A study material for M.Sc. Biochemistry (Semester: III) Students
on the topic (CC-13; Unit I)

Cosmid and Phagemid as Vector

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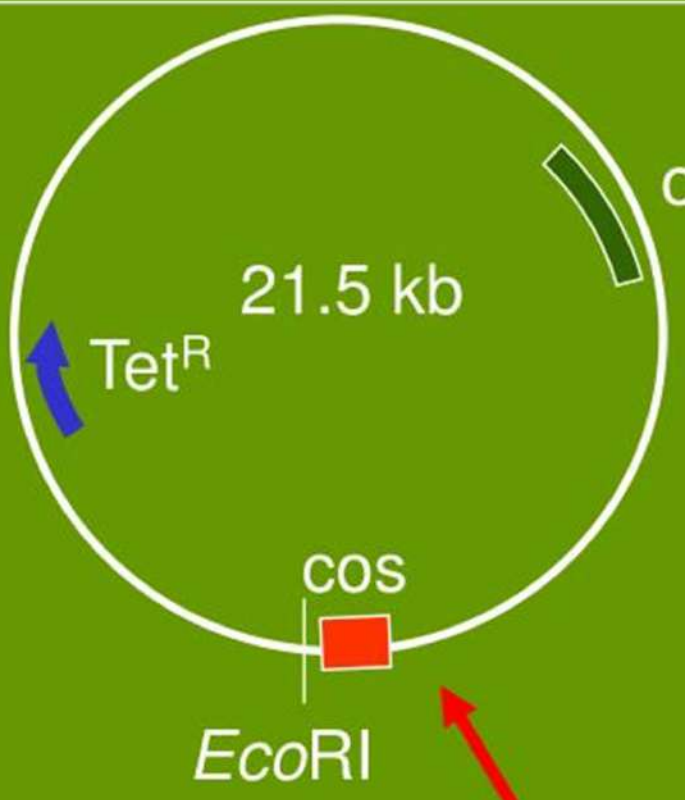
Cosmids

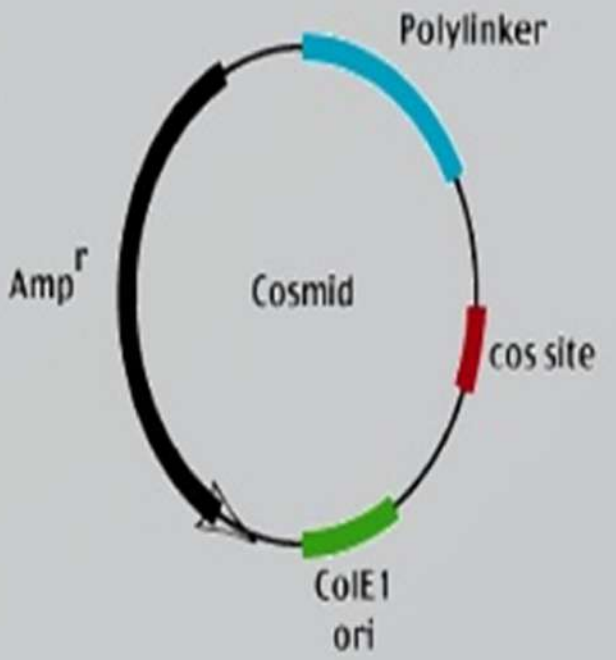
- Hybrid vectors: plasmids that contain bacteriophage lambda cos sites
- DNA (~ 33-48 kb) cloned into restriction site, the cosmid packaged into viral particles and these phages used to infect *E.coli*
- Cosmid can replicate in bacterial cell, so infected cells grow into normal colonies
- Insert DNA limited by the amount of DNA that can fit into phage capsule
- Somewhat unstable, difficult to



Cos site is the o requirement for packaging into

Cosmid

- Cosmids are plasmids that incorporate a segment of **bacteriophage λ DNA** that has the **cohesive end site (cos)** which contains elements required for packaging DNA into λ particles.
- It is normally used to clone large DNA fragments between **25 and 45 Kb.**
- They can replicate as plasmids if they have a suitable origin of replication.
- They can also be packaged in phage capsids, which allows the foreign genes to be transferred into cells by **transduction.**



Advantages :

- High transformation efficiency.
- The cosmid vector can carry up to 45 kb whereas plasmid and Lambda phage vectors are limited to 25 kb.

Cosmid vector

- Plasmids have been constructed which contain a fragment of lambda DNA including the *cos* site
- Cosmids are used as a gene cloning vector in conjunction with *in vitro* packaging system
- Ex.pJB8 (5.4 kb)

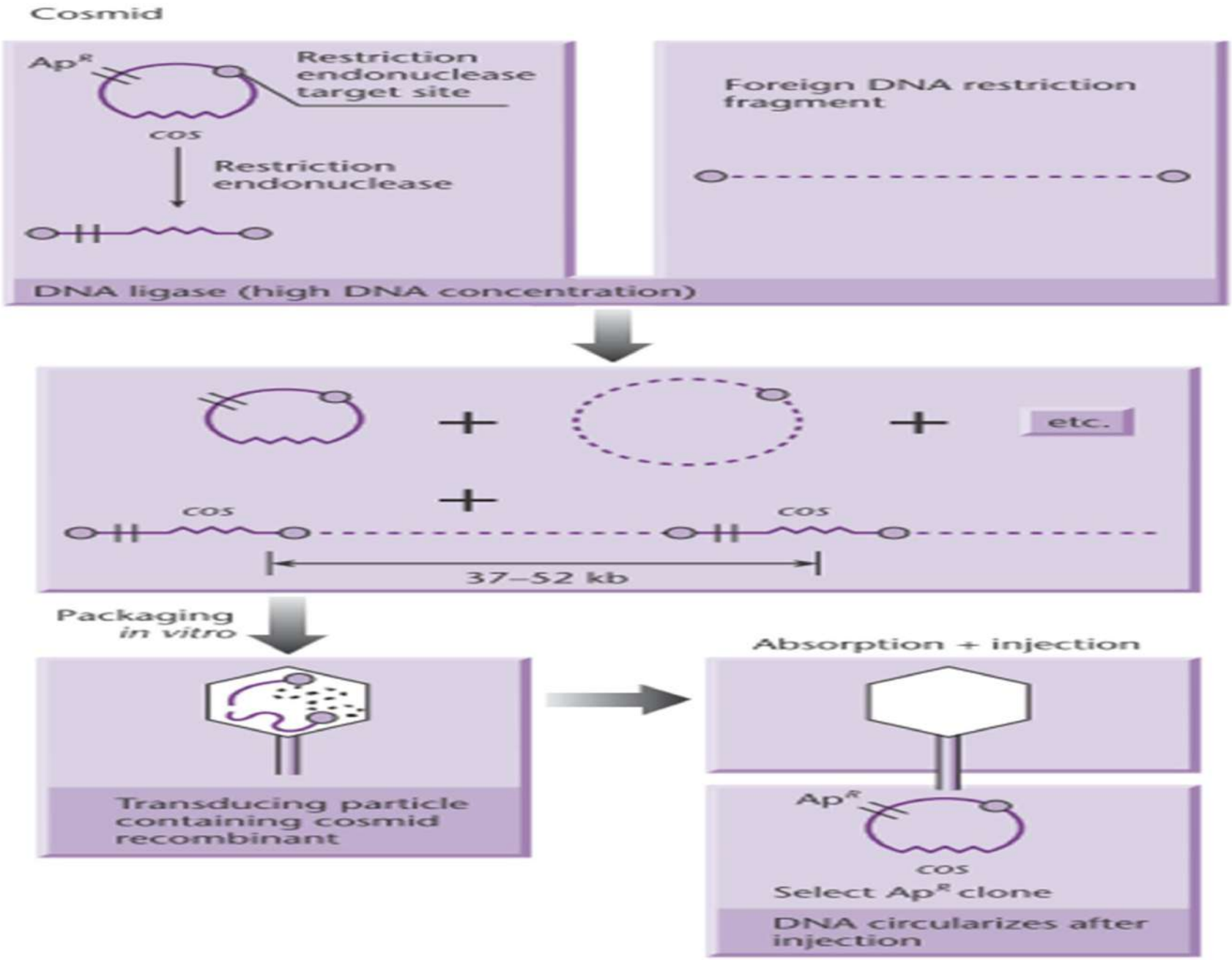


Fig. 5.1 Simple scheme for cloning in a cosmid vector. (See text for details.)

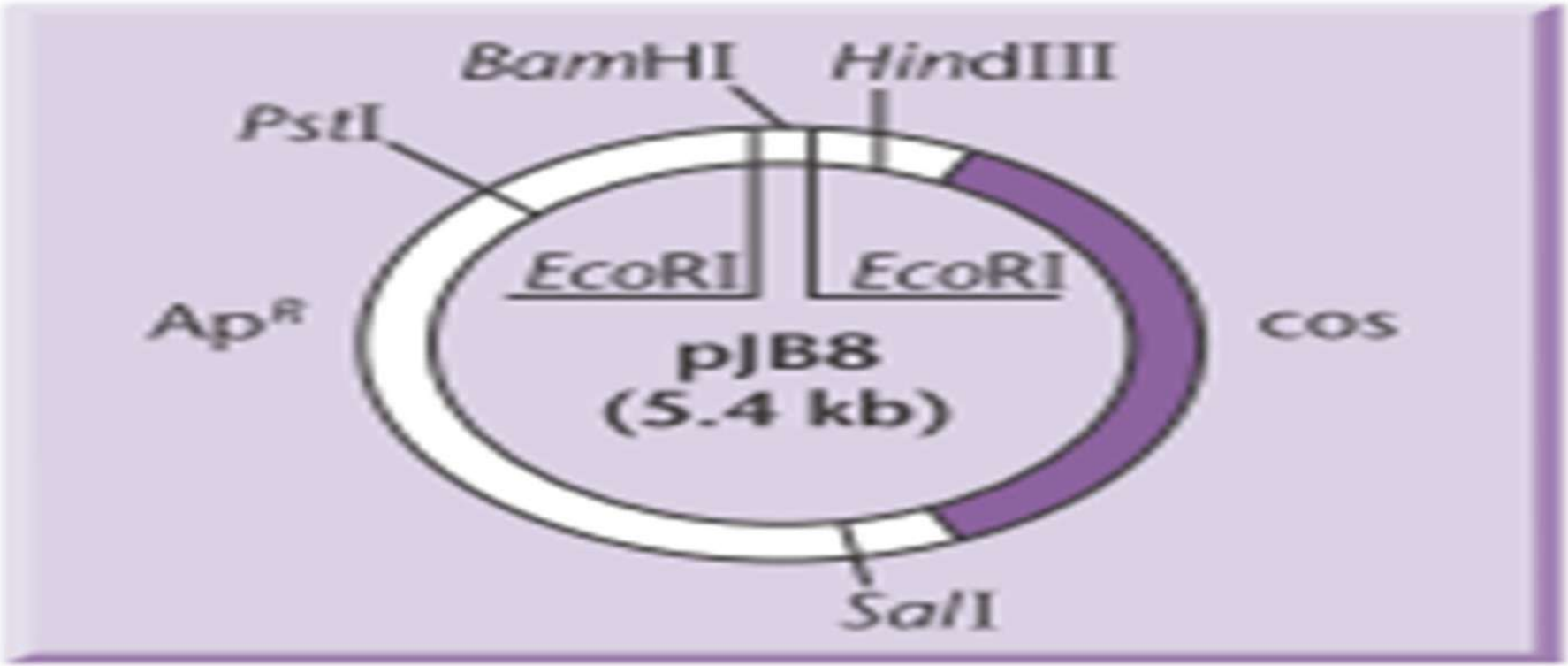


Fig. Map of cosmid pJB8 vector

Advantage:

Getting large DNA fragments inside the cell

~50 kb

Genomic library construction

Disadvantage:

Don't accept more than 50 kb fragment

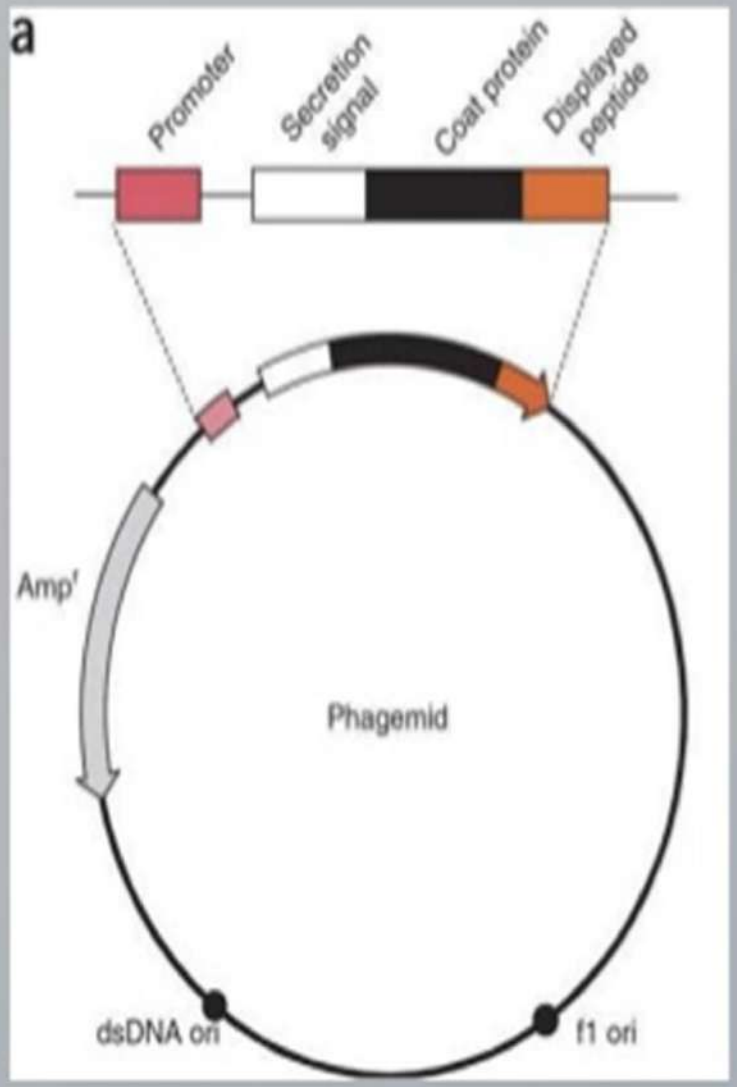
Plasmid vs Cosmid

More Information Online WWW.DIFFERENCEBETWEEN.COM

	Plasmid	Cosmid
DEFINITION	Plasmid is small extrachromosomal DNA present in prokaryotes.	Cosmid is a hybrid vector constructed by joining lambda phage DNA and plasmid DNA.
COMPOSITION	Only plasmid DNA	cos sequences of bacteriophage lambda and plasmid DNA
NATURE	Natural vectors	Constructed vectors
LENGTH OF INSERTING FRAGMENT	Up to 25 kb	Up to 45 kb
TRANSFORMATION EFFICIENCY	Comparatively less transformation efficiency	High transformation efficiency
COS SITES	Absent	Present

Phagemid

- A **phagemid** or **phasmid** is a plasmid that contains an f1 origin of replication from an f1 phage.
- It can be used as a type of cloning vector in combination with filamentous phage M13.
- A **phagemid** can be replicated as a plasmid, and also be packaged as single stranded DNA in viral particles.



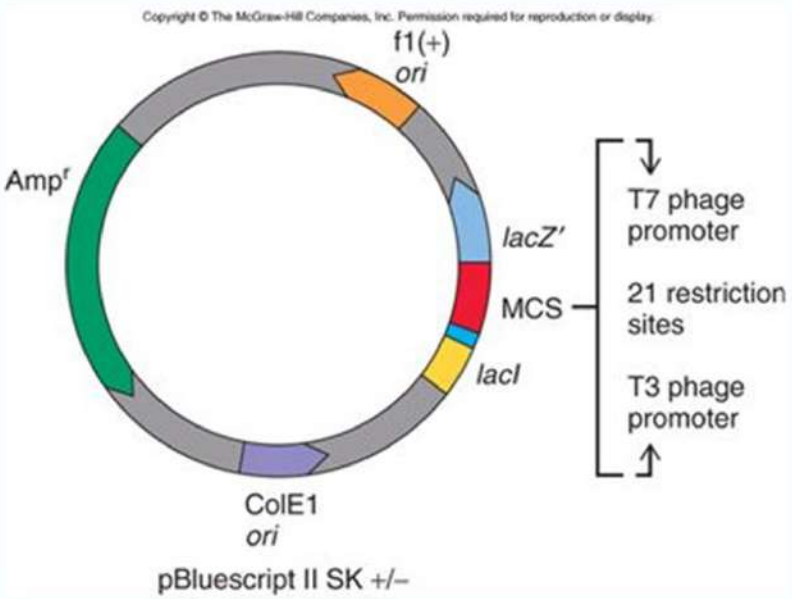
Phagemids

- Plasmid +*ori* sequence of lambda phage= phasmid
- Plasmid+ *ori* sequence of m13=phagemids
- Ex. pBluescriptKS+/- (2961 bp)
- Features:
- Derived from pUC vector
- KS represents orientation of polylinker such that transcription of *lacz* gene proceeds from the restriction site for *Kpn*I to that *Sca*I

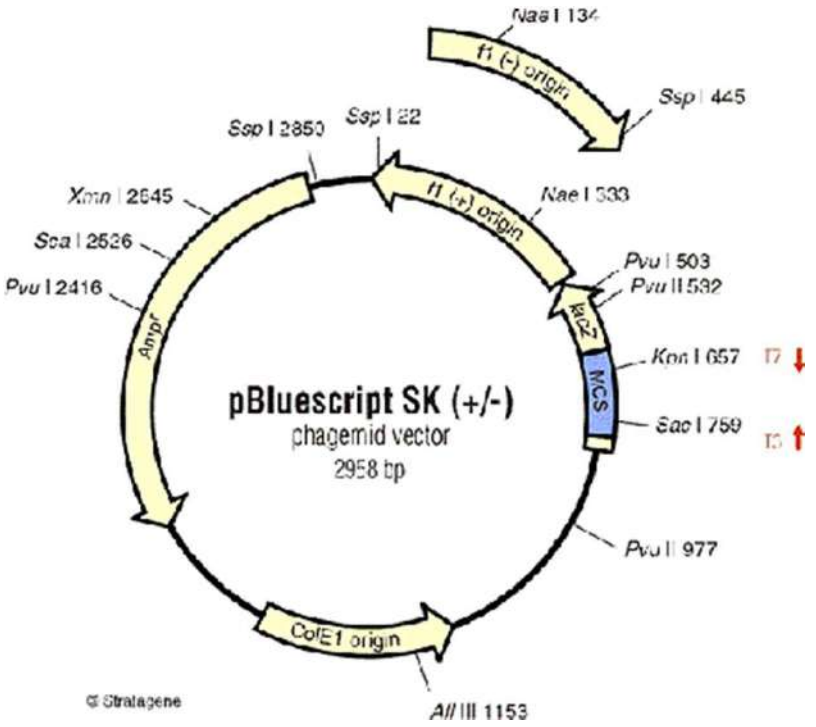
Phagemids

Phagemids are also vectors

- Like cosmids have aspects of both phages and plasmids
- Has MCS inserted into *lacZ'* gene to screen blue/ white colonies
- Has origin of replication of single-stranded phage f1 to permit recovery of single-stranded recombinant DNA
- MCS has 2 phage RNA polymerase promoters, 1 on each side of MCS



- MCS flanked by T3 and T7 promoters
- An inducible lac promoter (lacI) present
- F1 ori sequence derived from filamentous phage present
- An ori of replication (ColE1) derived from pBR322
- Ampicillin resistant gene present as selective marker



Acknowledgement and Suggested Readings:

1. Gene Cloning and DNA Analysis: An Introduction; Sixth Edition ; T. A. Brown; Wiley – Blackwell Publications
2. Principles of Gene Manipulation; Sixth Edition; Sandy B Primrose, Richard M Twyman and Robert W. Old; Wiley – Blackwell Publications
3. Biotechnology: Applying the Genetic Revolution; David P. Clark and Nanette J. Pazdernik; Academic Press (Elsevier)

Thanks