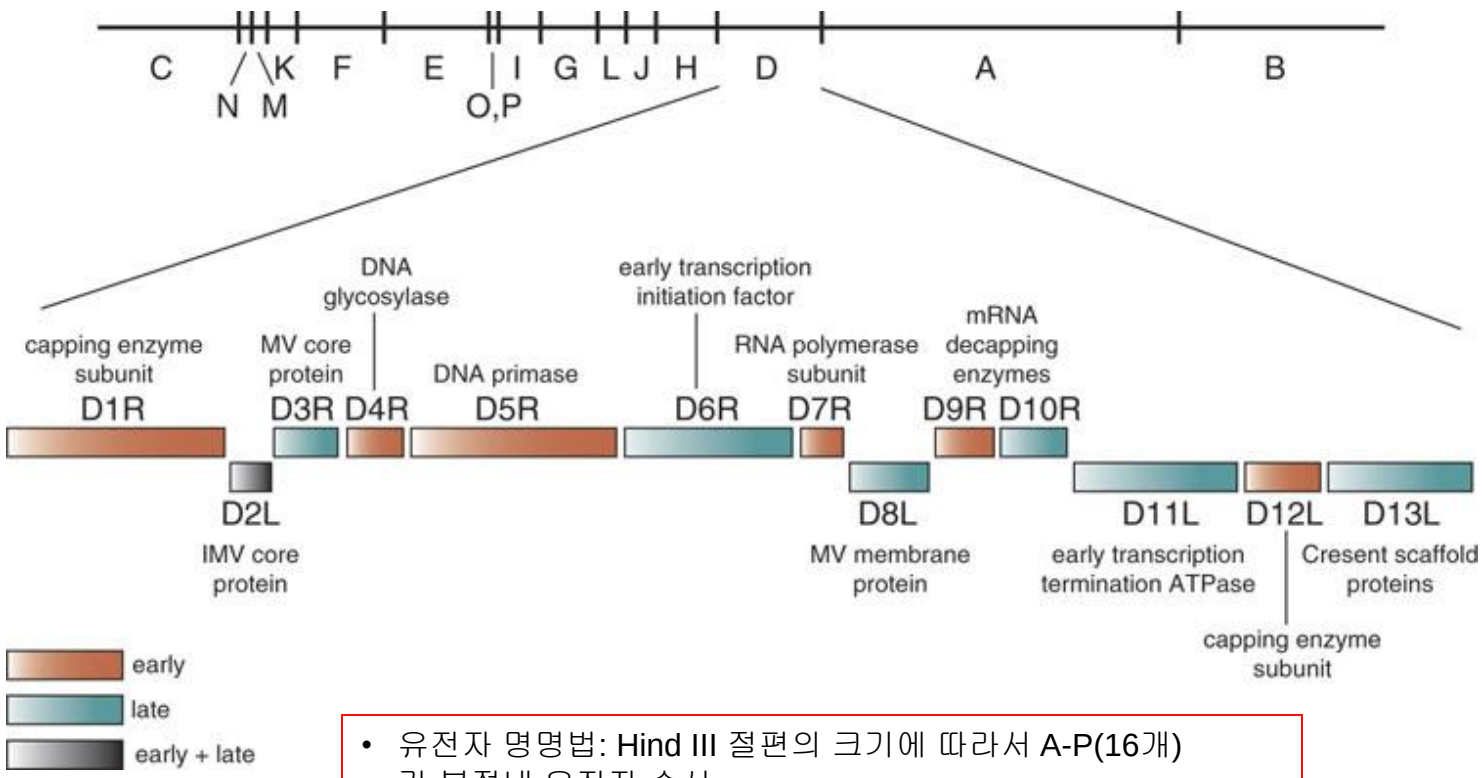


Genes and proteins

Table 26.2 Enzymes packaged in vaccinia virions

Function	Protein	No. of subunits	Mol. wt. (kD)
RNA synthesis	RNA polymerase	8	410
Transcription initiation	Early gene specificity factor	1	94
	Early gene initiation factor	2	166
mRNA 5' end capping	Cap guanylyltransferase	2	130
	Ribose-methyltransferase	1	39
Transcription termination	Cap guanylyltransferase	2	130
	DNA-dependent ATPase	1	72
mRNA 3' end polyadenylation	Poly(A) polymerase	2	94
Nucleic acid topology	RNA helicase	1	77
	DNA helicase	1	56
	DNA topoisomerase I	1	37
Protein modification	Protein kinase I	1	34
	Protein kinase II	1	52
	Protein phosphatase	1	20

Genes and proteins



- 유전자 명명법: Hind III 절편의 크기에 따라서 A-P(16개)
- 각 분절내 유전자 순서
- 전사방향

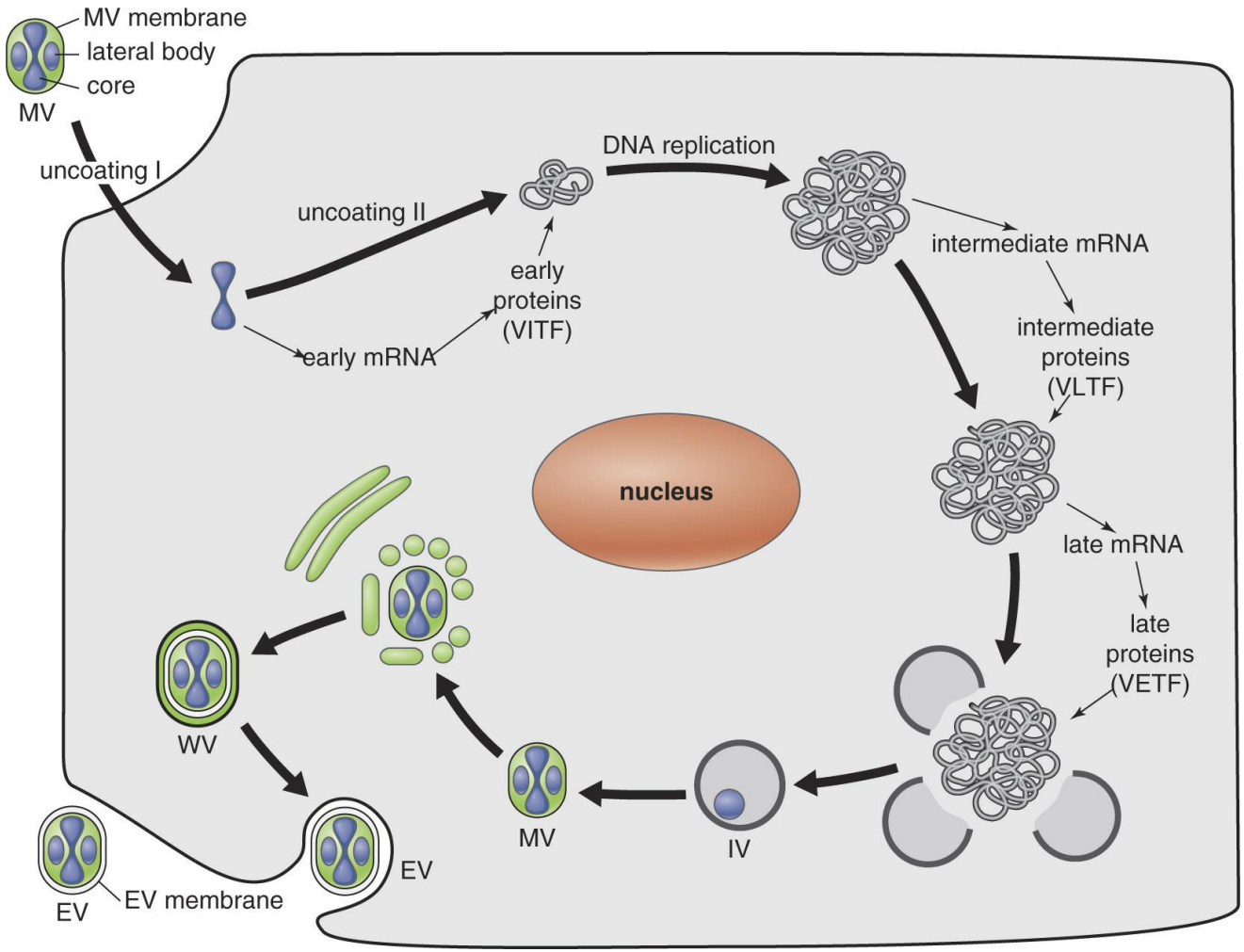
Genes and proteins

- Poxvirus genes are expressed in a regulated transcriptional cascade controlled by viral transcription factors
  - Virus early transcription factors (VETF) are packaged into virion and act upon release into cytoplasm
  - Viral intermediate transcription factors (VITF) trigger intermediate gene expression
  - Viral late transcription factors (VLTF) activate late genes

- Virus-coded enzymes packaged in the core carry out early RNA synthesis and packaging
  - Enzymes transcribe, cap, methylate and polyadenylate mRNA
  - Capping requires a **guanylytransferse** and a **ribose-methyltransferase**

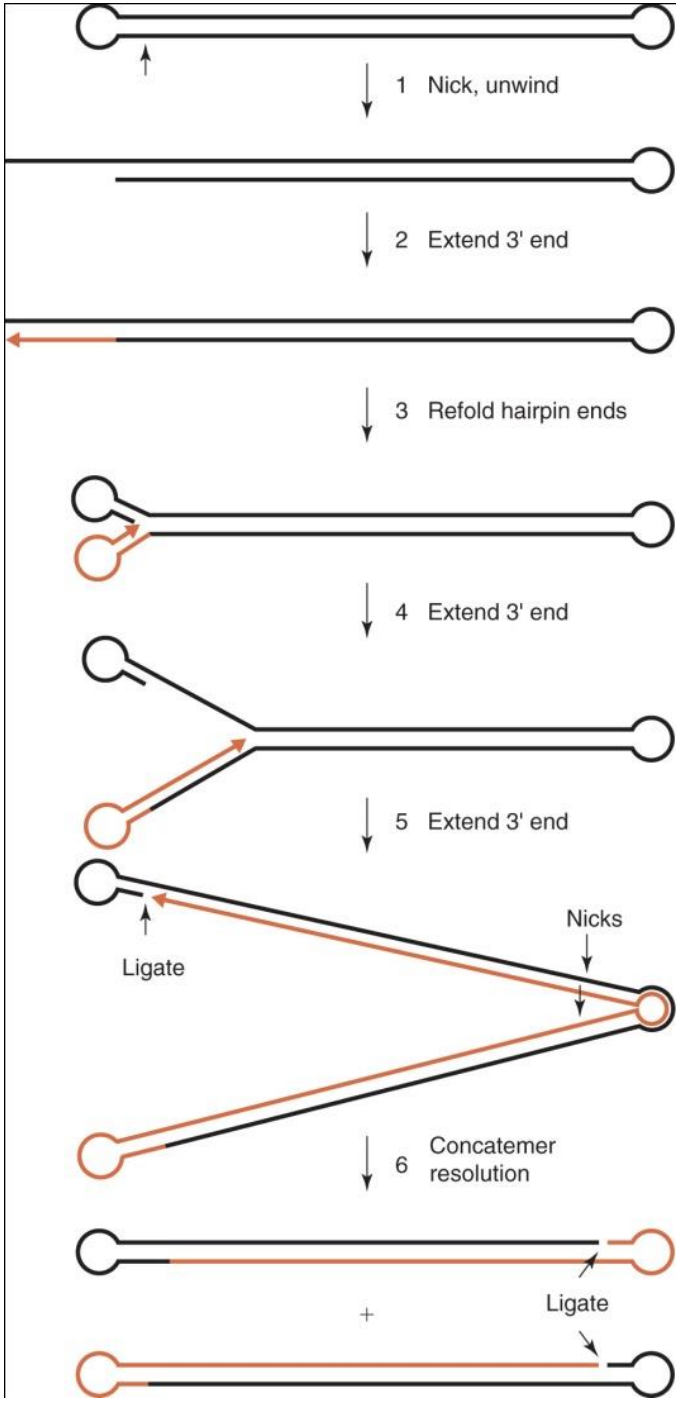
Life cycle

- Poxviruses replicate in the cytoplasm



DNA replication

- Poxviruses produce large concatemeric DNA molecules that are resolved into monomers
  - Rolling hairpin replication produces head-to-tail and tail-to-tail **concatemers**
  - Concatemers are resolved by virus-coded **resolvase**
- A model for vaccinia virus DNA replication

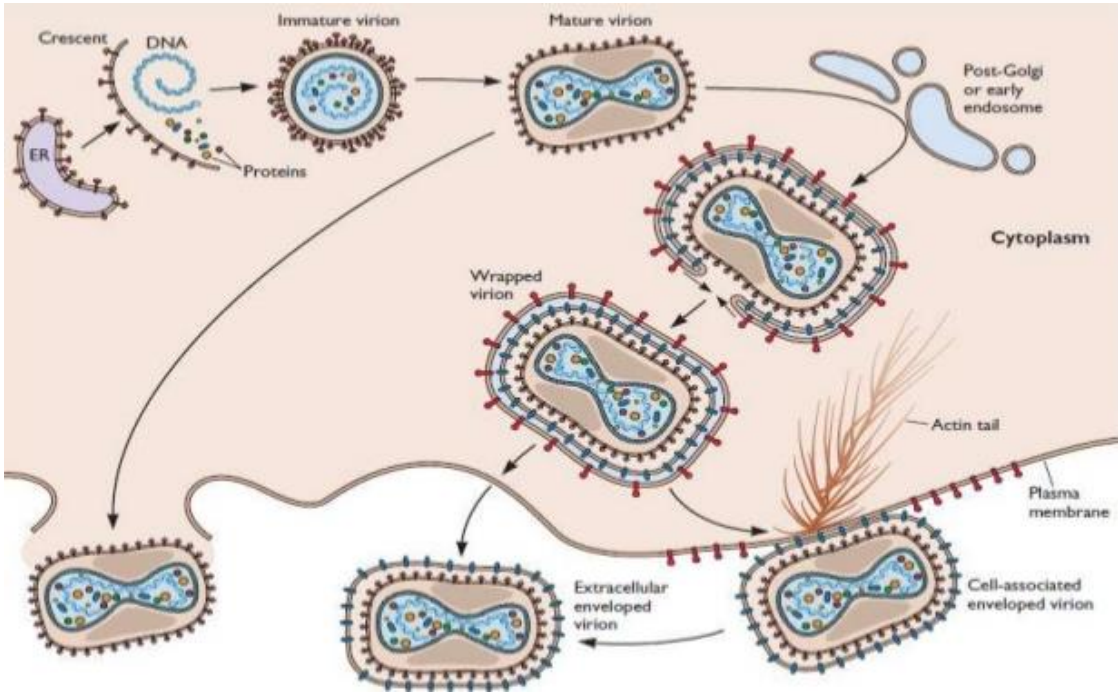


Genes and proteins

- Postreplicative mRNAs have 5’ end poly(A) extensions and 3’ end heterogeneity
  - A slippage mechanism during transcription initiation adds “poly(A) heads”
  - Heterogenous 3’ ends result from inefficient transcription termination at multiple sites

Assembly

- Mature virions are formed within virus “factories”
  - Crescent-shaped membrane structures form de novo and are destined to become the mature viral envelope
  - Crescents mature into spheres and enclose **viroplasm**
- Extracellular virions are extruded through the plasma membrane by actin tails
  - Mature viruses are wrapped in Golgi derived cisternae
  - Wrapped viruses are transported to plasma membrane
  - Membranes fuse, releasing EV which is now attached to cell membrane
  - Actin tails assemble beneath forming **microvilli**



Distinctive characteristics

- Poxviruses make several proteins that target host defenses against invading pathogens
  - Poxviruses K3L and E3L inhibit interferon response by inhibiting protein kinase (PKR) and 2′,5′-oligoadenylate synthetase
  - Serpins and poxvirus soluble receptors block cytokines like IL-1 and TNF
  - Vaccinia C3L gene product binds to and inactivates components of the complement system, blocking phagocytosis of the virus

Table 26.4 **Poxvirus genes that interfere with host defense systems**

System	Virus gene	Activity	Protection against host defense
Interferon	K3L	Homologue of eukaryotic initiation factor 2α (eIF-2α)	Pseudosubstrate for interferon-induced protein kinase (PKR)
	E3L	Double-stranded RNA binding protein	Sequesters dsRNA, an activator of interferon-induced PKR and 2',5'-oligo A synthetase
	B8R	Secreted, soluble interferon-γ receptor	Binds to and inhibits interferon-γ
Interleukin-1	B14R	Serine protease inhibitor (serpin)	Inhibits proteolytic activation of interleukin-1, decreasing inflammatory response
	B16R	Soluble interleukin-1 receptor	Binds to and inhibits interleukin-1β
Tumor necrosis factor (TNF)	B28R, A53R	Soluble TNF receptor	Binds to and inhibits TNF-α and β
Complement	C3L	C4B and C3B binding protein	Inhibits classical and alternative complement pathways
Chemokines	B29R/C23L	Chemokine binding protein	Interferes with leukocyte migration