UNIT I
DNA REPLICATION

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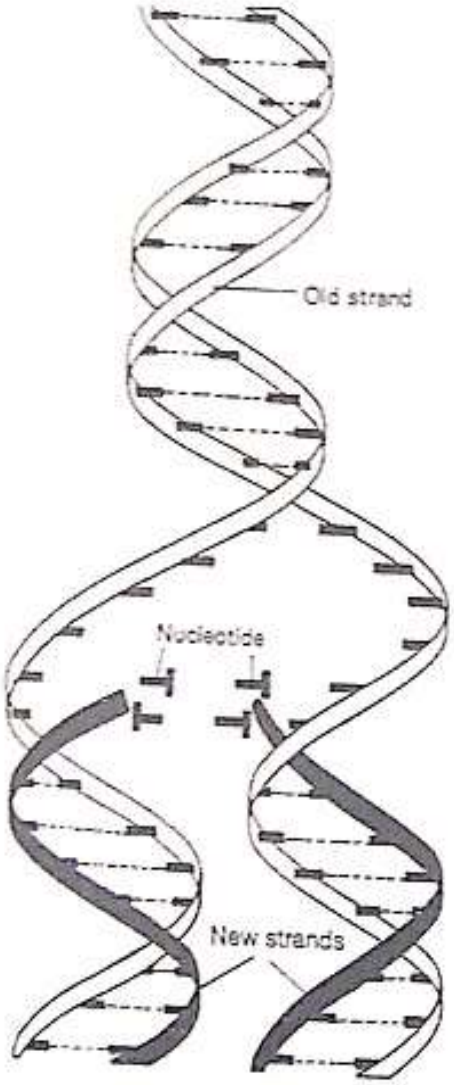
➢ Basic rules of replication:

• Replication is semi conservative process.
• Replication initiates at a DNA sequence called the origin of replication.
• The units of replication are the replicons.
• Replication requires priming.
• Replication takes place in the 5′ to 3′ direction.
• Replication forks have leading and lagging arms.
• Replication is predominantly bidirectional but may be unidirectional in some cases.



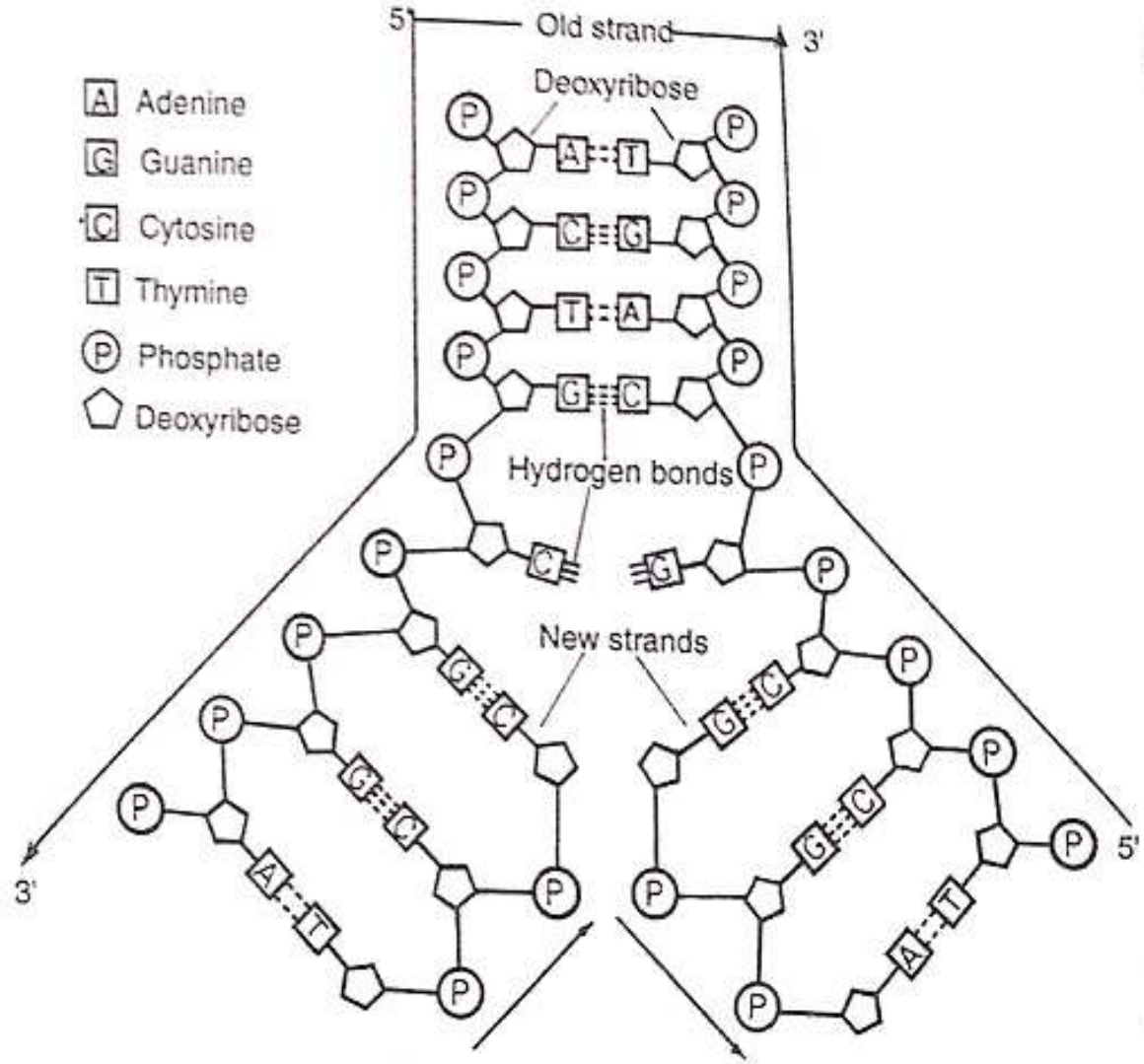
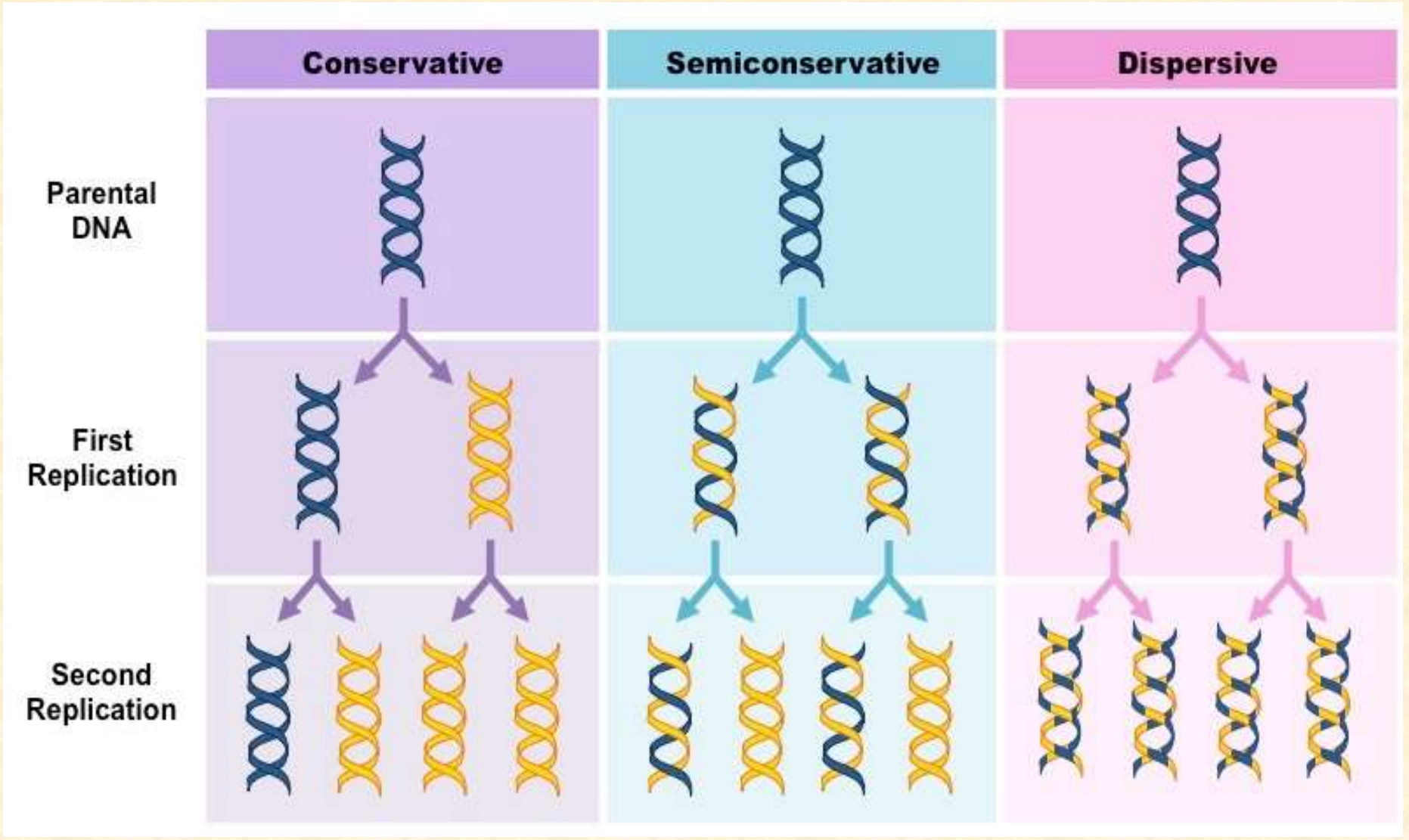


Fig. 14.1. Replication of DNA.

➢ Replication is a semi-conservative process:

• Watson and Crick were aware that any model of DNA structure should be able to explain replication.

• Delbruck suggested that the Watson – Crick model of DNA could theoretically replicate by three modes, conservative, semi-conservative and dispersive.


Conservative
Semiconservative
Dispersive
Parental DNA
First Replication
Second Replication

1) Conservative mode: Of the two double helices formed one would be entirely of old material and the other entirely of new material.

2) Semi conservative mode: Each strand of the two double helices formed would have one old and one new strand.

3) Dispersive mode : The DNA double helix would break at several points forming many pieces. Each piece would replicate, and then the pieces would reconnect at random. Thus, the two double helices formed would have a patch work of old and new pieces.

➢ Meselson – Stahl experiment:

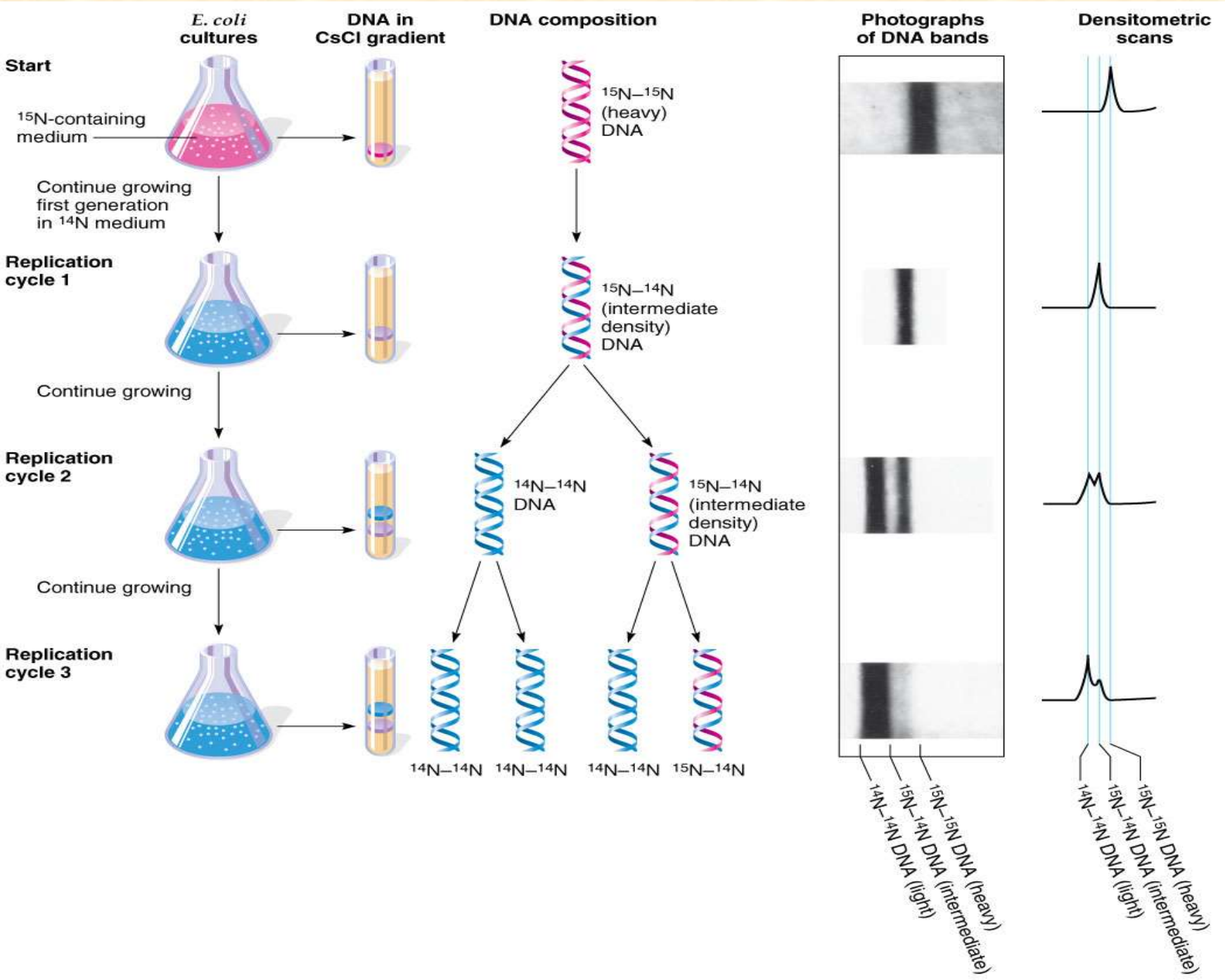
• Escherichia coli bacteria were grown for several generations in a medium containing “heavy” nitrogen (N_{15}), an isotope of nitrogen.
• All the nitrogen of the bacteria, including that of DNA, became N_{15} .
• This DNA is heavier than ordinary DNA, from which it can be distinguished by an ultracentrifuge.
• The labelled N_{15} cells were now grown in ordinary “light” N_{14} media, and allowed to divide several times.

• After the first division, the DNA was extracted and all of it was found to be a hybrid (N_{14} + N_{15}) .

• This hybrid was not as heavy as N_{15} nor as light as N_{14}, but has an intermediate density.

• After the second division, two kinds of DNA were found, normal N_{15} DNA (half) and hybrid (N_{14} + N_{15}) DNA (half).

• After the third division ¾ of the DNA was normal N_{14} and ¼ was hybrid (N_{14} + N_{15}).



• If DNA replicated conservatively one would expect to find only two layers, one of N_{14} and the other of N_{15}, in the first generation, and similarly for subsequent generations.

• With dispersive replication, tubes of all generations would be expected to show a single layer ($N_{14} + N_{15}$), since the DNA would contain both new and old material mixed up.

• In semi-conservative replication the first generation would be expected to show a hybrid $N_{14} + N_{15}$ layer. With each generation after the second the N_{14} layer would show a greater accumulation of material.

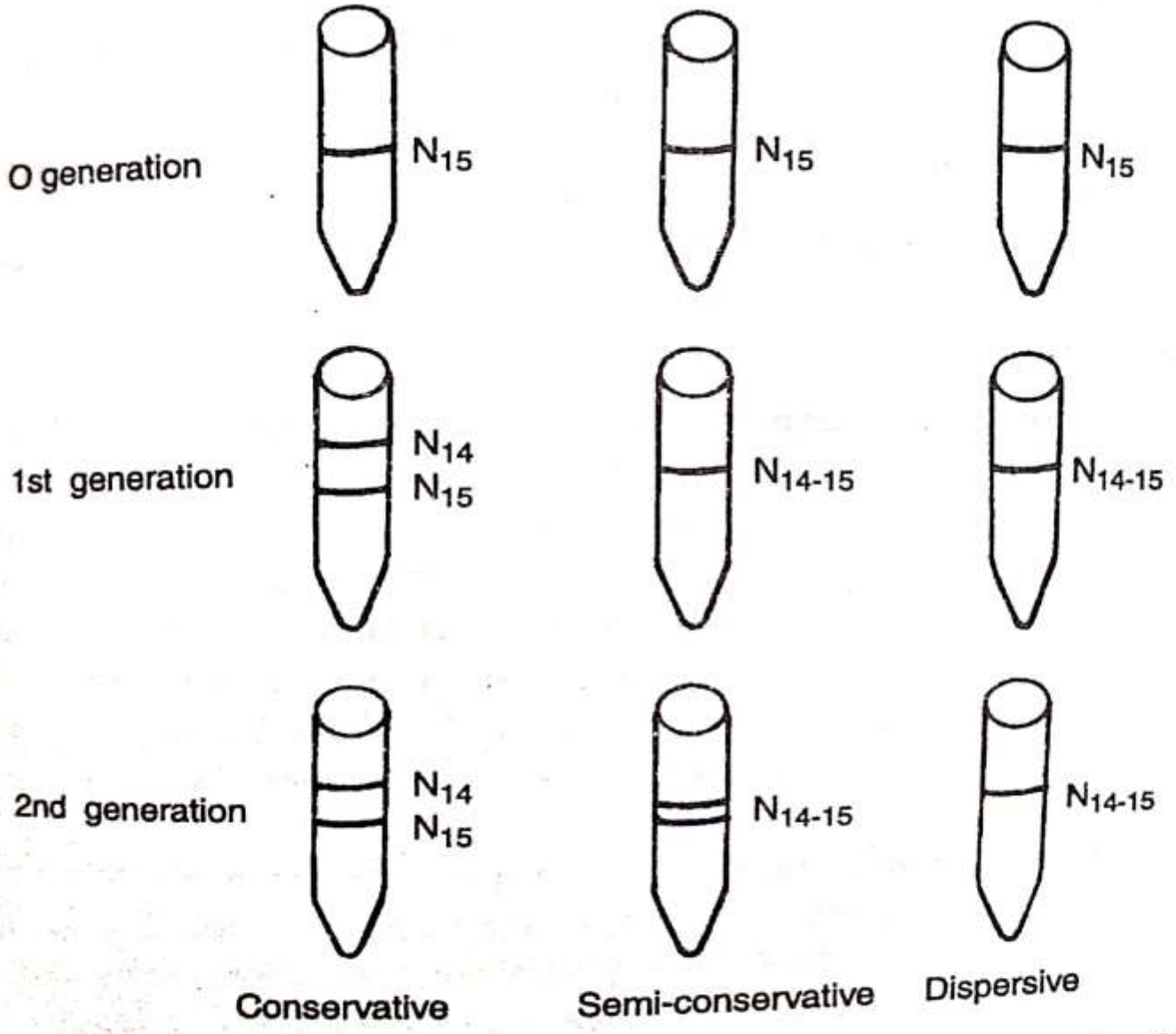


Fig. 14.4. The Meselson-Stahl experiment: expected results from conservative, semi-conservative and dispersive modes of replication.