MEGA LECTURE

IB DP BIOLOGY

Theme D: Continuity and Change

D1.1 DNA Replication

Revision Notes · Standard and Higher Level

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Original notes prepared for the IB Diploma Programme Biology course (first assessment 2025)

What the syllabus requires

By the end of D1.1 you should be able to explain how a cell copies its DNA accurately before it divides. Use this checklist as a final revision map; the rows flagged (HL) are examined only at Higher Level.

Understanding	You should be able to...
Purpose and timing of replication	State that DNA is copied during the S phase of interphase, before cell division, so that each daughter cell receives a complete genome.
Semi-conservative replication	Explain that each new DNA molecule contains one conserved (template) strand and one newly synthesised strand.
Complementary base pairing	Use the base-pairing rules (A−T, G−C) to show how each template directs an identical copy.
Role of helicase and DNA polymerase	Describe unwinding of the helix and the assembly of a new strand from free nucleotides.
Directionality and enzymes (HL)	Explain 5′→3′ synthesis, primase, leading and lagging strands, Okazaki fragments, ligase and proofreading.
Meselson–Stahl evidence (HL)	Interpret density-gradient results that distinguish the semi-conservative model from the alternatives.
Polymerase chain reaction (PCR)	Outline denaturation, annealing and extension, and explain why a thermostable polymerase is required.

> **Exam note:** The SL mechanism is deliberately simplified (helicase + DNA polymerase). Directionality, primers, Okazaki fragments, ligase, proofreading and the Meselson–Stahl experiment are all HL-only detail — do not lose SL marks by over-complicating a 2-mark description.

Background — the molecule being copied

Replication makes far more sense once you can picture the DNA that is being copied. A quick recap of the structure (covered fully in A1.2):

- DNA is a **double helix** of two polynucleotide strands. Each nucleotide has a phosphate group, the sugar **deoxyribose**, and one of four **nitrogenous bases**: A, T, G or C.
- The strands are joined by **hydrogen bonds** between complementary bases: **A pairs with T** (two H-bonds) and **G pairs with C** (three H-bonds).
- The two strands are **antiparallel**: one runs 5′→3′ and the other runs 3′→5′. The 5′ and 3′ labels refer to carbon atoms of the sugar and fix each strand's direction.
- Because pairing is specific, the two strands carry the **same information in complementary form** — this is exactly what makes accurate copying possible.

Base pair	Hydrogen bonds	Note
Adenine – Thymine (A−T)	2	Weaker pairing; easier to separate
Guanine – Cytosine (G−C)	3	Stronger; GC-rich DNA needs more heat to melt

> **Why it matters here:** The number of hydrogen bonds explains why denaturation in PCR needs heat, and why the pairing rules force the new strand's sequence. Keep A−T = 2 bonds and G−C = 3 bonds at your fingertips.

1. Why DNA must be replicated

DNA is the store of genetic information. Every time a cell divides by mitosis, the two daughter cells must each receive a **complete and identical** copy of that information, so the DNA has to be duplicated *before* division takes place. Without prior replication, division would simply halve the amount of DNA and the daughter cells would be missing genes.

When does it happen?

Replication occurs during the **S (synthesis) phase** of interphase, the long growth stage of the cell cycle that precedes mitosis. The sequence of events is:

- **G1 phase** → cell grows, makes proteins and organelles; each chromosome is a single DNA molecule.
- **S phase** → DNA is replicated; each chromosome now consists of two identical sister chromatids.
- **G2 phase** → further growth and checking of the copied DNA before division.
- **M phase** (mitosis) → sister chromatids are separated into the two daughter cells.

Because replication and division are separated in time, each daughter cell ends up with exactly one copy of every DNA molecule — the basis of genetic continuity.

> **Key idea:** Replication makes the copy; mitosis distributes the copies. The two are distinct stages, and DNA is only ever copied once per cell cycle.

2. Semi-conservative replication

DNA replication is described as **semi-conservative**. The prefix *semi* means 'half': each new double helix is built from one **old** strand (conserved from the parent molecule and used as a template) and one **newly synthesised** strand. So each of the two daughter molecules is *half original and half new*.

Why one strand is kept as a template

Because the strands are complementary and antiparallel, each one carries the full sequence information needed to specify its partner: a base on one strand always pairs with one particular base on the other. When the strands separate, the old strand therefore acts as a precise mould for building a new, complementary partner, and no information is lost.

Model	What it would predict	Observed?
Semi-conservative	Each daughter molecule = 1 old strand + 1 new strand	Yes — this is what happens
Conservative	Parent molecule stays intact; a completely new molecule is made	No
Dispersive	Old and new DNA are broken up and mixed along both strands	No

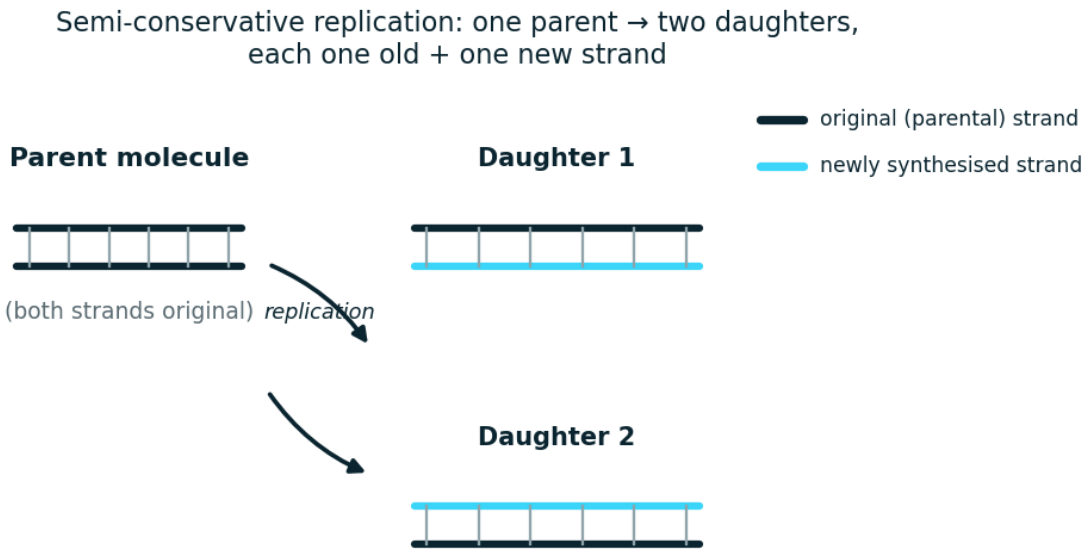

Figure 1. Semi-conservative replication: one parent molecule → two daughters, each with one original (dark) strand and one newly synthesised (cyan) strand.

Evidence — the Meselson–Stahl experiment (HL)

The semi-conservative model was confirmed by growing bacteria for many generations in a medium containing a **heavy** nitrogen isotope (^{15}N), so that all their DNA became dense. The bacteria were then switched to a **light** (^{14}N) medium, and DNA was extracted after each round of replication and spun in a density gradient, where molecules settle at a position matching their density.

- **Generation 0:** all DNA is heavy ($^{15}N/^{15}N$) — a single low band.
- **After 1 round:** all DNA is of *intermediate* density (one ^{15}N + one ^{14}N strand) — a single middle band.
- **After 2 rounds:** two bands appear — half intermediate, half fully light ($^{14}N/^{14}N$).

The single intermediate band after one round rules out the conservative model (which predicts a heavy band plus a light band). The appearance of a distinct light band after two rounds, with the intermediate band persisting, rules out the dispersive model (which predicts all molecules becoming progressively and uniformly lighter). Only the semi-conservative model fits both results.

> **Exam technique (HL):** Learn to reason from the band positions, not just to recall them. 'One intermediate band after one generation' is the single most important observation — it is exactly what semi-conservative replication predicts and what conservative replication cannot produce.

3. The mechanism at SL

At Standard Level you need a clear, ordered account using two named enzymes. The process converts one parent double helix into two identical daughter molecules.

- **Unwinding:** the enzyme **helicase** moves along the DNA, unwinding the double helix and breaking the hydrogen bonds between complementary bases, separating the two strands.
- **Templating:** each separated strand is exposed and acts as a **template** that carries the information for building a new partner strand.

- **Base pairing:** free DNA nucleotides in the nucleus line up against the template, pairing complementarily — **adenine with thymine** (A−T) and **guanine with cytosine** (G−C).
- **Joining:** the enzyme **DNA polymerase** catalyses the formation of covalent (phosphodiester) bonds between adjacent nucleotides, building a continuous new sugar–phosphate backbone.
- **Result:** two DNA molecules are formed, each identical to the original and to each other, and each with one old and one new strand.

Notice that because both strands are copied at the same time, one round of replication turns one molecule into two. The accuracy comes entirely from the base-pairing rules described in the next section.

> **SL mark scheme tip:** A full-mark SL description usually needs: helicase unwinds / breaks hydrogen bonds; strands act as templates; complementary base pairing (A−T, G−C); DNA polymerase joins nucleotides; two identical molecules with one old and one new strand.

4. Complementary base pairing ensures accuracy

The reason replication produces two *identical* copies is the strict complementary base pairing between the template and the incoming nucleotides. Because A only pairs with T, and G only pairs with C, the sequence of the template strand completely determines the sequence of the new strand.

Worked logic: if a stretch of template reads

template 3′ – T A C G G A T – 5′

then the only sequence the new strand can have is

new 5′ – A T G C C T A – 3′

Any other base would fail to pair, so — barring rare errors — the copy must match the sequence that was originally opposite the template. Since both original strands are copied, the two daughter molecules carry exactly the same genetic information as the parent. This faithful copying is what allows genes to be passed unchanged from a cell to its descendants.

> **Why 'complementary', not 'identical':** The new strand is complementary to its template (A opposite T, etc.), but the resulting double-stranded molecule is identical to the parent double helix. Keep the two ideas separate in your wording.

5. Enzymes and directionality (HL)

At Higher Level you must explain *how* the strands are actually built, which requires the concept of directionality and several more enzymes. The site where the double helix is opened and copied is called the **replication fork**.

The 5′ → 3′ rule

DNA polymerase can add new nucleotides **only** to the 3′ end of a growing strand — that is, it synthesises exclusively in the 5′→3′ direction. Because the two template strands are **antiparallel** (one runs 3′→5′, the other 5′→3′), the two new strands cannot be made in the same simple way.

Enzyme / component	Role in replication
Helicase	Unwinds the double helix and breaks hydrogen bonds at the replication fork.
Primase	Synthesises a short **RNA primer** that provides a free 3′ end for DNA polymerase to build on.
DNA polymerase III	Adds free DNA nucleotides to the 3′ end of the primer, extending the new strand 5′→3′; also proofreads.
DNA polymerase I	Removes RNA primers and replaces them with DNA nucleotides.
DNA ligase	Seals the remaining gaps (nicks) in the sugar–phosphate backbone, joining fragments into a continuous strand.

Leading and lagging strands

- The **leading strand** is built **continuously**, following the fork as it opens, because its 5′→3′ synthesis runs in the same direction as the fork moves.
- The **lagging strand** runs the opposite way, so it is built **discontinuously**, in short pieces called **Okazaki fragments**, each begun with its own RNA primer and made away from the fork.
- DNA polymerase I removes the primers, and **DNA ligase** then joins the Okazaki fragments into one continuous lagging strand.

The two new strands at a fork, side by side:

Feature at the fork	Leading strand	Lagging strand
Synthesis vs fork movement	same direction (toward the fork)	opposite direction (away from the fork)
Continuity	continuous	discontinuous — Okazaki fragments
Primers required	one, at the start	one for every fragment
Finishing step	already continuous	DNA ligase joins the fragments

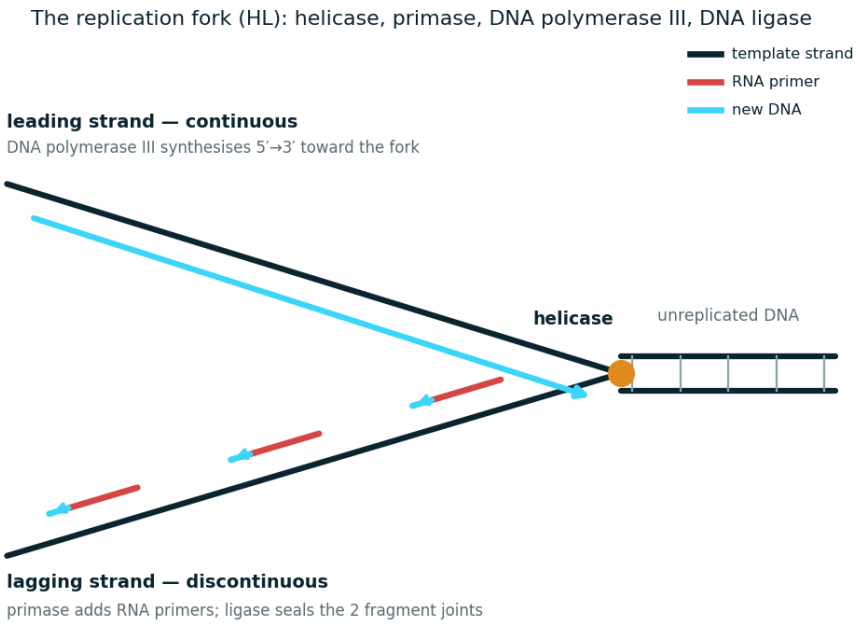

Figure 2. The replication fork (HL): helicase unwinds the helix; the leading strand is synthesised continuously toward the fork; the lagging strand is built away from the fork as primed Okazaki fragments later sealed by ligase.

Proofreading and accuracy

DNA polymerase also has a **proofreading** function: as it works it checks that each newly added nucleotide is correctly paired, and it can remove a mismatched base and replace it before moving on. This lowers the error rate dramatically and is a major reason replication is so accurate. Errors that slip through become the raw material of mutation (Section 7).

> **Common HL confusion:** Leading vs lagging is defined relative to the *fork direction*, not to any fixed strand. At the fork, one template gives a leading strand and the other gives a lagging strand — both new strands still grow 5′→3′.

6. Application — the polymerase chain reaction (PCR)

PCR uses the natural machinery of replication to make millions of copies of a chosen DNA sequence **in vitro** (in a test tube). It is one of the most important techniques in modern biology. A single reaction mixture contains the target DNA, a supply of free nucleotides, two short primers that flank the target, and a heat-stable DNA polymerase; it is then cycled through three temperatures.

Step	Temperature	What happens
1. Denaturation	≈ 95 °C	Heat breaks the hydrogen bonds and separates the double helix into two single strands (the natural role of helicase is done by heat).
2. Annealing	≈ 55 °C	Cooling lets the short primers bind (anneal) to their complementary sequences at the ends of the target region.
3. Extension	≈ 72 °C	**Taq polymerase** extends each primer 5′→3′, building new complementary strands from free nucleotides.

Why Taq polymerase?

The denaturation step reaches about 95 °C, which would destroy a normal (human or bacterial) enzyme. **Taq polymerase**, obtained from a heat-tolerant bacterium that lives in hot springs, is **thermostable** — it survives the high temperatures and stays active through every cycle, so fresh enzyme does not have to be added each time. This is what made automated, repeated cycling practical.

Amplification and uses

Each cycle doubles the amount of the target sequence, so after *n* cycles the number of copies rises to about 2^n — roughly a million-fold after 20 cycles. PCR is used to:

- amplify tiny DNA samples for **forensic** analysis and DNA profiling;
- **diagnose** infections by detecting a pathogen's DNA or RNA, and to detect genetic disorders;
- provide enough DNA for **gene cloning, sequencing and research**.

> **Link:** PCR mimics replication but replaces helicase with heat and uses primers that the scientist designs to select exactly which region is copied.

7. When replication goes wrong

Replication is extraordinarily accurate, but not perfect. Occasionally an incorrect nucleotide is inserted and escapes proofreading. A change in the base sequence of DNA is a **mutation** (studied in detail in D1.3). Key points to connect:

- Uncorrected errors alter the base sequence, and are then copied faithfully in every subsequent round of replication — the change becomes permanent.
- If the error occurs in a body (somatic) cell it affects only that cell's descendants; if it occurs in a gamete-forming cell it can be inherited.
- Most errors are silent or harmful, but some produce new alleles that are the ultimate source of **genetic variation** and hence the raw material for evolution.
- Proofreading by DNA polymerase and other repair systems keep the natural error rate very low, which is why offspring resemble their parents so closely.

> **Exam framing:** If a question asks why replication must be accurate, tie it to consequences: errors change genes, are inherited by all daughter cells, and can disrupt protein structure or cause disease — while occasionally driving variation.

8. Worked examples and exam skills

Worked example 1 — strands after several rounds

A single DNA molecule with two original (parental) strands is replicated for 3 complete rounds. (a) How many double-stranded molecules result? (b) How many of them contain an original parental strand?

(a) Each round doubles the number of molecules: after *n* rounds there are 2^n molecules, so after 3 rounds there are 2^3 = **8 molecules**.

(b) There are only **2** original strands in total, and each stays intact and ends up in a separate molecule. So exactly **2 of the 8 molecules** contain an original strand; the other 6 are made entirely of new DNA. This is a direct consequence of semi-conservative replication.

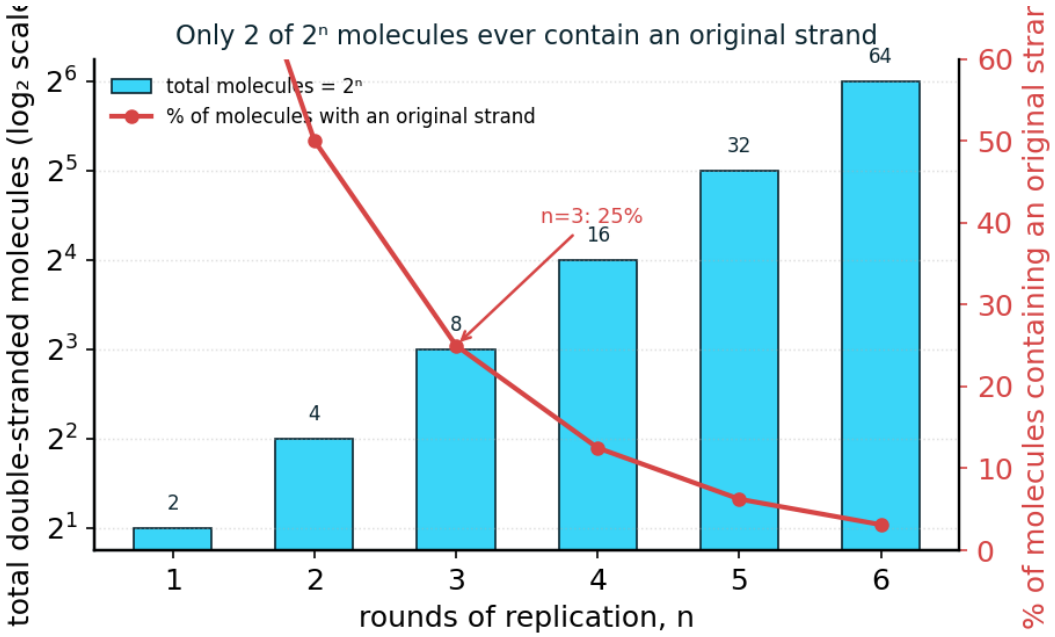

Figure 3. As replication proceeds, the number of molecules grows as 2^n but only 2 molecules ever retain an original strand -- so the fraction with an original strand falls as $2/2^n$ (25% at n = 3, matching Worked Example 1).

Worked example 2 — writing the complementary strand

A template strand reads 3′ – T A C G A C T T G – 5′. Write the sequence of the new strand produced, with polarity labelled.

Apply the pairing rules base by base (A−T, G−C) and remember the new strand is antiparallel:

new strand: 5′ – A T G C T G A A C – 3′

Check: the new strand read 5′→3′ is the mirror-complement of the template read 3′→5′. Each A faces a T and each G faces a C.

Worked example 3 — order the steps (HL)

Put the following into the correct sequence for one region of the lagging strand: DNA ligase seals nicks; primase lays an RNA primer; helicase unwinds the helix; DNA polymerase III extends from the primer; DNA polymerase I replaces the primer with DNA.

Correct order: **helicase unwinds → primase lays an RNA primer → DNA polymerase III extends** the Okazaki fragment 5′→3′ → **DNA polymerase I replaces the primer** with DNA → **DNA ligase seals** the remaining nick.

Worked example 4 — interpreting a Meselson–Stahl result (HL)

Bacteria grown in ^{15}N are moved to ^{14}N. After the FIRST round of replication all the extracted DNA forms a single band of intermediate density. What does this show, and what does it rule out?

A single band of intermediate density means every molecule contains one heavy (^{15}N) template strand and one light (^{14}N) new strand — exactly the prediction of the **semi-conservative** model.

It **rules out the conservative model**, which would give two separate bands after one round (an intact heavy parent plus a fully light new molecule). The result is also consistent with dispersive at this stage, which is why a second round is needed to distinguish them.

Worked example 5 — counting copies and nucleotides

A 500 base-pair target is amplified by PCR. (a) Approximately how many copies exist after 10 cycles from a single starting molecule? (b) Roughly how does the copy number change if you run 20 cycles instead?

(a) Copies $\approx 2^n = 2^{10} =$ **1024** (about a thousand) copies.

(b) $2^{20} \approx 1.05 \times 10^6$, i.e. roughly a **million-fold** — doubling the cycle count squares the number of copies, showing why PCR amplifies so powerfully.

Worked example 6 — base composition (Chargaff)

A sample of double-stranded DNA is found to contain 30% adenine. Calculate the percentage of each of the other three bases, and explain your reasoning.

Because A pairs only with T, %T = %A = **30%**. That leaves 100 − 30 − 30 = 40% shared equally between G and C, which also pair 1:1.

So %G = %C = 40 / 2 = **20% each**. In any double-stranded DNA, %A = %T and %G = %C — a direct consequence of complementary base pairing (Chargaff's rule).

Common pitfalls

- Saying 'semi-conservative' means half of *each strand* is new. It means each daughter molecule keeps one *whole* old strand and gains one *whole* new strand.

- Claiming DNA polymerase can build in either direction. It adds nucleotides **only** to a 3′ end, so synthesis is always 5′→3′. (HL)
- Mixing up leading and lagging strands, or forgetting the lagging strand is made in Okazaki fragments joined by ligase. (HL)
- Confusing complementary with identical: the new strand is complementary to the template; the finished double helix is identical to the parent.
- Forgetting that replication happens in S phase, *before* mitosis — not during it.
- In PCR, writing that helicase separates the strands. In PCR the strands are separated by **heat**, and Taq polymerase must be thermostable to survive it.

Quick reference

Term / step	Summary
Timing	S phase of interphase, before cell division.
Semi-conservative	Each daughter molecule = 1 old (template) strand + 1 new strand.
Base pairing	A—T (2 H-bonds) and G—C (3 H-bonds); ensures accuracy.
Helicase	Unwinds helix, breaks hydrogen bonds.
DNA polymerase	Joins free nucleotides into the new strand (5′→3′); proofreads.
Primase (HL)	Lays RNA primers giving a 3′ start point.
Leading / lagging (HL)	Continuous vs discontinuous (Okazaki fragments).
DNA ligase (HL)	Seals nicks / joins Okazaki fragments.
Meselson–Stahl (HL)	One intermediate band after 1 round = semi-conservative.
PCR	95 °C denature → 55 °C anneal → 72 °C extend (Taq); copies ≈ 2^n.

Test yourself

Attempt these without notes; full answers follow. HL-only items are flagged.

1. State precisely when in the cell cycle DNA replication occurs and explain why it must happen before division.

2. Explain what is meant by semi-conservative replication.

3. Describe, at SL, the roles of helicase and DNA polymerase in replication.

4. A template strand reads 3′ – C C G A T A G C T – 5′. Write the new strand with its polarity.

5. (HL) Explain why the lagging strand is synthesised discontinuously.

6. (HL) In a Meselson–Stahl experiment, predict the banding pattern after two rounds of replication in ^{14}N and explain what it shows.

7. Outline the three temperature steps of one PCR cycle and give the approximate temperature of each.

8. Explain why a thermostable polymerase such as Taq is essential for PCR, and give one use of PCR.

Answers

1. DNA is replicated in the **S phase** of interphase. It must occur before mitosis so that each daughter cell receives a complete, identical copy of the genome; otherwise division would leave cells lacking genes.

2. Each new double helix consists of one **original (conserved) strand** used as a template and one **newly synthesised strand**; thus each daughter molecule is half old and half new.

3. **Helicase** unwinds the double helix and breaks the hydrogen bonds between paired bases, separating the strands. **DNA polymerase** joins free complementary nucleotides (A−T, G−C) that pair with each template, forming the new strand.

4. New strand: **5′ – G G C T A T C G A – 3′** (each base complementary to the template, antiparallel).

5. (HL) DNA polymerase only synthesises 5′→3′. The lagging-strand template runs the 'wrong' way relative to the fork, so the strand is built away from the fork in short **Okazaki fragments**, each primed separately and later joined by DNA ligase.

6. (HL) After two rounds there are **two bands**: one **intermediate** (one ^{15}N + one ^{14}N strand) and one **fully light** ($^{14}N/^{14}N$). The persistence of the intermediate band alongside a new light band confirms semi-conservative replication and rules out the dispersive model.

7. **Denaturation** ≈ 95 °C (strands separate) → **annealing** ≈ 55 °C (primers bind) → **extension** ≈ 72 °C (Taq extends the primers).

8. Denaturation reaches ≈ 95 °C, which would denature an ordinary enzyme. **Taq** is thermostable, so it stays active through every cycle without replacement. Use: forensic DNA profiling, or diagnosis of infection/genetic disease (any valid use).