Collegedunia NCERT Formula Sheet

Class 12 Biology Formula Sheet — Chapter 10 Biotechnology and its Applications (NCERT 2026-27)

Chapter 10: Biotechnology and its Applications

Quantitative biotechnology — Bt toxin & *cry* genes, RNAi, recombinant insulin, ADA gene therapy, ELISA, PCR, transgenic animals, GEAC norms

Also see for this chapter: **NCERT Solutions** | **Revision Notes** | **Exemplar Solutions**

Chapter-Wide Key Quantitative Reference

Parameter / Fact	Value / Range	Significance
Recombinant therapeutics approved	~ 30 worldwide; 12 marketed in India	rDNA-derived drugs (insulin, GH, EPO, ...)
Bt toxin gut-activation pH	≥ 9.5 (alkaline insect midgut)	solubilises Cry crystal $\rightarrow$ active toxin
Bt toxin selectivity	lepidoptera, coleoptera, diptera	harmless to mammals; gut pH ~ 2 in humans
Human insulin chain A length	21 amino acids	joined to chain B by S–S bridges
Human insulin chain B length	30 amino acids	joined to chain A by S–S bridges
Disulphide bridges in insulin	3 (two interchain + one intrachain on A)	confer mature 3-D fold
First gene therapy (ADA-SCID)	1990, 4-year-old girl, USA	ADA deficiency cured with retroviral cDNA
Insulin commercial launch	Eli Lilly, 1983, USA	A and B chains in *E. coli* plasmids
Transgenic Rosie milk (1997)	2.4 g human α-lactalbumin / L	human-protein-enriched cow milk
Transgenic animals — mice share	$> 95\,\%$ of all existing	disease models, drug testing
Basmati biopiracy patent	1997 USA patent on Indian Basmati	landmark biopiracy case
Rice varieties in India	$\sim 200{,}000$	27 documented Basmati varieties
Indian regulator	GEAC (Genetic Engineering Approval Committee)	approves GM research and release

1 10.1 Biotechnology in Agriculture — Bt Crops

Three options for raising food output — agrochemicals, organic agriculture, and GM crops — with focus on **Bt cotton** (*cry* genes from *Bacillus thuringiensis*) and tissue-culture micropropagation as supporting tools.

Bt toxin from *Bacillus thuringiensis* — activation

Source: sporulating *Bacillus thuringiensis* forms parasporal **Cry protein crystals**.

Activation sequence in insect midgut:

Step 1. Cry protoxin (crystal, ~ 130 kDa) — solubilised by **alkaline pH** ≥ 9.5 of midgut juice.

Step 2. Soluble protoxin — cleaved by **midgut serine proteases** to active toxin (~ 65 kDa).

Action: active toxin binds midgut epithelial receptors $\rightarrow$ inserts into membrane $\rightarrow$ pores $\rightarrow$ osmotic cell swelling and lysis $\rightarrow$ larva dies in ~ 48 h.

Toxin does **not** kill the host *Bacillus* because it exists as an **inactive protoxin crystal** inside the spore. Human gut pH (~ 2, acidic) cannot solubilise the crystal, so Bt is **safe to ingest**.

cry gene catalogue — Bt crops

cry gene	Encodes toxin against	Crop carrying the gene
cryIAc	cotton bollworm (*Helicoverpa*)	**Bt cotton**
cryIIAb	cotton bollworm	**Bt cotton**
cryIAb	corn borer (*Ostrinia*)	**Bt corn** (Bt maize)
cryIIIA	coleopteran beetles	potato, brinjal

Naming: lower-case *cry* (gene) gives upper-case Cry (protein); roman numeral = family, capital letter = subfamily. Each Cry is **insect-order specific**: lepidoptera, coleoptera, or diptera only — non-target species are unaffected.

RNAi — silencing the nematode

Pest: *Meloidogyne incognita* (root-knot nematode) infects tobacco roots.

Strategy: introduce nematode-specific gene into host using *Agrobacterium* vector so the plant cell makes **both sense and antisense RNA**:

$$\text{sense RNA} + \text{antisense RNA} \longrightarrow \text{dsRNA}$$

Step 1. dsRNA is cleaved by **Dicer** (RNase III enzyme) into siRNA duplexes (21–23 nt).

Step 2. siRNA is loaded onto **RISC** (RNA-induced silencing complex) and the antisense strand guides cleavage / translational block of the complementary mRNA.

Nematode's specific mRNA is silenced inside the host plant cell $\Rightarrow$ parasite **cannot survive** in the transgenic root. RNAi is a **cellular defence** present in all eukaryotes; natural triggers are viral dsRNA or transposons.

Three options for raising food output

NCERT lists: **(i) agrochemical-based agriculture** (Green Revolution; high yield but costly inputs), **(ii) organic agriculture** (no synthetic chemicals; lower yield but eco-friendly), **(iii) genetically engineered crops** (Bt, RNAi, golden rice; high yield with reduced inputs but raises biosafety / ethical concerns).

GMO benefits — five NCERT-listed outcomes

GM has: (i) made crops **tolerant to abiotic stresses** (cold, drought, salt, heat); (ii) **reduced reliance on chemical pesticides** (pest-resistant Bt crops); (iii) **reduced post-harvest losses**; (iv) **increased mineral-use efficiency** (slows soil fertility loss); (v) **enhanced nutritional value** — e.g. **golden rice** engineered to make β-carotene (provitamin A).

Tissue culture toolkit — micropropagation, somaclones, somatic hybrids

Totipotency = any plant cell/explant can regenerate the whole plant. **Micropropagation** = mass production of plants from explants on nutrient medium (sucrose + salts + vitamins + amino acids + **auxins, cytokinins**). All offspring are genetically identical $\Rightarrow$ **somaclones**

(tomato, banana, apple). **Meristem culture** (apical / axillary) gives **virus-free plants** since meristems are virus-free. **Somatic hybridisation** = fusion of naked protoplasts from two varieties $\Rightarrow$ somatic hybrids (e.g. **pomato** = potato + tomato).

NEET extension — Cry crystal structure

A typical Cry1A protoxin is a ~ 130 kDa polypeptide. Proteolytic activation removes the C-terminal half, leaving a ~ 65 kDa active toxin with three domains: **Domain I** (pore-forming α-helices), **Domain II** (receptor binding), **Domain III** (β-sandwich, structural). Receptor on midgut = **cadherin-like protein**; secondary receptor = aminopeptidase-N.

Bt crystals do NOT kill the bacterium

A common Board MCQ: *Why don't Cry crystals kill* ***Bacillus thuringiensis*** *itself?* The answer is **(c) toxin is inactive (protoxin)** — it exists as a crystalline pro-form that only becomes lethal after alkaline solubilisation and proteolytic cleavage in the insect midgut. Do **not** answer "bacterium is resistant" or "toxin is immature".

2 10.2 Biotechnology in Medicine

Recombinant therapeutics (~ 30 approved worldwide, 12 in India), genetically engineered insulin, gene therapy for ADA-SCID, and molecular diagnosis by PCR, autoradiographic probes, and ELISA.

Human insulin — chain composition

Structure: two polypeptide chains held together by disulphide bridges.

Chain	Length	Role
A chain	21 amino acids	contains one **intra-chain** S–S bond
B chain	30 amino acids	linked to A by **two inter-chain** S–S bonds
Total	51 residues	3 disulphide bridges total

Pro-insulin in mammals: A chain — **C-peptide** — B chain (single chain), folded and bridged; C-peptide is then **cleaved** during maturation.

Mature insulin has **no C-peptide**. Recombinant production must reproduce the disulphide pattern correctly, which was the key engineering challenge.

Recombinant human insulin — Eli Lilly 1983 process

Step 1: Two DNA sequences corresponding to **chain A** and **chain B** are chemically synthesised.

Step 2: Each sequence is inserted into a separate plasmid of *E. coli*.

Step 3: Bacteria express **chain A and chain B separately**; chains are extracted.

Step 4: Chains are combined *in vitro* and **disulphide bonds are formed** chemically $\rightarrow$ mature human insulin (*Humulin*).

Recombinant insulin is **structurally identical** to natural human insulin $\Rightarrow$ no immunogenic

/ allergic response unlike older porcine or bovine insulin extracted from slaughterhouse pancreas.

Gene therapy for ADA deficiency — protocol

Disease: adenosine deaminase (ADA) deficiency $\rightarrow$ severe combined immuno-deficiency (**SCID**) because lymphocytes cannot detoxify deoxyadenosine.

First case: 1990, USA, a **4-year-old girl**.

Three escalating treatment options:

Approach	Curative?	Limitation
Bone-marrow transplant	sometimes	donor match needed
ADA enzyme replacement (injection)	no, palliative	repeated doses
Gene therapy (somatic)	partial	cells not immortal $\Rightarrow$ periodic infusion

Gene-therapy sequence: lymphocytes drawn from patient blood $\rightarrow$ grown in culture $\rightarrow$ **functional ADA cDNA** delivered using a **retroviral vector** $\rightarrow$ engineered lymphocytes returned to patient.

Permanent cure: insert ADA gene into cells at **early embryonic stages** (germline / stem-cell-level correction). Somatic therapy alone is not curative because mature lymphocytes are short-lived.

PCR for early molecular diagnosis

Why PCR works for early detection: amplifies pathogen DNA / RNA exponentially even when the organism is below the symptom-producing threshold.

Amplification: $N_n = N_0 \cdot 2^n$, where N_0 = starting copies, n = number of cycles, N_n = copies after n cycles.

Typical 30-cycle run: $N_n/N_0 = 2^{30} \approx 1.07 \times 10^9$ (about a billion-fold amplification).

Clinical uses: (i) detect **HIV** in suspected AIDS patients, (ii) detect mutations in suspected **cancer** patients, (iii) screen many other **genetic disorders**.

PCR uses thermostable **Taq polymerase**, two primers, dNTPs, and template DNA. Cycle = denaturation ($94\,^\circ$C) $\rightarrow$ annealing ($\sim 55\,^\circ$C) $\rightarrow$ extension ($72\,^\circ$C).

ELISA — antigen–antibody assay

Principle: antigen–antibody binding, made visible by an **enzyme-linked** reporter.

Standard sandwich-ELISA signal:

$$\text{Capture Ab} + \text{Antigen} + \text{Enzyme-linked Ab} + \text{Substrate} \longrightarrow \text{coloured product}$$

Detection law (typical): $A = \varepsilon \cdot c \cdot l$ (Beer–Lambert), where A = absorbance, ε = molar absorptivity of the coloured product, c = concentration, l = path length.

Antigens (proteins, glycoproteins) **or** antibodies against the pathogen can be measured. Routinely used for **HIV, hepatitis, dengue, COVID-19** serology. Highly sensitive and specific.

DNA probe — autoradiography

Reagent: single-stranded DNA or RNA tagged with a **radioactive label** (probe).

Protocol: probe hybridises by **complementary base-pairing** to the matching gene in a clone of cells → photographic film exposure detects the hybridisation site.

A clone carrying a **mutated** gene will **not** appear on the film — the probe is no longer fully complementary, so it does not hybridise. This is the molecular basis of probe-based mutation screening.

NEET extension — three pillars of biotechnology research

NCERT names three critical research areas: (i) **best catalyst** — improved organism (microbe or pure enzyme), (ii) **optimal conditions** via engineering (bioreactor design, pH, T, O_2), (iii) **downstream processing** (purification of protein / organic compound). All three together define modern industrial biotechnology.

Insulin cannot be taken orally

Insulin is a **polypeptide hormone** (51 aa). If swallowed, stomach proteases (pepsin, trypsin) digest it into amino acids before it can reach the bloodstream. Hence it must be **injected subcutaneously**. The same logic explains why most protein therapeutics are not orally bio-available.

Three molecular-diagnosis modalities in NCERT

NCERT names three rDNA-era diagnostic tools: **(i) PCR** — amplifies pathogen nucleic acid to detectable levels before symptoms appear; **(ii) radioactive DNA / RNA probes + autoradiography** — detect mutations or specific genes in cell clones by complementary hybridisation; **(iii) ELISA** — detects antigens or antibodies via enzyme-coupled colour signal. All three replace slower, less-sensitive serum / urine biochemistry for early diagnosis.

Why recombinant therapeutics are safer than animal-source biologics

Pre-1980s insulin came from **slaughterhouse pancreas** of cattle and pigs. Although chemically close to human insulin, these animal forms differ by 1–3 amino acids ⇒ elicit immune / allergic responses in some patients, and supplies were limited. Recombinant *E. coli*-derived insulin is **structurally identical** to human insulin, **free from contamination by animal pathogens**, and can be scaled to any volume by fermenter expansion.

3 10.3 Transgenic Animals

Transgenic animals carry and express a foreign gene. Over $95\,\%$ of all transgenic animals are **mice**; transgenic rats, rabbits, pigs, sheep, cows, and fish also exist. Five NCERT-named reasons for making them.

Five NCERT applications of transgenic animals

Application	Example
1. Normal physiology & development	study insulin-like growth factor (IGF) by introducing or knocking out related genes
2. Study of disease	transgenic models for **cancer, cystic fibrosis, rheumatoid arthritis, Alzheimer's**
3. Biological products	**α-1-antitrypsin** for emphysema; PKU and cystic-fibrosis biologics; transgenic cow **Rosie** (1997) produced milk with 2.4 g human α-lactalbumin / L
4. Vaccine safety testing	transgenic mice testing **polio vaccine** safety $\Rightarrow$ may replace monkey-based testing
5. Chemical safety / toxicity testing	mice sensitised to toxins give faster toxicology read-outs than non-transgenic animals

Acronym to remember the five — **PDBVC**: **P**hysiology, **D**isease, **B**iologicals, **V**accine safety, **C**hemical safety.

Rosie — first transgenic dairy cow (1997)

Year: 1997 **Animal:** *Bos taurus* (Holstein cow named Rosie).

Transgene: human gene for **α-lactalbumin**.

Yield: 2.4 g human α-lactalbumin / litre of milk.

Human-protein-enriched milk is **nutritionally more balanced for human infants** than untreated cow milk. Same general strategy ("pharming") is used to make human clotting factors, antibodies, and antitrypsin in goat / sheep milk.

Why model human disease in transgenic mice?

Mice share ~ 99 % of human genes (by class) and have a short generation time (~ 10 weeks). Inserting or knocking out a single human disease-allele yields a living model on which therapies can be screened. This drives the dominance of mice (> 95 % of transgenic animals).

NEET extension — knock-in vs knock-out vs transgenic

Transgenic = foreign gene **added** to the genome (e.g. human α-lactalbumin gene in Rosie). **Knock-in** = a specific endogenous gene **replaced** by a designed allele (point mutation, reporter fusion). **Knock-out** = a specific endogenous gene **disabled** (null allele). Methodology has shifted from random pronuclear injection to **CRISPR/Cas9**, which makes all three faster and more precise.

Pharming vs farming

Pharming (with a 'ph') = use of transgenic plants or animals to produce **pharmaceuticals** (e.g. Rosie's α-lactalbumin, ATryn antithrombin in goat milk). **Farming** = conventional cultivation / animal husbandry for food. Both can use the same species, but pharming output is a **biologic drug**, not a food product.

Stem-cell / germline vs somatic gene therapy

Somatic gene therapy targets non-reproductive cells (lymphocytes, bone marrow); the correction is **not heritable** and lasts only as long as the modified cells survive. **Germline** therapy targets embryos or gametes; the correction **is heritable** and represents a permanent cure but raises sharp ethical concerns. NCERT explicitly notes that introducing the ADA gene into early embryonic cells **could be a permanent cure** — this is the germline route.

4 10.4 Ethical Issues & Biopiracy — GEAC Norms

The regulatory and ethical frame around GM organisms in India: **GEAC** approval, the Patents (Second Amendment) Act, biopiracy cases (Basmati, turmeric, neem) and TRIPS-style international concerns.

GEAC — regulatory mandate

Full name: Genetic Engineering Approval Committee (now **Genetic Engineering Appraisal Committee**).

Set up by: Indian Government, under the Ministry of Environment, Forest & Climate Change.

Two-fold remit:

Function 1	validity of GM **research** (lab-scale, contained use)
Function 2	**safety of release** of GM organisms for public services (commercial / environmental release)

Any new GM crop (Bt cotton variant, GM mustard, GM brinjal, ...) requires GEAC clearance before commercial release. The committee also reviews GM imports and field-trial proposals.

Biopiracy — definition and landmark cases

Definition: the use of **bio-resources** by multinational companies (or others) **without proper authorisation** from the source countries / people and **without compensatory payment**.

Resource	Year / Place	Issue
Basmati rice	1997, USA	US firm patented an Indian-derived variety
Turmeric (haldi)	1990s, USA	patent on wound-healing use; later revoked
Neem	1990s, USA / EU	patent on antifungal extract; revoked

Industrialised nations are **rich financially but poor in biodiversity**; developing nations are **rich in biodiversity and traditional knowledge**. India responded with the **Patents (Second Amendment) Act** addressing patent terms, emergency provisions, and biopiracy.

Three pillars of safe biotechnology

(i) Safety: no allergenicity, no horizontal gene transfer, no harm to non-target species. **(ii) Ethics:** informed consent, biopiracy prevention, equitable benefit sharing. **(iii) Regulation:**

GEAC, IBSC (Institutional Biosafety Committees), RCGM (Review Committee on Genetic Manipulation), Patents (Second Amendment) Act, and the Cartagena Protocol.

NEET extension — Indian regulatory pyramid

Below GEAC are two layers: **RCGM** (DBT, reviews ongoing research) and **IBSC** (institute-level safety committee). Any laboratory using rDNA must first clear IBSC, then RCGM, then GEAC for release. Cartagena Protocol on Biosafety (India ratified 2003) governs international transboundary movement of LMOs (living modified organisms).

GM crop $\neq$ hybrid crop

A **hybrid** arises from conventional sexual crossing of two parental varieties. A **GM** (transgenic) crop carries a gene transferred by **recombinant DNA technology**, often across species (e.g. a bacterial gene in cotton). The Basmati patent example is contentious precisely because it claimed novelty for an output that was *largely conventional crossing* of an Indian variety with a semi-dwarf — not a transgene.

Cartagena Protocol on Biosafety — at a glance

Adopted under the **UN Convention on Biological Diversity (CBD)** at Cartagena (Colombia) in January 2000; entered into force September 2003. India ratified it the same year. It governs the safe **transboundary movement, handling, and use of LMOs** (living modified organisms — i.e. GMOs that can transmit genetic material). Key procedure: **Advance Informed Agreement (AIA)** — the exporter notifies the importing country, which can accept, reject, or impose conditions before the first shipment of a LMO meant for environmental release.

NEET extension — Bt cotton vs HT cotton

Bt cotton is **pest-resistant** (Cry kills bollworm). **HT (herbicide-tolerant) cotton / soy / maize** carry a different transgene — e.g. the *cp4-epsps* gene from *Agrobacterium* that confers tolerance to **glyphosate** (Roundup). Many commercial seeds are **stacked** — both Bt and HT in the same plant. Bt focuses on insect pests; HT focuses on weed competition; together they cut both insecticide and labour cost.

Recall — biotechnology applications by domain

"A-M-T-E" = **A**griculture, **M**edicine, **T**ransgenic animals, **E**thics.
Inside each, the headline keywords:
Agriculture $\rightarrow$ **Bt cotton (cryIAc, cryIIAb)**, **RNAi (Meloidogyne)**, golden rice.
Medicine $\rightarrow$ **recombinant insulin (Eli Lilly 1983)**, **ADA gene therapy (1990)**, **PCR**, **ELISA**.
Transgenic $\rightarrow$ **Rosie (1997, 2.4 g/L)**, α-1-antitrypsin, $> 95\,\%$ mice.
Ethics $\rightarrow$ **GEAC**, biopiracy (Basmati, turmeric, neem), Patents 2nd Amendment.

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Quick Reference — Fact / Sequence / Convention Index

Topic / Entity	Key Identifier	Key Numerical / Factual Datum
Bt toxin source	*Bacillus thuringiensis*	Cry crystal forms during sporulation
Cry activation pH	alkaline insect midgut	pH ≥ 9.5 solubilises crystal
cryIAc, cryIIAb	cotton bollworm toxin	engineered into **Bt cotton**
cryIAb	corn borer toxin	engineered into **Bt corn**
RNAi target	*Meloidogyne incognita*	root-knot nematode in tobacco
RNAi machinery	dsRNA $\rightarrow$ Dicer $\rightarrow$ siRNA $\rightarrow$ RISC	21–23 nt siRNA silences mRNA
Vector for RNAi transgene	*Agrobacterium*	delivers sense + antisense DNA
Golden rice	β-carotene biosynthesis	vitamin-A enriched rice
Pomato	somatic hybrid	potato + tomato protoplast fusion
Tissue-culture growth regulators	auxins + cytokinins	for shoot/root regeneration
Virus-free plants	meristem culture	meristems are virus-free
Recombinant therapeutics approved	~ 30 globally / 12 in India	rDNA-derived drugs
Insulin chain A	21 aa	one intrachain S–S
Insulin chain B	30 aa	two interchain S–S to A
Pro-insulin extra peptide	C-peptide	removed during maturation
Insulin commercial host	*E. coli* plasmids	Eli Lilly, 1983
ADA gene therapy first case	4-year-old girl, 1990 USA	retroviral cDNA in lymphocytes
ADA permanent cure route	insert gene at early embryonic stage	germline correction
PCR amplification	$N_n = N_0 \cdot 2^n$	$\sim 10^9$-fold in 30 cycles
PCR enzyme	Taq DNA polymerase	thermostable, from *T. aquaticus*
ELISA principle	antigen–antibody + enzyme	coloured product, Beer–Lambert
ELISA clinical use	HIV, hepatitis, dengue, COVID-19	serology, antigen / antibody
Probe-based diagnosis	radioactive ssDNA / RNA	autoradiography on cell clones
Transgenic animals — share of mice	$> 95\,\%$	disease models, vaccine testing
First transgenic dairy cow	Rosie, 1997	2.4 g human α-lactalbumin / L milk
Emphysema biologic	human α-1-antitrypsin	made in transgenic animals

Topic / Entity	Key Identifier	Key Numerical / Factual Datum
Disease models in mice	cancer, CF, RA, Alzheimer's	rheumatoid arthritis, cystic fibrosis
Indian GMO regulator	**GEAC**	approves research and release
Indian patents act revision	Second Amendment	emergency provisions, R&D
Biopiracy — Basmati	1997 USA patent	landmark case
Rice diversity in India	$\sim 200{,}000$ varieties	27 documented Basmati
Three biotech research pillars	catalyst / conditions / downstream	defines industrial biotech
Five uses of transgenic animals	PDBVC mnemonic	Physiology, Disease, Biologicals, Vaccine, Chemical