A

A (1) Adenine (as a base, or the corresponding nucleoside or nucleotide).
(2) L-Alanine (alternative to Ala).

Å Ångström unit, 10^{-10} m; a unit of length used e.g. to indicate intermolecular distances.

A_{260} See the entry ULTRAVIOLET ABSORBANCE.

A box The adenine riboswitch aptamer (see RIBOSWITCH).

A-DNA One of the conformations adopted by dsDNA: a right-handed helix with ~11 base-pairs per turn.
(Note that aDNA is used to refer to ANCIENT DNA.)
(cf. B-DNA and Z-DNA.)

A family (of DNA polymerases) A group of DNA-DEPENDENT DNA POLYMERASES that include prokaryotic, eukaryotic and viral enzymes. Members of the A family include some phage polymerases (although not those from phages φ29 or T4) and the *Escherichia coli* pol I (involved e.g. in the maturation of Okazaki fragments and in BASE EXCISION REPAIR).
Also included in this family is POLQ (= pol θ; pol theta), an enzyme found in human and other eukaryotic cells. POLQ is able to carry out translesion synthesis of DNA. It may participate in base excision repair, a suggestion supported by the *in vitro* demonstration of its 5′-deoxyribose phosphate lyase activity, a role apparently involved in single-nucleotide base excision repair [Nucleic Acids Res (2009) 37(6):1868–1877].
(See also B FAMILY, X FAMILY and Y FAMILY.)

A site (of a ribosome) The aminoacyl or 'acceptor' site at which tRNA molecules carrying the second and subsequent amino acids bind during translation. (cf. P SITE.)

A-tract In genomic DNA: a nucleotide motif that is reported to be associated with regions of the most pronounced curvature of the molecule; an A-tract is a poly(A) (i.e. poly-adenosine) sequence. In the genome of *Escherichia coli*, A-tracts were reported to be distributed 'quasi-regularly', in both coding and non-coding sequences; the A-tracts occur in clusters ~100 bp long, with consecutive A-tracts exhibiting a periodicity of 10 to 12 bp. It was suggested that the clusters of A-tracts may constitute a form of 'structural code' for DNA compaction in the NUCLEOID [Nucleic Acids Res (2005) 33:3907–3918].
Studies on the mechanics and dynamics of DNA suggested a rationale, incorporating A-tracts, for the stable bending of DNA [Nucleic Acids Res (2008) 3:2268–2283].
Studies on *eukaryotic* genomes have reported that A-tracts are absent specifically in those coding sequences (exons) that correspond to the locations of nucleosomes. It was concluded that the pattern of absence/presence of A-tracts in the genome constitutes a code for the presence/absence – respectively – of nucleosome locations. [The coexistence of the nucleosome positioning code with the GENETIC CODE on eukaryotic genomes: Nucleic Acids Res (2009) doi: 10.1093/nar/gkp689.]
(cf. CLASS A FLEXIBLE PATTERNS.)

AAA ATPases 'ATPases associated with diverse cellular activities' – ATPases which are found in various locations, such as proteasomes and peroxisomes. They have been categorized as AAA+ PROTEINS.

AAA+ proteins A family of NTPases whose members include proteins with diverse functions; AAA ATPASES are examples of this group.
[Review of AAA+ proteins: Genome Biol (2008) 9(4): 216.]

AAAVs Avian adeno-associated viruses (see the entry AAVS).

AAS Aminoalkylsilane (3-aminopropyltriethoxysilane; APES): a reagent used e.g. to bind tissue sections to glass (for *in situ* hybridization etc.).
[Uses (e.g.): Am J Pathol (2006) 169(1):258–267; Nucleic Acids Res (2008) 36(16):5335–5349.]

***aat* gene** In *Escherichia coli*: a gene encoding the enzyme that catalyzes addition of a leucine or phenylalanine residue to the N-terminal of proteins that are synthesized with either an N-terminal arginine or a lysine residue; such addition facilitates degradation of the protein.
(See also N-END RULE.)

AatII A RESTRICTION ENDONUCLEASE from *Acetobacter aceti*. Recognition sequence/cutting site: GACGT↓C.

AAUAAA In a pre-mRNA: a polyadenylation signal upstream of the site at which the molecule is cut and polyadenylated; the polyadenylation sequence is similar in various organisms, although there are variations.
Other *cis*-acting elements may have roles in regulating the polyadenylation of human mRNAs – including upstream U-rich sequences similar to those which have been identified in yeast and plants.
As well as acting as a polyadenylation signal, this sequence was reported to affect the *rate* of transcription [RNA (2006) 12(8):1534–1544].

AAV Adeno-associated virus: see the entry AAVS.

AAV Helper-Free System A commercial gene-delivery system (Stratagene, La Jolla CA) in which the genes in two plasmids provide functions necessary for production of infective AAV virions (see AAVS) without the need for a helper virus; these virions are used to deliver genes to target cells within which viral DNA – containing the gene of interest – integrates in the host cell's DNA.
Essentially, the gene/fragment of interest is first cloned in a plasmid cloning vector in which the insert is bracketed by a pair of inverted terminal repeats (ITRs) which are necessary for subsequent viral packaging. This plasmid is then used to transfect PACKAGING CELLS – which are *co-transfected* with two other plasmids: (i) a plasmid containing the genes that encode viral capsid and replication functions, (ii) a plasmid containing genes that encode the lytic phase of AAV. The resulting infective (*but still replication-deficient*) virions that are produced in the packaging cells can then be used to infect the required target cells (in which the gene of interest can be expressed).
This GENE-DELIVERY SYSTEM has been used e.g. to express

siRNAs [Mitochondrion (2007) 7(4):253–259]; to deliver an anti-angiogenic gene (for investigating age-related macular degeneration) [Mol Vision (2008) 14:471–480]; and to study some features of food/energy metabolism [J Neurosci (2009) 29(1):179–190].

(See also: VIRAPORT RETROVIRAL GENE EXPRESSION SYSTEM and VIRAPOWER LENTIVIRAL EXPRESSION SYSTEM.)

AAVs Adeno-associated viruses (also known as: adeno-satellite viruses): defective viruses that are able to replicate only when certain functions are provided by a co-infecting *helper virus* (adenovirus or herpesvirus) – or, in certain *in vitro* systems, when these functions are provided by plasmid-borne genes (as e.g. in the AAV HELPER-FREE SYSTEM).

Functions provided by adenovirus type 5 (for AAV type 5) include both positive and negative effects. For example, the E4Orf6 function (involved in replication of AAV5 genomic DNA) – together with E1b – degrades AAV5 capsid proteins and Rep52 [J Virol (2007) 81(5):2205–2212]. The functions provided by herpes simplex virus type 1 (for the early stages of AAV replication) were reported to involve nine proteins from the helper virus [PLoS Pathog (2009) 5(3):e1000340].

The AAVs are parvoviruses in which the genome is linear ssDNA. Positive and negative strands of the viral DNA are encapsidated in separate virions.

The AAVs infect a wide range of vertebrates. Initial stages of infection, including internalization of DNA, occur without a helper virus. [Cloning an *avian* AAV (an AAAV) and the generation of recombinant AAAVs: J Virol (2003) 77:6799–6810.]

AAVs are used, for example, in GENE THERAPY. Efforts are being made to increase the efficacy of AAV vectors in gene therapy by designing the CAPSID on the basis of e.g. information obtained from studies on the naturally occurring capsid variants of AAVs in mammals [see: Gene Therapy (2009) 16: 311–319]. (See also KU70.)

An inducible and highly efficient system was reported for the production of recombinant AAV vectors in insect (Sf9) cells [Proc Natl Acad Sci USA (2009) 106(13):5059–5064].

AAV vectors, encoding genes of the α and the β subunits of hexosaminidase, were inoculated, *intracranially*, into mice in order to assess the potential of gene therapy for treatment of the human GM2 gangliosidoses such as Tay–Sachs disease and Sandhoff disease [Proc Natl Acad Sci USA (2006) 103 (27):10373–10378]. A simpler method for delivering genes to brain cells was reported later (see below).

AAV9 has been used, in mice, for gene delivery to cells of the central nervous system (brain and spinal cord) *by intravenous injection*. It was thought that this approach may allow the development of gene therapy for e.g. some human neurodegenerative diseases [Nature Biotechnol (2008) 27:59–65].

AAV vectors were also used for the genetic manipulation of cultured neurons [Brain Res (2008) 1190:15–22].

It was reported earlier that, in human cells, AAV DNA (in the absence of helper virus) integrates in the genome with an apparent preference for CPG ISLANDS. More recently, AAVs have been reported to integrate, site-specifically, into a locus on chromosome 19, and the occurrence of such integration is apparently influenced by the TRP-185 protein [J Virol (2007) 81(4):1990–2001]. Palindromes of length greater than about 40 bp are reported to be significant targets for the integration of recombinant AAV vectors [J Virol (2007) 81(20):11290–11303].

The site of insertion of AAVs within chromosome 19 was reported to contain a 347-bp sequence capable of enhancing the promoter and transcriptional functions of AAV vectors in liver cells; inclusion of this small fragment in AAV vectors may thus facilitate their use for the delivery and expression of transgenes in liver cells [Gene Therapy (2009) 16:43–51].

AB1380 A strain of the yeast *Saccharomyces cerevisiae* (see the entry SACCHAROMYCES for some details).

(See also YEAST ARTIFICIAL CHROMOSOME.)

abacavir A NUCLEOSIDE REVERSE TRANSCRIPTASE INHIBITOR used e.g in antiretroviral therapy; CSF–plasma ratios indicate that it may reach therapeutic levels in the cerebrospinal fluid (CSF).

A trial that compared abacavir with nevirapine (as part of a combined therapy) reported that abacavir tended to produce a lower rate of serious adverse effects, suggesting a wider use of this drug in resource-limited settings [Trop Med Int Health (2008) 13(1):6–16].

abasic site *Syn*. AP SITE.

abasic-site mimic See the entry RPA.

ABC excinuclease See UVRABC-MEDIATED REPAIR.

Abelson murine leukemia virus See the entry ABL.

aberrant RNA (aRNA) See the entry ARNA (sense 2).

abl (*ABL*) An ONCOGENE first identified in the Abelson murine leukemia virus. The v-*abl* product has TYROSINE KINASE activity. The human homolog of v-*abl*, c-*abl*, is usually present on chromosome 9; however, in the majority of patients with CHRONIC MYELOGENOUS LEUKEMIA it has been translocated to chromosome 22, forming a chimeric gene, known as *bcr-abl*, that encodes a tumor-specific tyrosine kinase (designated P210). Chromosome 22 containing the chimeric *bcr-abl* gene is called the Philadelphia chromosome (also called Ph^1).

Subcellular localization of c-Abl protein at an early stage in myogenic differentiation was reported to be influenced by its acetylation [EMBO Rep (2006) 7(7):727–733].

abortive transduction TRANSDUCTION in which the transduced DNA persists in a recipient cell as a stable, extrachromosomal but non-replicating molecule; when the recipient divides only one daughter cell receives the DNA fragment.

absorbance (ultraviolet) See ULTRAVIOLET ABSORBANCE.

abzyme *Syn*. CATALYTIC ANTIBODY.

Abzyme® A reagent kit (Abbott Laboratories) used for detecting antibodies in the context of hepatitis B.

acceptor splice site (acceptor splice junction) In a pre-mRNA: the splice site (consensus AG) at the 3′ end of an intron.

(cf. DONOR SPLICE SITE.)

accession number A number which refers to a unique database entry for a given sequence or gene. Some examples include:

(i) GenBank® accession number X17012, referring to data on the gene for rat insulin-like growth factor II (IGF II); (ii) GenBank® accession number AY024353, referring to data on the *ftsZ* gene of the bacterium *Sodalis glossinidius;* (iii) GenBank® accession number AM160602, referring to data on mRNA of the gene for cinnamyl alcohol dehydrogenase in a species of oak (*Quercus ilex*).

(See also ANNOTATION.)

AccuPrime™ GC-rich DNA polymerase A DNA polymerase (Invitrogen, Carlsbad CA) optimized for DNA synthesis on 'difficult-to-amplify' templates, including those with a GC content >65%. Targets up to 5 kb may be amplified with this polymerase.

[Uses (e.g.): J Bacteriol (2008) 190(24):8096–8105; J Exp Clin Cancer Res (2008) 27:54; FEMS Microbiol Lett (2009) 294(1)32–36.]

AccuProbe® A family of PROBES (Gen-Probe, San Diego CA) used for identifying certain medically important bacteria by detecting specific sequences of nucleotides from lysed cells. The method involves a *hybridization protection assay*. In this assay, an added reagent cleaves the acridinium ester label on all *unbound* probes. Labels on the *bound* probes (which are protected from cleavage by virtue of their position in the probe–target duplex) react with a second reagent, producing a chemiluminescent (light) signal. The light produced by this reaction is measured in RLUs (i.e. relative light units). The threshold value (in RLUs) for a positive result must be carefully examined [see for example: J Clin Microbiol (2005) 43:3474–3478].

[Use for *Staphylococcus aureus*: J Clin Microbiol (2008) 46(6):1989–1995. Use for *Streptococcus pneumoniae* (as a reference): J Clin Microbiol (2008) 46(7):2184–2188. Use for *Mycobacterium avium*: J Clin Microbiol (2008) 46 (8):2790–2793. Use for identifying *Mycobacterium* spp: Emerg Infect Dis (2009) 15(1):53–55, and Emerg Infect Dis (2009) 15(2):242–249.]

(See also PACE 2C and TMA.)

acetosyringone A phenolic substance which promotes activity of the *vir* operon in species of the plant-pathogenic bacterium *Agrobacterium* (see CROWN GALL).

(See also AGROINFILTRATION.)

Acetosyringone has been used e.g. for studies on terpenoid metabolism in the tomato plant [Plant Physiol (2009) 149(1): 499–514], and studies on the transformation of wheat [Plant Cell Rep (2009) 28(6):903–913].

Agrobacterium can also transfer T-DNA to other types of cell, including e.g. human and fungal cells; acetosyringone was used to promote transfer of T-DNA from *Agrobacterium* to the fungus *Aspergillus fumigatus* for (random) insertional mutagenesis [PLoS ONE (2009) 4(1):e4224].

***N*-acetyl-L-cysteine** See MUCOLYTIC AGENT.

acetylation (of histones) HISTONE acetylation is regulated e.g. by the opposing effects of histone acetyltransferases (HATs) and histone deacetylases (see HDAC); the (de)acetylation of histones can affect CHROMATIN structure, and may therefore alter the accessibility of DNA for events such as transcription and repair.

The acetylation of histones can be studied/manipulated e.g. by using HDAC *inhibitors* (e.g. TRICHOSTATIN A).

[Genome-wide analysis of histone acetylation and its effect on gene expression in (the protozoan) *Entamoeba histolytica*: BMC Genomics (2007) 8:216.]

A general perception is that transcription of genes requires – as a pre-condition – an 'open' form of CHROMATIN (the so-called *euchromatin*) in the vicinity of the given genes; acetylation of vicinal histone(s) is usually regarded as an important factor associated with the presence of euchromatin. (In some types of chemotherapy – EPIGENETIC THERAPY – an inhibitor of HDACs is sometimes included in order to promote 'open' chromatin in the vicinity of specific gene(s) with the object of contributing to de-repression of the genes.) However, histone acetylation is only one factor that regulates gene expression; for example, it was reported that the drug-induced formation of 'open' chromatin (involving hyper-acetylation of vicinal histone(s)) was not, on its own, sufficient to de-repress lytic-cycle genes in the Epstein–Barr virus [J Virol (2008) 82(10): 4706–4719]. Nevertheless, the complexity of this issue may be indicated by a study in which histone acetylation – but not DNA demethylation – *was* found to be sufficient to break the latency of gamma-herpesvirus 68 in a mouse cell line [PLoS ONE (2009) 4 (2):e4556].

In a genomewide study of HDACs in *Schizosaccharomyces pombe* (a fission yeast), the patterns of histone acetylation, HDAC binding and nucleosome density were compared with gene expression profiles; it was found that different HDACs may have different roles in repression and activation of genes [EMBO J (2005) 24(16):2906–2918]. Following *damage* to DNA in *S. pombe*, the restoration of chromatin structure was reported to involve deacetylation of histone H3 by Hst4 (a putative HDAC) [Eukaryotic Cell (2008) 7:800–813], while recovery from DNA damage was reported to involve Mst1 (a histone acetyltransferase) [Genetics (2008) 179(2):757–771].

In (human) nucleosomes, the acetylation of certain lysine residues depends primarily on HATs, but the effect of these enzymes appears to be promoted by binding protein HMGN1 [EMBO J (2005) 24(17):3038–3048].

Acetylation of the histone *chaperone* NUCLEOPHOSMIN, as well as histone acetylation, apparently promotes transcription [Mol Cell Biol (2005) 25(17):7534–7545], while chaperone-stimulated, histone-acetylation-independent transcription has also been reported [Nucleic Acids Res (2007) 35:705–715].

HDACs in (human) development and physiology have been reviewed within the context of implications for disease and therapy [Nature Rev Genetics (2009) 10:32–42].

The c-Abl protein (see ABL) was reported to be a substrate for the p300 and other histone acetyltransferases.

***N*-acetylmuramidase** See LYSOZYME.

***N*-acetylneuraminic acid** (NANA) See NEURAMINIDASE.

ACF APOBEC-1 complementation factor: see RNA EDITING.

Achilles' heel technique A technique in which a RESTRICTION ENDONUCLEASE is targeted to one *particular* recognition site when multiple copies of that site are freely available. In one method, a triplex-forming oligonucleotide (see TRIPLEX DNA) is used to mask the required cleavage site. While this site is masked, the remaining sites are methylated in order to inhibit subsequent cleavage; the triplex is then removed and specific cleavage can be carried out.

(See also PROGRAMABLE ENDONUCLEASE.)

aciclovir Alternative spelling for ACYCLOVIR.

acid-fast bacilli Those bacilli (i.e. rod-shaped bacteria) which, when stained with the Ziehl–Neelsen (or similar) stain, resist decolorization with mineral acid or an acid–alcohol mixture. This kind of staining method is used for screening respiratory specimens, e.g. samples of sputum, and for examining other types of specimen, for *Mycobacterium tuberculosis* (an acid-fast species).

Ac*M*NPV *Autographa californica* NPV: see NUCLEAR POLYHEDROSIS VIRUSES.

AcNPV *Syn*. Ac*M*NPV – see entry NUCLEAR POLYHEDROSIS VIRUSES.

acridines Heterocyclic, fluorescent compounds which bind to dsDNA (primarily as an INTERCALATING AGENT) and also to single-stranded nucleic acids (and to the backbone chains of double-stranded nucleic acids). Acridines have antimicrobial activity and they are mutagenic; they are also used as stains for nucleic acids and can be used for CURING plasmids.

acridinium ester label (on probes) See ACCUPROBE.

acrocentric Refers to a CHROMOSOME in which the CENTROMERE is located close to one end.

acrydite hybridization assay An assay in which molecules of labeled ssDNA or ssRNA, passing through a polyacrylamide gel by electrophoresis, are captured (bound) by complementary oligonucleotides immobilized in a (central) 'capture zone' within the gel; all the molecules of nucleic acid that are *not* complementary to the capture oligos pass through the central capture zone and continue their migration to the end of the gel strip. The complementary oligos are synthesized with a 5′ terminal acrydite group which binds them to the polyacrylamide matrix so that they are immobilized in the gel. (Note that the central region of the gel strip is prepared separately.)

acrylamide A toxic, water-soluble agent (CH_2=CH—$CONH_2$) which can be polymerized to POLYACRYLAMIDE by catalysts such as *N*,*N*′-methylene-*bis*-acrylamide ('Bis') which promote cross-linking.

actinomycin C_1 *Syn*. ACTINOMYCIN D.

actinomycin D An antibiotic (a substituted phenoxazone linked to two pentapeptide lactone rings) produced by some species of *Streptomyces;* it acts as an INTERCALATING AGENT, binding to DNA and inhibiting DNA-dependent RNA polymerase. The drug has low affinity for AT-rich promoter regions; hence, *initiation* of transcription from such promoters may be little affected by the antibiotic.

activation domain (AD) See YEAST TWO-HYBRID SYSTEM.

activation-induced cytidine deaminase (AID) An enzyme that occurs in germinal center B lymphocytes (B cells) and which is an absolute requirement for affinity maturation and class switching in normal development of antibodies.

The (autosomal recessive) form of HYPER-IGM SYNDROME has been linked to a deficiency of AID (see the table in entry GENETIC DISEASE).

AID also inhibits retrotransposition of L1 – suggesting that it has function(s) in addition to its role of creating antibody diversity [Nucleic Acids Res (2009) 37(6):1854–1867].

(See also CYTIDINE DEAMINASE AND RNA EDITING.)

activation/regulation of genes (*DNA technol*.) See e.g. entry CONDITIONAL GENE ACTIVATION/REGULATION.

activity-based probe A type of probe used for the real-time study of APOPTOSIS (q.v.).

acyclonucleotide Any analog of a deoxyribonucleotide or ribonucleotide in which a non-cyclic moiety carries the base. One example is a monomer of glycerol nucleic acid (see the entry GNA). Polymerization of certain acyclonucleotides on a DNA template has been achieved with THERMINATOR DNA POLYMERASE.

acyclovir (alternative spelling: aciclovir) 9-(2-hydroxyethoxymethyl)guanine: an antiviral agent which is active against a number of herpesviruses, including herpes simplex. In cells, acyclovir is phosphorylated to the monophosphate by (viral) thymidine kinase; subsequently it is converted to the (active) triphosphate form via host-encoded enzymes. The active drug inhibits *viral* DNA polymerase; the host cell's polymerase is much less sensitive.

In cells which are not virally infected, acyclovir appears not to be significantly phosphorylated.

Acyclovir has been used topically and systemically.

***N*-acyl-homocysteine thiolactone** See QUORUM SENSING.

***N*-acyl-L-homoserine lactone** (AHL) See QUORUM SENSING.

acylneuraminyl hydrolase *Syn*. NEURAMINIDASE.

AD primer (arbitrary degenerate primer) See TAIL-PCR.

Ada protein (in *Escherichia coli*) See DNA REPAIR.

adaptamer See ORFMER SETS.

adaptive response (to alkylating agents) See DNA REPAIR.

adaptor A short, synthetic, double-stranded fragment of DNA which is similar, in principle, to a LINKER but which generally offers more flexibility. Thus, for example, the two ends of a given adaptor may consist of dissimilar STICKY ENDS – one end able to bind to a (complementary) sticky end on a DNA fragment and the other able to bind to a different sticky end on a vector molecule, facilitating the integration of fragments and vectors which were cut by different restriction enzymes. An adaptor may also include one or more internal restriction sites, offering the chance to select alternative sticky ends at a later stage in the work, and there may also be primer-binding sites and/or a promoter sequence etc.

(See also NOTI.)

ADAR1 A dsRNA ADENOSINE DEAMINASE which is involved e.g. in RNA EDITING.

(See also Zα in the entry Z-DNA.)

AdEasy™ XL adenoviral vector system A GENE-DELIVERY

SYSTEM (marketed by Stratagene, La Jolla CA, USA) which uses ADENOVIRUS-based vectors. Essentially, it facilitates the preparation of a recombinant adenoviral vector containing the gene/fragment of interest.

Initially, the gene/insert is cloned in a small shuttle vector (~7 kb) which includes: (i) the left and right ITRs (inverted terminal repeats) of the adenovirus genome; (ii) two regions homologous to two sequences in another plasmid, pAdEasy-1 (see later), (iii) a gene encoding resistance to kanamycin; and (iv) a recognition site for the restriction endonuclease PmeI.

After cloning, the shuttle vector is linearized (by cleavage with PmeI); linearization leaves the two homologous regions (see above) in terminal positions.

The linearized shuttle vector is inserted, by transformation, into a strain of *Escherichia coli*, BJ5183-AD-1, that already contains the (circular) plasmid vector pAdEasy-1. pAdEasy-1 (~33 kb) includes modified genomic DNA of human adenovirus serotype 5 (containing deletions in both the E1 and E3 regions). Homologous recombination occurs (within *E. coli*) between the linearized shuttle vector and homologous regions in pAdEasy-1. Cells containing the recombinant plasmids are selected on kanamycin-containing media.

The recombinant plasmids are isolated, and the next stage is conducted *in vitro*.

Recombinant plasmids are cleaved by a restriction enzyme, PacI, at selected sites, yielding a linear construct with adenoviral terminal sequences (i.e. ITRs). This construct is used to transfect specialized, competent AD-293 cells – within which infective adenovirus virions (containing the gene/fragment of interest) are produced; the virions are released for subsequent use in gene-expression studies in mammalian cells.

The principle of the AdEasy™ system was exploited in the production of oncolytic adenovirus [BMC Biotechnol (2006) 6:36].

The AdEasy™ XL vector system has been used in a range of studies [see e.g. Nucleic Acids Res (2006) 34(12):3577–3584; Vaccine (2007) 25(52):8687–8701; Mol Ther (2008) 16(5):886–892; J Clin Invest (2009) 119:976–985; Cell Cycle (2009) 8(2):257–267; Biochim Biophys Acta (2009) 1793(8): 1379–1386; J Exp Clin Cancer Res (2009) 28(1):75; BMC Immunol (2009) 10:14].

adefovir A NUCLEOSIDE REVERSE TRANSCRIPTASE INHIBITOR.

adenine phosphoribosyltransferase An enzyme (EC 2.4.2.7) which forms adenosine monophosphate (AMP) from adenine and 5-phosphoribosyl-1-diphosphate.

In humans, a deficiency of adenine phosphoribosyltransferase (an autosomal recessive disorder) can cause excretion of adenine (in the urine) and the formation of a highly insoluble product, 2,8-dihydroxyadenine, which can give rise to kidney stones and renal failure.

adeno-associated viruses See AAVS.

adeno-satellite viruses See AAVS.

adenosine A RIBONUCLEOSIDE.

adenosine deaminase An enzyme (EC 3.5.4.4) which catalyzes the conversion of adenosine to inosine.

(See also ADAR1.)

adenosine deaminase deficiency The congenital deficiency of a functional adenosine deaminase (EC 3.5.4.4) characterized by lack of normal development of T cells and a (consequent) marked immunodeficiency in which the patient is susceptible to infection by opportunist pathogens. This disorder has been treated successfully by GENE THERAPY.

(See also GENETIC DISEASE (table).)

***S*-adenosyl-L-methionine** A methyl group (CH_3–) donor that is used in various reactions.

Adenovirus A genus of icosahedral, non-enveloped viruses of the family Adenoviridae; the genome is linear dsDNA. These viruses infect mammals and birds; each type of adenovirus is usually specific for one, or a limited range, of closely related host species.

Adenovirus pathogenesis (in humans) commonly involves respiratory-tract infections, while some adenoviruses are able to induce tumors in rodents (rats). The *oncogenic* potential of adenoviruses has been investigated by using Ad5 adenovirus *dl520* to infect human U2OS cells, and monitoring levels of the Myc protein (see MYC); it was reported that the (virus-encoded) E1A protein interacts with the cell's p400, resulting in the stabilization of Myc and induction of Myc target genes [Proc Natl Acad Sci USA (2008) 105(16):6103–6108)]. (See also HEPATOCELLULAR CARCINOMA SUPPRESSOR 1.)

The adenovirus virion is ~70–90 nm in diameter; the capsid encloses a core containing genomic DNA (which is closely associated with an arginine-rich polypeptide). The 5′ end of each strand of the DNA is covalently linked to a hydrophobic 'terminal protein' (TP).

Both ends of the genomic DNA are characterized by an inverted terminal repeat (ITR) – which varies in length in the different types of adenovirus; the 5′ end residue is commonly dCMP.

During infection, the core enters the nucleus, releasing viral DNA. Replication of viral DNA involves TP and also a virus-encoded DNA polymerase, as well as other virus- and host-encoded proteins. TP, synthesized in precursor form, binds covalently to DNA during replication and is later cleaved to the mature (DNA-bound) TP. A TP-mediated form of DNA replication also occurs in PHAGE φ29 (q.v. for details).

Expression of late viral genes, encoding structural proteins, is accompanied by the cessation of cellular protein synthesis.

Some 10^5 virions may be formed within a single host cell.

Adenoviruses as vectors

Adenoviruses are employed as VECTORS in various types of investigation, including GENE THERAPY. (See also the entry ADEASY XL ADENOVIRAL VECTOR SYSTEM.) Re-targeting of adenovirus type 5 vectors to cell-surface $\alpha_v\beta 6$ integrin molecules resulted in reduced hepatotoxicity and improved uptake by tumor cells following systemic delivery [J Virol (2009) 83 (13):6416–6428].

Titers of recombinant adenoviruses in packaging cells may be optimized in various ways: see e.g. PACKAGING CELL.

Variable genome size in replication-deficient vectors may

affect viral stability [J Virol (2009) 83(4):2025–2028].

[Vaccine vectors: Mol Therapy (2009) 178:1333–1339.]

Adenovirus dodecahedron base

A construct, which consists of twelve copies of a pentameric adenoviral capsid protein, involved in the penetration of cells, has been used as a vehicle for increasing the uptake, by cells, of the antitumor agent bleomycin (which was tethered to the construct) [PLoS ONE (2009) 4(5):e5569].

adenoviruses Viruses of (i) the genus ADENOVIRUS – *or* (ii) the family Adenoviridae.

adenylate cyclase An enzyme (EC 4.6.1.1) which catalyzes the conversion of ATP to CYCLIC AMP.

In *Escherichia coli*, the activity of adenylate cyclase (the *cya* gene product) is regulated e.g. in association with the CATABOLITE REPRESSION system.

(See also BACTERIAL TWO-HYBRID SYSTEM.)

In mammals, the enzyme forms part of a plasma membrane complex and is regulated e.g. via certain G proteins; it is activated by some bacterial exotoxins (e.g. PERTUSSIS TOXIN).

Anthrax toxin (EF component) and *cyclolysin* (a virulence factor synthesized by the Gram-negative bacterial pathogen *Bordetella pertussis*) both exhibit adenylate cyclase activity, which is stimulated by CALMODULIN.

adenylate kinase An enzyme (EC 2.7.4.3) which catalyzes the (reversible) conversion: 2ADP⟷ATP+AMP.

aDNA *Ancient* DNA – see the entry ANCIENT DNA.

ADO ALLELE DROP-OUT.

AdoMet Abbreviation for *S*-adenosyl-L-methionine.

ADP-ribosylation The transfer, to a protein, of an ADP-ribosyl group from NAD^+, mediated by ADP-ribosyltransferase (EC 2.4.2.30).

In eukaryotes ADP-ribosylation can (for example) regulate the properties of HISTONES.

In *Escherichia coli*, RNA polymerase is ADP-ribosylated (with change in activity) following infection with bacteriophage T4.

ADP-ribosylation is an intracellular effect of some bacterial exotoxins (e.g. cholera toxin and PERTUSSIS TOXIN).

Polymerized ADP-ribosyl subunits (up to 50) may be found on certain eukaryotic proteins.

adult stem cell See STEM CELL.

Aedes aegypti A species of mosquito which can act as a vector in the transmission of e.g. dengue and yellow fever [genome: Science (2007) 316(5832):1718–1723].

(See also ARBOVIRUSES.)

It was reported that NUMTS are prevalent in the genome of *A. aegypti*, suggesting that future phylogenetic or population genetic studies should be carried out with nuclear, rather than mitochondrial, DNA markers [BMC Genet (2009) 10:11].

affinity capture electrophoresis Electrophoresis in a medium containing immobilized capture probes; it is used e.g. for the isolation of a given fragment of ssDNA, or a fragment of triplex-forming dsDNA.

(See also ACRYDITE HYBRIDIZATION ASSAY.)

affinity chromatography Chromatography in which particular molecules are isolated (adsorbed) owing to their affinity for an immobilized ligand – any non-specific unbound molecules being removed from the immobilized matrix. This procedure may be used e.g. for isolating/purifing a given type of molecule (see e.g. GENE FUSION (uses)).

Affinity® protein expression and purification A product of Stratagene (La Jolla CA) designed to facilitate the expression and purification of proteins expressed in prokaryotic systems; the product includes various pCAL plasmid vectors, shown in the diagram – each encoding a CALMODULIN-binding affinity tag.

Protein expression is maximized by a vector that includes a T7/LacO promoter system. In suitable strains of *Escherichia coli* (e.g. BL21(DE3)), T7 RNA polymerase is expressed in the presence of the inducer IPTG and drives expression from the T7/LacO promoter system on the plasmid. Tight control of expression is achieved with a plasmid-borne copy of $lacI^q$. Efficient translation of the protein of interest is promoted by using the strong ribosome-binding site (RBS) of T7 gene 10.

pCAL vectors contain a ColE1 origin of replication and an ampicillin-resistance gene.

All pCAL vectors encode a CALMODULIN-binding peptide (CBP) tag which forms a fusion product with the expressed protein and permits high-level purification following a single passage through CALMODULIN-AFFINITY RESIN. The (small) size of the CBP tag (about 4 kDa) may be expected to have a smaller effect on the protein of interest compared with larger tags – such as the (26-kDa) glutathione *S*-transferase (GST) affinity tag.

One of the pCAL vectors includes a KEMPTIDE SEQUENCE which can be used e.g. for *in vitro* labeling of the expressed protein with protein kinase A (PKA) and ^{32}P.

All of the pCAL vectors include a cleavage site (for enterokinase or thrombin proteinase) for removal of the CBT tag.

One of the pCAL vectors includes a FLAG sequence.

Affinity® vectors were used e.g. in studies on a fluorescent reporter protein [Proc Natl Acad Sci USA (2008) 105:20038–20043]; a light-activated DNA-binding protein [Proc Natl Acad Sci USA (2008) 105(31):10709–10714; and HMG box proteins [Mol Endocrinol (2008) 22(5):1141–1153].

affinity resin See e.g. NICKEL-CHARGED AFFINITY RESIN.

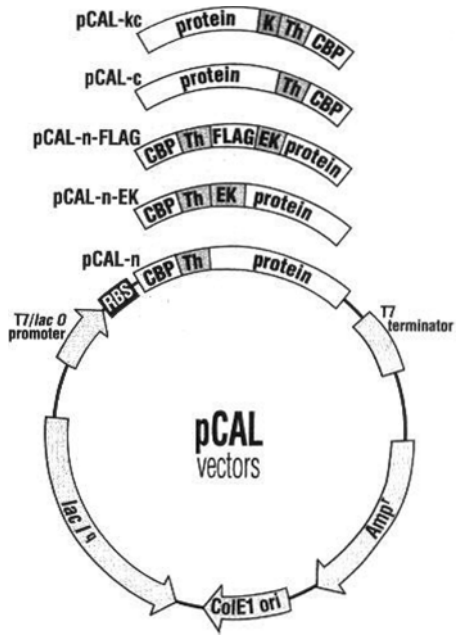
affinity tag *Syn*. AFFINITY TAIL.

affinity tail (affinity tag) Part of a FUSION PRODUCT which can facilitate detection/isolation of a (recombinant) protein e.g. by AFFINITY CHROMATOGRAPHY or by the use of an affinity resin (such as a NICKEL-CHARGED AFFINITY RESIN).

Some affinity tails are small peptides. One advantage of a small affinity tail is that it is less likely to interfere with the function of the fusion protein – so that its removal may not be necessary.

(See also FLAG, PESC VECTORS and SIX-HISTIDINE TAG.)

Large (protein) tails, for example glutathione *S*-transferase, may improve the solubility of the fusion protein but they may need subsequent removal in order to avoid interference with the function of the recombinant target protein.



Affinity® PROTEIN EXPRESSION AND PURIFICATION The range of pCAL vectors (see entry for details of the method).CBP (in each vector) refers to calmodulin-binding peptide. EK = enterokinase; K = Kemptide sequence; Th = thrombin proteinase. (See also entry for FLAG.)

Courtesy of Stratagene, La Jolla CA, USA.

(See also CHAMPION PET SUMO VECTOR.)

A highly temperature-stable affinity tail (a lectin, stable up to 80°C) may be useful for proteins originating from thermophilic organisms; the fusion proteins bind specifically to an agarose matrix that contains D-mannose, and the affinity tail can by cleaved by an enterokinase [BioTechniques (2006) 41 (3):327–332].

[Affinity as a tool in life science (a review): BioTechniques (2008) 44(5):649–654.]

(See also AFFINITY PROTEIN EXPRESSION AND PURIFICATION.)

aflatoxins Heat-stable toxins produced by certain fungi (strains of *Aspergillus flavus* and *A. parasiticus*); the molecule of an aflatoxin contains a bifuran moiety fused with a (substituted) coumarin.

[Genes for aflatoxin biosynthesis: Appl Environ Microbiol (2005) 71:3192–3198.]

Aflatoxins damage DNA (producing e.g. mutations, strand breakage and inhibition of repair) and they are also highly carcinogenic in mammalian species; the aflatoxins are associated e.g. with cases of hepatocellular carcinoma. Different species may be affected in different ways by these toxins.

In mouse hepatocytes, it was found that α-mannan protects against DNA damage from aflatoxin B_1 [Int J Mol Sci (2009) 10(2):395–406].

AFLP Either of *two distinct* PCR-based approaches for TYPING bacteria.

One approach ('amplified fragment length polymorphism') includes a number of variant forms of arbitrarily primed PCR (AP-PCR), including e.g. RAPD analysis.

The other approach, outlined here, involves initial digestion of genomic DNA with two types of RESTRICTION ENDONUCLEASE; it is sometimes called 'amplified restriction fragment length polymorphism' – but this is not recommended by the original authors [see comments in: Nucleic Acids Res (1995) 23:4407–4414].

In the digested genome each fragment is flanked by STICKY ENDS produced by one or other of the two types of restriction enzyme. Two types of adaptor molecule (A, B) are added; the A molecules have *one* sticky end which binds to sites cleaved by one of the restriction enzymes, and B molecules have *one* sticky end that binds to sites cleaved by the other enzyme. A site cleaved by EcoRI (left) and a matching adaptor (right) is shown below:

```
5′-----NNG          AATTGNNNNN-3′
  -----NNCTTAA            CNNNNN
```

Fragment–adaptor binding is followed by ligation – but the cleavage site of EcoRI is *not* regenerated, avoiding repeated restriction. Fragments (with their adaptors) are amplified by PCR. Each primer is complementary to a sequence covering part of an adaptor and the (contiguous) restriction site of the fragment. Each primer's 3′ end extends beyond the restriction site for one (or several) nucleotides; thus, a given primer will be extended only if the primer's 3′ terminal 'selective' nucleotide(s) align with *complementary* base(s) in the fragment. In the example given above, one template strand is:

```
5′-----NNGAATTGNNNNN-3′
```

and a primer with a deoxycytidine (C) selective 3′ nucleotide will bind as follows:

```
5′-----NNGAATTGNNNNN-3′
       CCTTAACNNNNN-5′   ← primer
```

The same principle holds for each primer-binding site: only some of the primers will be extended owing to their selective 3′ end.

The PCR products undergo gel electrophoresis, and bands of products (made visible e.g. by the use of labeled primers) form the *fingerprint*.

AFLP discriminated easily between *Clostridium botulinum* types A, B, E and F, but only limited intraspecies differences

were reported in isolates of *Brucella*.

AFM ATOMIC FORCE MICROSCOPY.

agar A complex mixture of galactans obtained from certain red algae (e.g. *Gelidium*); these compounds form part of the cell wall and/or intercellular matrix.

Agar comprises two main components: AGAROSE and agaropectin. Agaropectin is a mixture of sulfated galactans, some of which contain glucuronic acid and/or other constituents.

An agar *gel* is used as a matrix in many types of solid and semi-solid microbiological medium. This translucent, jelly-like material is prepared by heating a mixture of agar – e.g. 1.5% w/v – and water to >100°C and then cooling it to room temperature; gelling begins at ~40–45°C.

Agar can be an inhibitory factor in PCR (cf. GELLAN GUM).

One report indicated that agar promotes transformation in *Escherichia coli* [J Bacteriol (2009) 191(3):713–719].

agarose The major constituent of AGAR: a non-sulfated linear polymer consisting of alternating residues of D-galactose and 3,6-anhydro-L-galactose. Agarose is used e.g. as a medium in GEL ELECTROPHORESIS for the separation of large fragments of nucleic acid.

(cf. POLYACRYLAMIDE.)

age-related macular degeneration See the entry AMD.

agnoprotein A regulatory protein, encoded by JC VIRUS, which plays a major role in the infective cycle; it may be involved in facilitating the transport of virions from the nucleus (where virus assembly occurs) to the cytoplasm.

Mutant, phosphorylated forms of agnoprotein are unable to sustain the infective cycle [J Virol (2006) 80(8):3893–3903].

Expression of a transfected agnoprotein-encoding gene was reported to give rise to inhibition of the low-fidelity pathway of DSB (double-strand break) repair, involving protein KU70.

Ago protein See ARGONAUTE.

Agrobacterium A genus of motile, Gram-negative bacteria that are found primarily in the rhizosphere (root environment of plants).

GC% of genomic DNA: 57–63.

Optimal temperature for growth: 25–28°C. Various mono- and disaccharides can be metabolized; glucose is metabolized e.g. via the Entner–Doudoroff pathway and the hexose monophosphate pathway.

Colonies which develop on media containing carbohydrates are generally mucilaginous, copious slime being formed.

Most species are pathogenic to plants – *A. tumefaciens* and *A. vitis* are causal agents of CROWN GALL.

(Some relevant entries: AGROINFECTION, BINARY VECTOR SYSTEM, FLORAL DIP METHOD AND TRIFOLITOXIN.)

Agrobacterium was used for random insertional mutagenesis in the fungus *Aspergillus fumigatus* (by the insertion of T-DNA: see CROWN GALL) [PLoS ONE (2009) 4(1):e4224].

Taxonomic note

One taxonomic study has classified all species of *Agrobacterium* in the genus *Rhizobium* [Int J Syst Evol Microbiol (2001) 51: 89–103]. According to this paper, *A. tumefaciens* is renamed *Rhizobium radiobacter*, and *A. vitis* is renamed as *Rhizobium vitis*. (The name *Agrobacterium tumefaciens* has been – and continues to be – widely used in publications.)

agroinfection A method for introducing viral DNA (or cDNA) into plant cells.

In the original procedure, viral DNA is inserted into the T-DNA region of the Ti plasmid of bacterium *Agrobacterium tumefaciens* (see the entry CROWN GALL for details of the Ti plasmid); thus, when infecting a plant, *A. tumefaciens* injects viral DNA (within the T-DNA) into plant cells.

Currently, the commonly used procedure involves the use of a *binary vector* system. In this approach the gene/sequence of interest is inserted into a small (binary) vector in which it is flanked, on each side, by the left border and right border of T-DNA. The binary vector, containing the gene, is inserted into an engineered strain of *Agrobacterium tumefaciens* that contains the *vir* (virulence) region of the Ti plasmid, which is concerned with transfer of DNA into plant cells. This strain is then used to infect a plant. When the *vir* genes are activated, DNA from the *binary vector* (specifically, the DNA from the section bracketed by the left and right borders of T-DNA) is transferred into the plant cells – such transfer being mediated by factors encoded by the *vir* region acting in *trans*.

For developing transgenic plants it is desirable to minimize the content of extraneous DNA which is transferred to plant cells through the vector system, particularly when such plants are to be made available in a general agricultural setting; the content of extraneous DNA is covered by certain laws which relate to transgenic plants. To this end, a number of minimal T-DNA vectors have been suggested [BioTechniques (2006) 41(6):708–710].

Agroinfection was used e.g. to study resistance to disease in the potato and the avirulence genes in *Phytophthora infestans* (the late blight pathogen) [PLoS ONE (2008) 3(8):e2875].

[High-efficiency expression of proteins in plants from agroinfection-compatible TMV (tobacco mosaic virus) expression vectors: BMC Biotechnol (2007) 7:52.]

(See also FLORAL DIP METHOD; cf. AGROINFILTRATION.)

agroinfiltration A procedure, used for inserting a vector into plant cells, in which cells of the plant-pathogenic bacterium *Agrobacterium tumefaciens* (containing the vector, and in the presence of ACETOSYRINGONE) are injected, with a syringe, through leaf stomata.

[Uses (e.g.): Plant Physiol (2007) 145(1):5–16 and (2009) 149(2):1005–1016.]

(cf. AGROINFECTION.)

AGT O^6-alkylguanine-DNA alkyltransferase: see *Uses of gene fusion* in the entry GENE FUSION.

AHL (*N*-acyl-L-homoserine lactone) See QUORUM SENSING.

***ahpC* gene** See ISONIAZID.

AHT (*N*-acyl-homocysteine thiolactone) See QUORUM SENSING.

AID ACTIVATION-INDUCED CYTIDINE DEAMINASE.

AIDS Acquired immune deficiency syndrome. An HIV^+ person

with AIDS has counts of $CD4^+$ T cells below a certain level and, additionally, the presence of one or more types of AIDS-defining disease (such as: candidiasis of the *lower* respiratory tract, retinitis with CMV (cytomegalovirus), *extra*pulmonary infection with *Mycobacterium tuberculosis*, pneumonia due to *Pneumocystis carinii* etc.).

The causal agent is generally HIV-1 (q.v.); the disease can also be caused by HIV-2. (See RETROVIRUSES for a general background to HIV-1 and other, related viruses.)

Chemotherapy used in the treatment of AIDS may include various ANTIRETROVIRAL AGENTS (q.v.). (See also HAART.)

Quiescent HIV-1

The chemotherapeutic agents currently in use aim primarily to break the replicative cycle of the virus (e.g. by inhibiting maturation of the virions). However, this does not address the covert problem of viral *persistence* resulting from the 'silent' *integrated* HIV genomes that pose an ongoing threat (as they escape the immune system as well as therapeutic agents) [see e.g. Nature Rev Microbiol (2009) 7:798–812].

In one approach to this problem, siRNAs have been used to investigate those particular histone deacetylases (see HDAC) that contribute to HIV-1 repression in latently infected cells. From the results of these studies it was suggested that HDAC inhibitors, specific for particular HDACs, may be useful for the targeted disruption of persistent HIV-1 infection [J Virol (2009) 83(10):4749–4756]. (In an analogous (murine) study, histone acetylation, but not DNA demethylation, was found to reactivate a latent gammaherpesvirus (MHV-68) in a cell line [PLoS ONE (2009) 4(2):e4556].)

In a different approach to the disruption of latency in HIV, an alternatively spliced transcription factor (ΔVII-Ets-1) was found to activate transcription in latent HIV-1 in cells derived from patients on a HAART chemotherapeutic regime. Unlike some other approaches, this method did not have the major disadvantage of causing significant T cell activation [Proc Natl Acad Sci USA (2009) 106(15):6321–6326].

aiRNA Asymmetric interfering RNA: see the entry SIRNA.

alanine scan mutagenesis A method that is used for studying the binding properties (or other characteristics) of particular residues in a protein by replacing them with alanine residues. Replacement of residue(s) may be achieved e.g. by synthesizing the protein from a modified (engineered) mRNA. After insertion of the alanine residues the protein is examined for specific properties/functions.

alarmone Any of various low-molecular-weight molecules that are able to mediate some change in cellular metabolism as a response to a particular type of stress. One example is ppGpp which is formed in the stringent response in *Escherichia coli*.

albamycin *Syn*. NOVOBIOCIN.

(See also ANTIBIOTIC (table).)

Alexa Fluor® 488 See XERODERMA PIGMENTOSUM (diagnosis by the EdU method).

AlgR In *Pseudomonas aeruginosa*: the response regulator of a TWO-COMPONENT REGULATORY SYSTEM (q.v.), FimS/AlgR, which controls biosynthesis of type IV fimbriae (responsible for so-called 'twitching motility'); this regulation involves activating the pilVWXY1Y2E operon [J Bacteriol (2008) 190(6):2023–2030].

AlgR also has roles in regulating type III protein secretion genes and alginate biosynthesis.

AlgZ *Syn*. FimS: see entry ALGR.

AlkA protein See DNA REPAIR.

alkaline phosphatase (AP) An enzyme (EC 3.1.3.1; maximum stability at pH ~7.5–9.5) which is used e.g. for removing 5′ terminal phosphate groups from nucleic acids.

AP is also used in reporter systems. For example, when it is bound to antibodies, AP can act as a label to any molecule of the relevant antigen to which the antibodies bind; the AP is then detectable by using any of various types of substrate that yield a colored product or light. Substrates that yield colored products include NBT (nitroblue tetrazolium) + BCIP (5-bromo-4-chloro-3-indolyl phosphate). ATTOPHOS is cleaved by AP to yield a fluorescent molecule (fluorophore). Several 1,2-dioxetanes, such as CSPD® and AMPPD®, give rise to CHEMILUMINESCENCE when cleaved by AP.

AP is used as a reporter e.g. in ELISA.

alkaline stripping Stripping of hybridized RNA probes from a DNA MICROARRAY by degradation with buffers that contain NaOH (sodium hydroxide) under carefully regulated conditions (e.g. a temperature of 60–62°C).

A stripped microarray can be re-used – reducing costs – but a microarray cannot be stripped twice without loss of quality. Microarrays which had been stripped once were reported to give results similar to those from virgin (non-stripped) arrays.

***O^6*-alkylguanine-DNA alkyltransferase**(AGT) See *Uses of gene fusion* in the entry GENE FUSION.

allele (allelomorph) Any one of two or more different versions of a particular GENE; the product or function of a given allele may exhibit qualitative and/or quantitative difference(s) from the product or function of other alleles of that gene.

In a diploid cell or organism, if an *allelic pair* (i.e. the two alleles of a given gene) consists of two identical alleles then the cell/organism is said to be *homozygous* for that particular gene; if different, the cell/organism is said to be *heterozygous* for the gene.

(See also ALLELE-SPECIFIC GENE EXPRESSION.)

allele drop-out (ADO) The failure of PCR to amplify one of the two alleles of a given gene.

ADO can be problematical e.g. for preimplantation genetic diagnosis (see PGD) in which the sample cell is heterozygous for the relevant gene; in some cases a PGD misdiagnosis has been due to ADO.

allele-specific DNA methylation analysis A method designed for determining allele-specific methylation. It involves PCR-amplification of BISULFITE-treated DNA followed by PYROSEQUENCING of the amplicons of each allele using primers that include allele-specific single-nucleotide polymorphisms (see SNP). Use of the method was demonstrated by analyzing methylation of the *H19* gene – in which the paternal allele is usually imprinted [BioTechniques (2006) 41(6):734–739].

allele-specific gene expression The two alleles of a given gene (in a diploid cell) may be expressed at different levels for a number of reasons – e.g. one allele may be transcriptionally silent owing to X-INACTIVATION, or it may be expressed at a different level owing to the presence of one or more polymorphisms (see SNP) which affect transcriptional regulation. Such differential allelic expression may be detected e.g. by a method in which the levels of mRNA from each of the two alleles are compared – the transcripts from one allele being distinguished from those of the other allele by the presence of a specific polymorphism (acting as a marker) [PLoS Genetics (2008) 4(2):e1000006]. Other workers screened 11500 SNPs to identify differential allelic expression and concluded that allele-specific gene expression is widespread across the genome [PLoS ONE (2009) 4(1):e4150].

Allele-specific gene expression patterns were investigated by quantitative genotyping of 2529 genes from patients with acute lymphoblastic leukemia – revealing regulation of gene expression by methylation at CpG sites [Genome Res (2009) 19(1):1–11].

(See also EPIGENETIC ALLELIC RATIO ASSAY.)

allele-specific PCR A variant form of PCR designed to amplify a particular allele of a given gene – but no other allele(s) of that gene. One of the two primers is designed with a 3′ terminal nucleotide that pairs with a *specific* base in the required allele – a base which is known to be different from that in the other allele(s) at this location; this primer can be extended on the required allele, which will be amplified. Other alleles are not amplified as the mismatch at the primer's 3′ terminal will inhibit extension.

An essential requirement is the use of a polymerase which *lacks* proofreading ability (i.e. one which lacks 3′-to-5′ exonuclease activity); such an enzyme (e.g. the *Taq* polymerase) is not able to remove the primer's terminal nucleotide (i.e. it cannot correct the mismatch) and does not amplify unwanted alleles.

It is also important to carry out the reaction with an appropriate level of stringency.

A modified form of this method is found e.g. in SNP GENOTYPING.

allelic pair See ALLELE.

allelic ratio assay See e.g. EPIGENETIC ALLELIC RATIO ASSAY.

allelomorph *Syn.* ALLELE.

allolactose β-D-Galactopyranosyl-(1→6)-D-glucopyranose: the natural inducer of the LAC OPERON in *Escherichia coli;* it is formed as a minor product during the cleavage of lactose by β-galactosidase.

(See also IPTG.)

allosteric effect The effect which is produced when the binding of a ligand to a given target molecule affects the properties of other site(s) on the same molecule; allosteric effects are due to conformational changes that result from the binding of the ligand.

allosteric nucleic acid enzymes Nucleic acid enzymes (see e.g. APTAZYME) whose function can be subject to regulation e.g. by the activity of a low-molecular-weight molecule – or the activity of an oligonucleotide.

(See BINARY DEOXYRIBOZYME LIGASE and MAXIZYME.)

alpha (α, *Lk*) A symbol for LINKING NUMBER.

alpha peptide (α-peptide) See the entry α-PEPTIDE.

Alphanodavirus See NODAVIRUSES.

Alphavirus A genus of viruses that include many which cause disease (involving e.g. encephalitis and arthralgia) in humans and animals. The (enveloped) virion is spherical, ∼50–70 nm diam., and the (monopartite) genome is linear, positive-sense ssRNA.

Alphaviruses include e.g. the SINDBIS VIRUS and Western equine encephalomyelitis virus.

alternation of generations See PLOIDY.

alternative polyadenylation See POLYADENYLATION.

alternative splicing SPLICING of a given pre-mRNA which can proceed in at least two different ways, the different modes of splicing producing mRNAs with different overall sequences.

Alternative splicing is a common (i.e. natural) phenomenon [Nucleic Acids Res (2004) 32(13):3977–3983] which allows a given gene to encode extra information. Thus, e.g. a given gene may encode two mutually antagonistic messages (whose expression must be under appropriate control). Exceptionally, it was reported that a single gene may potentially give rise to >100 different transcripts.

The regulation of (normal) alternative splicing involves a balance between certain factors which promote or inhibit the use of specific splicing sites in pre-mRNA.

Events that may occur during alternative splicing include the splicing out (loss) of an exon (*exon skipping*), inclusion of an intron (*intron retention*) and/or splicing at a site *within* an exon.

Differential, SNP-associated alternative splicing may occur in different tissues [PLoS Biol (2008) 6(12):e1000001].

A comparative study on eight organisms found alternative splicing to be more extensive in vertebrates than in invertebrates [Nucleic Acids Res (2007) 35(1):125–131].

Atypical (U1-*independent*) splicing may be a characteristic feature of some types of alternative splicing event [Nucleic Acids Res (2009) 37(6):1907–1914].

The occurrence of alternative splicing, in respect of a given gene, may be investigated (e.g.) by initially isolating mRNAs from an extract of the relevant (living) cells. For eukaryotic cells, this can be done most easily by exploiting the poly(A) tail which characteristically forms the 3′ end of mRNAs; the cell extract is passed through a column containing the ligand oligo(dT)-cellulose that binds mRNAs (by their poly(A) tails) and allows other types of RNA (e.g. ribosomal RNA) to be eliminated. When eluted from the column, the population of mRNAs is subjected to GEL ELECTROPHORESIS and the gel is then used for NORTHERN BLOTTING; individual mRNAs can be identified (on the membrane) by highly specific, labeled PROBES. This procedure can thus reveal mRNAs *of different sizes* that were derived from a given gene.

Alternative splicing may also be investigated by the use of

an EXON ARRAY (q.v.).

The choice of splice site in a transcript can be modulated, *in vitro* and *in vivo*, by using a specific antisense molecule. Thus, splicing of pre-mRNA of gene *bcl-x* normally produces a long anti-apoptotic mRNA as well as a short pro-apoptotic mRNA, and the ratio of these two mRNAs in the cell is a key determinant of cancer progression. By using a short antisense PNA conjugated to an oligopeptide containing eight serine–arginine repeats (cf. SR PROTEINS), it was possible to modify the splicing pattern so that the short (pro-apoptotic) mRNA became the dominant influence; this procedure was able to produce APOPTOSIS in HeLa (cancer) cells [Nucleic Acids Res (2005) 33(20):6547–6554].

siRNA-controlled alternative splicing has been reported in which the siRNAs targeted appropriate sequences in introns or exons [Nature Struct Mol Biol (2009) 16:717–724].

Aberrant splicing causes many types of disease. Correction of aberrant splicing has been demonstrated both *in vitro* and *in vivo* by using specifically designed *splice-switching oligonucleotides* (SSOs) that can re-direct the splicing machinery to normal splice sites. Positive results were reported with the use of SSOs in patients with Duchenne muscular dystrophy. [Therapeutic potential of splice-switching oligonucleotides (a review): Oligonucleotides (2009) 19(1):1–14.]

An alternatively spliced transcription factor was reported to disrupt latency (i.e. activate transcription) of HIV-1, offering a potential approach to the problem of quiescent infection in AIDS [Proc Natl Acad Sci USA (2009) 106(15):6321–6326].

(See also the entries CRYPTIC SPLICING, EXON TRAPPING, and *alternative polyadenylation* in POLYADENYLATION.)

Alu sequences (*Alu* sequences) In (at least) some mammalian genomes: a family of related sequences, each typically about 300 nt long and commonly having a recognition site for the RESTRICTION ENDONUCLEASE AluI (AG↓CT); the human genome may contain about one million copies. Alu sequences are RETROTRANSPOSONS; owing to their ability to generate insertional mutations they are regarded as potential factors in genetic disorders.

Most Alu sequences in the human genome (those from subfamilies S and J) seem to be largely inactive – transpositional activity being essentially confined to a much smaller group of 'younger' Alu sequences in the Y family [Genome Res (2009) 19:545–555].

Some Alu sequences with regions similar to splice sites are found in certain genes and are recognized as exons. (See also EXONIZATION.) Splicing patterns of exonized Alu sequences have been described in (human) tissues [PLoS Genet (2008) 4(10):e1000225].

A study of exonizing and non-exonizing Alu sequences has reported various features required for precise recognition of exons by the splicing machinery [PLoS Comput Biol (2009) 5(3):e1000300].

(See also SINE and LINE.)

AluI A RESTRICTION ENDONUCLEASE from *Arthrobacter luteus;* recognition site: AG↓CT.

α-amanitin A complex, substituted cyclic peptide (one member of a family of toxins produced e.g. by the 'death cap fungus', *Amanita phalloides*) which, at low concentrations, can inhibit (eukaryotic) DNA-dependent RNA polymerase II.

This agent was used e.g. for studies on RNA polymerase II [PLoS ONE (2008) 3(2):e1661; Acta Crystallogr (Section D) (2009) 65(2):112–120].

amantadine (1-adamantanamine hydrochloride) A polycyclic ANTIVIRAL AGENT used (e.g.) for prophylaxis and early treatment of infection by type A INFLUENZAVIRUS; *rimantadine*, α-methyl-1-adamantane methylamine hydrochloride, has similar activity.

Amantadine inhibits viral replication by blocking a proton channel formed by the M2 protein of influenza virus A [Proc Natl Acad Sci USA (2008) 105(5):1483–1488].

(See also *swine flu* in the entry INFLUENZAVIRUS.)

Amaxa Nucleofector® See NUCLEOFECTION.

amber codon See NONSENSE CODON.

amber mutation A mutation that creates an amber codon (see NONSENSE CODON).

amber suppressor See SUPPRESSOR MUTATION.

ambisense RNA Viral ssRNA in which some gene(s) occur in positive-sense form and other(s) occur in negative-sense form (in the same strand).

Ambisense RNA occurs e.g. in the genome in viruses of the Arenaviridae, including Lassa fever virus, and in the genome in viruses of the genus *Phlebovirus*, e.g. the Rift Valley fever virus; in Rift Valley fever virus the tripartite genome consists of two fragments of negative-sense ssRNA and one fragment of ambisense ssRNA.

In many ambisense viruses, the junction between opposite-sense ORFs in the genome exhibits a hairpin structure which acts as a termination signal. The hairpin structures appear to be absent in the small (S) ambisense fragment in Rift Valley fever virus; instead, a conserved motif was reported to act as a termination signal in both the negative-sense and ambisense fragments [J Virol (2007) 81(10):5246–5256].

AMD Age-related macular degeneration: any of certain forms of pathologic condition affecting the eyes in the elderly. One form involves the development of new blood vessels (angiogenesis) in retinal tissue; the potential for GENE THERAPY in this condition has been investigated – see e.g. AAV HELPER-FREE SYSTEM.

amelanotic melanoma See MELANOMA.

amelogenin A protein associated with dental development. A gene encoding human amelogenin (*AMELX*) occurs on the X chromosome (at location Xp22.3–Xp22.1) and another gene encoding amelogenin (*AMELY*) occurs on the Y chromosome (location Yp11.2).

The amelogenin genes are exploited e.g. in a gender identification assay: see DNA SEX DETERMINATION ASSAY.

***AMELX* gene** See AMELOGENIN.

***AMELY* gene** See AMELOGENIN.

Ames strain (of *Bacillus anthracis*) See BACTERIA (table).

Ames test (Mutatest; *Salmonella*/microsome assay) A test used

to determine whether a given agent is mutagenic (and therefore possibly carcinogenic) by investigating its ability to *reverse* an auxotrophic mutation in *Salmonella typhimurium;* the mutation in *S. typhimurium* makes the organism dependent on an exogenous source of histidine, and reversal of the mutation would allow the organism to synthesize its own histidine (i.e. it would revert to prototrophy). Different strains of *S. typhimurium* may be used, each with a different type of mutation in the histidine operon. Some of these test strains may also contain mutations which make them more permeable to certain chemicals and/or which prevent them from carrying out DNA repair. Moreover, the test strains may contain the plasmid pKM101 which includes genes for so-called error-prone repair and which therefore promotes the mutagenic effects of any DNA-damaging agents present in the reaction.

Because some chemical agents exhibit mutagenic activity only after their metabolic activation, the test system generally includes microsomal enzymes from a liver homogenate (the 9000 ***g*** supernatant, fraction 'S9') from rats pre-treated with a carcinogen to induce production of the appropriate enzymes.

When performed as a 'plate incorporation test', a culture of *S. typhimurium*, an S9 preparation and the substance under test are mixed with soft agar (which includes a low level of histidine) and this is poured onto a plate of *minimal agar* – which is then incubated at 37°C in the dark. (Minimal agar permits the growth of prototrophs but does not permit growth of the (auxotrophic) test strain of *S. typhimurium*.) The low level of histidine in the soft agar allows (only) limited growth of the (auxotrophic) *S. typhimurium* and this results in a light, confluent growth of this organism in the upper layer of agar (the 'top agar').

If the test substance had caused reversion to prototrophy in any cells of the test strain, those cells (whose growth would not be limited) can grow and form visible colonies.

When interpreting the results, several factors must be borne in mind. An absence of growth in the 'top agar' would suggest that the substance under test has general antibacterial activity, and that any colonies which develop on the plate are unlikely to be true revertants. Again, before drawing conclusions from a number of *apparent* revertant colonies, it is necessary to take into account the known *spontaneous* reversion rate of the particular mutation in the given test strain.

Various modifications of the basic Ames test are used for specific purposes.

[Ames test with a derivative of carbamic acid: Antimicrob Agents Chemother (2005) 49:1160–1168.]

An Ames test was employed in an assay of mutagenicity on certain sulfur analogs of polycyclic aromatic hydrocarbons [J Org Chem (2007) 72(22):8383–8393].

[Comparison of Ames II and traditional Ames test responses with respect to mutagenicity, strain specificities, need for metabolism and correlation with rodent carcinogenicity: Mutagenesis (2009) 24(4):359–366.]

(See also SOS CHROMOTEST.)

amethopterin (methotrexate) 4-Amino-10-methylfolic acid: an agent which blocks nucleotide synthesis; it is used e.g. in the chemotherapy of leukemia.

(cf. AMINOPTERIN.)

amikacin See AMINOGLYCOSIDE ANTIBIOTIC.

amino acid A term which (in the context of DNA technology) generally refers to one of 20 compounds (listed in the table) whose residues are common components of oligopeptides, polypeptides and proteins. Each amino acid is represented by at least one CODON (see the table, and see GENETIC CODE).

Ornithine is just one example of an amino acid which is not represented by a CODON but which is nevertheless found e.g. in certain oligopeptide antibiotics; this kind of oligopeptide is synthesized by a ribosome-free enzyme system (rather than by translation) (see NON-RIBOSOMAL PEPTIDE SYNTHETASE).

4-amino-10-methylfolic acid See AMETHOPTERIN.

aminoalkylsilane See AAS.

4-aminofolic acid See AMINOPTERIN.

aminoglycoside antibiotic Any of a group of (broad-spectrum) antibiotics in which the molecular structure typically includes an aminosugar and either 2-deoxystreptamine or streptidine; these antibiotics bind to the bacterial 30S ribosomal subunit and inhibit protein synthesis.

The aminoglycosides include amikacin, framycetin, gentamicin, hygromycin B, kanamycin, neomycin, streptomycin and tobramycin; they are typically bactericidal (at appropriate concentrations) and are active against a wide range of Gram-positive and Gram-negative species.

Resistance to aminoglycosides can arise e.g. by (i) mutation in proteins of the ribosomal 30S subunit (affecting the binding of antibiotics); (ii) inactivation of antibiotics by bacterial enzymes which e.g. carry out *N*-acetylation or *O*-phosphorylation; (iii) decreased uptake by the cell.

(See also G418 SULFATE.)

aminopterin 4-Aminofolic acid: an agent that blocks the pathway of nucleotide synthesis. Aminopterin is used e.g. in HAT medium for preparing a HYBRIDOMA (q.v.).

[Example of the use of aminopterin (for the preparation of a hybridoma): BMC Immunol (2009) 10:16.]

(cf. AMETHOPTERIN.)

AMO Anti-miRNA oligonucleotide: see e.g. MICRORNAS.

AMP CT Amplified *Chlamydia trachomatis* test: a TMA-based assay used for detecting the pathogen *Chlamydia trachomatis* in clinical specimens (Gen-Probe, San Diego CA). One early study [J Clin Microbiol (1997) 35:676–678] examined urine specimens from female patients as a non-invasive method of diagnosing chlamydial infection; both the AMP CT assay and a PCR-based method were found to be sensitive and specific methods for detecting *C. trachomatis*, and it was concluded that both methods were suitable screening procedures.

ampholyte Any electrolyte with both acidic and basic groups.

ampicillin 6(α-aminobenzylamido)-penicillanic acid: a semi-synthetic PENICILLIN used in media e.g. as a selective agent for bacteria containing a vector with an ampicillin-resistance marker gene.

amplicon (1) A specific (precise) sequence of nucleotides, part

AMINO ACIDS: symbols, molecular weights and codons

Amino acid	1-letter symbol	3-letter symbol	Molecular weight	Codons
Alanine	A	Ala	89	GCA, GCC, GCG, GCU
Arginine	R	Arg	174	AGA, AGG, CGA, CGC, CGG, CGU
Asparagine	N	Asn	150	AAC, AAU
Aspartic acid	D	Asp	133	GAC, GAU
Cysteine	C	Cys	121	UGC, UGU
Glutamic acid	E	Glu	147	GAA, GAG
Glutamine	Q	Gln	146	CAA, CAG
Glycine	G	Gly	75	GGA, GGC, GGG, GGU
Histidine	H	His	155	CAC, CAU
Isoleucine	I	Ile	131	AUA, AUC, AUU
Leucine	L	Leu	131	CUA, CUC, CUG, CUU, UUA, UUG
Lysine	K	Lys	146	AAA, AAG
Methionine	M	Met	149	AUG
Phenylalanine	F	Phe	165	UUC, UUU
Proline	P	Pro	115	CCA, CCC, CCG, CCU
Serine	S	Ser	105	AGC, AGU, UCA, UCC, UCG, UCU
Threonine	T	Thr	119	ACA, ACC, ACG, ACU
Tryptophan	W	Trp	204	UGG
Tyrosine	Y	Tyr	181	UAC, UAU
Valine	V	Val	117	GUA, GUC, GUG, GUU

of a larger nucleic acid molecule, which is copied (amplified) by an *in vitro* amplification process such as NASBA or PCR.
(2) One of the copies of a sequence of nucleotides which has been copied (amplified) by methods such as NASBA or PCR.
(3) One of a number of elements of linear DNA (~100 kb) formed in studies on the *JBP1* gene of *Leishmania tarentolae* [term used in: Nucleic Acids Res (2005) 33(5):1699–1709].
(4) *Formerly* [Proc Natl Acad Sci USA (1985) 82:694–698]: a defective virus vector.
(5) Within a Y chromosome: a segment with >99% sequence identity to other region(s) in the given chromosome (hence the adjective *ampliconic*). [Use of term: BMC Genet (2007) 8:11.]
(6) A given sequence of nucleotides *within which* a specific sub-region may be identified as being of special interest and which may be treated as an amplicon (sense 1).

amplicon containment One approach to the minimization of contamination by amplicons from previous assays in methods such as PCR. Essentially, this involves division of the working environment into several dedicated areas, each of which is used for only certain specific stage(s) of the procedure. For example, when working with PCR, it is usual to carry out the thermal cycling and the analysis of products (e.g. electrophoresis) in separate areas, and separate areas may also be specified for extracting target nucleic acid and for preparing reagents.

(See also AMPLICON INACTIVATION.)

amplicon inactivation In PCR: any method which avoids contamination by destroying carry-over amplicons from previous assays. Such contamination can be a major problem e.g. in those clinical laboratories in which specimens are examined routinely for only a small number of target sequences; under these conditions new specimens may risk contamination if amplicons are allowed to build up in the laboratory environment (e.g. in/on the equipment or in reagents).

Methods for amplicon inactivation

The uracil-N-glycosylase method. In this method, *all* assays are conducted in the normal way except that deoxythymidine triphosphate (dTTP) is replaced by deoxyuridine triphosphate

(dUTP) in the reaction mixture. All amplicons produced in each assay therefore contain dUMP instead of dTMP. These amplicons can be analysed in the normal way by gel electrophoresis etc.

In addition to the use of dUTP, the reaction mixture also includes the enzyme URACIL-N-GLYCOSYLASE (UNG). Thus, if an assay is contaminated with amplicons from a previous assay, these amplicons will act as substrates for the enzyme, uracil being cleaved from each dUMP; this, in itself, does not bring about strand breakage, but the amplicons are degraded to non-amplifiable pieces by the high temperature used for the initial denaturation of target DNA. The high temperature also inactivates UNG; this is necessary in order to avoid degradation of amplicons from the current assay. Note that the *target* DNA in the reaction is not affected by UNG as it contains dTMP.

Normally, this method cannot be used in PCR-based studies of DNA methylation in which the sample DNA is treated with BISULFITE; this is because bisulfite treatment converts non-methylated cytosines to uracil, so that the template DNA itself would be subjected to degradation. However, unlike the usual form of bisulfite treatment (in which DNA is sulfonated and is subsequently desulfonated), it has been found that non-desulfonated DNA can be amplified by PCR with the UNG method of decontamination because this (sulfonated) form of DNA is resistant to UNG. Desulfonation of DNA is achieved by a prolonged (30-minute) initial stage of denaturation (at 95°C) [Nucleic Acids Res (2007) 35(1):e4].

The isopsoralen method. In this method isopsoralen is added to the reaction mixture. Isopsoralen is a heterocyclic compound which, when bound to DNA, can form covalent interstrand crosslinks when photoactivated by ultraviolet radiation (e.g. 365 nm/15 min/4°C); activation of isopsoralen at low temperatures was reported to be more efficient than at room temperature. As the double-stranded amplicons are *covalently* crosslinked they cannot be denatured to single strands; hence, they cannot serve as templates, so that, if they contaminate a subsequent assay the outcome would not be affected.

Amplicons produced by this method are suitable for examination by processes such as gel electrophoresis and staining (e.g. for confirming the presence of a given target sequence in the sample DNA). However, they cannot be used for any process (such as SSCP analysis) that requires single-stranded samples.

APSR. In another approach to amplicon inactivation, all the primers have a 5′ tag which incorporates a binding site for a type IIS restriction endonuclease; this enzyme, in the reaction mixture, cleaves any contaminating amplicons – and is itself inactivated at the initial high-temperature stage (see APSR).

amplicon primer site restriction See APSR.

ampliconic See AMPLICON (sense 5).

amplification (of DNA *in vitro*) See DNA AMPLIFICATION.

amplification (of RNA *in vitro*) See RNA AMPLIFICATION.

amplification-refractory mutation system See ARMS.

amplified fragment length polymorphism (AFLP) See AFLP.

amplified restriction fragment length polymorphism A non-recommended name which has been used for a method for TYPING bacteria – see the entry AFLP.

Ampligase® See DNA LIGASE.

amplimer Any primer used in PCR.

AmpliTaq^{TM} Gold DNA polymerase A 94-kDa thermostable, recombinant DNA polymerase (Applied Biosystems) used in PCR; its optimal extension temperature is reported to be 72–80°C, with an extension rate of 2 to 4 kb per minute at 72°C.

[Uses (e.g.): Malaria J (2009) 8:154; Mol Vision (2009) 15: 1620–1630; BMC Cancer (2009) 9:258.]

AmpliWax^{TM} See HOT-START PCR.

AMPPD® A 1,2-dioxetane substrate that emits light when dephosphorylated by ALKALINE PHOSPHATASE (AP). It is used e.g. for detecting AP-labeled probes.

(See also CHEMILUMINESCENCE.)

amprenavir See PROTEASE INHIBITORS.

AMTDT Amplified *Mycobacterium tuberculosis* direct test: a TMA-based assay for detecting *Mycobacterium tuberculosis* in clinical specimens (Gen-Probe, San Diego CA).

The AMTDT was approved in 1995 by the American FDA (Food and Drug Administration) for use with *smear-positive* respiratory specimens; a smear-positive specimen is one from which a smear showing ACID-FAST BACILLI can be prepared.

In the original test, the specimen was initially treated with a MUCOLYTIC AGENT, decontaminated with sodium hydroxide, and finally sonicated (to lyse organisms and release nucleic acids). The specimen was then heated (95°C/15 minutes) to remove intra-strand base-pairing in the rRNA.

The reaction mixture contained 45 μL or 50 μL of sample, and amplification (at 42°C) was conducted for 2 hours. The amplification product was detected by the addition of target-specific probes (for details see entry ACCUPROBE).

An attempt was made to adapt AMTDT for the detection of *Mycobacterium tuberculosis* in *non*-respiratory specimens [J Clin Microbiol (1997) 35:307–310], and studies were made to compare the original and subsequent (improved) versions of AMTDT for the detection of *M. tuberculosis* in respiratory and non-respiratory specimens [J Clin Microbiol (1998) 36: 684–689].

The new-format 'enhanced' AMTDT was approved by the FDA in 1998. Among other changes, this version involved the use of a 450 μL aliquot of the sample (instead of a 40–50 μL sample).

One study reported false-positive results in tests on sputa from patients infected with *Mycobacterium kansasii* and *M. avium;* these species of *Mycobacterium* are not infrequently isolated from infections in immunocompromised patients. The authors of this study suggested a change in the threshold value of luminometer readings considered to be an indication of a positive result [J Clin Microbiol (1999) 37:175–178].

anaerobic respiration RESPIRATION (q.v.) which occurs in the absence of oxygen.

analyte In a test system: that component whose properties are being studied/measured.

anchor primer A primer that binds to an ANCHOR SEQUENCE.

anchor sequence Commonly, a sequence of nucleotides, with a known composition, which is present in a given molecule or which is ligated to another sequence (or added by tailing) in order to serve a particular function – e.g. as a primer-binding site. For example, in ANCHORED PCR an anchor sequence is used to provide an otherwise unavailable site for priming the amplification of an unknown sequence.

Certain natural sequences, such as the poly(A) tail on many mRNA molecules, have been described as anchor sequences.

anchored PCR A form of PCR used for amplifying an unknown sequence of nucleotides adjacent to a known sequence on a fragment of DNA; this approach addresses the problem of the lack of a primer-binding site in the unknown sequence.

To each end of the fragment is ligated a short segment of DNA of known sequence (e.g. a linker). (If the fragment has 3′ or 5′ overhangs then these can be eliminated, enzymically, in order to prepare the fragment for blunt-end ligation to the linkers.) Following ligation, that linker which is contiguous with the unknown region provides an ANCHOR SEQUENCE which is able to serve as a primer-binding site for one of the PCR primers. The second primer is designed to bind at a site within the known sequence. If PCR is primed in this way, the resulting amplicons will include the unknown sequence and at least part of the known sequence. If the known sequence occurs in the *center* of the fragment, then amplification, as described above, can be carried out for both of the unknown flanking regions.

(See also TAIL-PCR and VECTORETTE PCR.)

anchoring enzyme In SAGE (q.v.): a name sometimes given to the enzyme used for initial cleavage of the cDNAs.

ancient DNA (aDNA) DNA recovered from specimens which are thousands or millions of years old.

[Isolation of ancient DNA (optimization of procedures): BioTechniques (2007) 42(3):343–352.]

(See also SPEX.)

Studies on (permafrost-derived) mammoth bones provided some indication that mitochondrial DNA is preserved better than nuclear DNA in permafrost [Nucleic Acids Res (2009) doi: 10.1093/nar/gkp159].

ANDENYALAA The (10-amino-acid) tag which functions as a proteolysis signal on an aborted protein in the TRANS TRANSLATION process (q.v.).

aneuploid Refers to a genome that has one or more chromosomes in excess of, or less than, the number characteristic of the species (see e.g. DOWN'S SYNDROME).

Angelman syndrome A disorder that may be caused by any of various genetic mechanisms – see the table in entry GENETIC DISEASE for further details.

angiogenesis (neovascularization) Development of (new) blood vessels – e.g. in an embryo or in a tumor.

(See also CANCER THERAPY.)

annealing The hybridization of two complementary (or near-complementary) sequences of nucleotides to form a double-stranded molecule or a double-stranded region within a larger molecule (e.g. the binding of primers to primer-binding sites in PCR).

annotation Information supplied with the database entry of a given sequence or gene.

For an uncharacterized gene, the annotation may include a predicted function and/or an attempted characterization of the gene's product based on sequence homology with a gene of known function present in another organism.

(See also ACCESSION NUMBER.)

antagomir Any of a range of synthetic, chemically engineered oligonucleotides that bind to, and antagonize, specific types of MICRORNA molecule. Intravenous administration (in mice) of antagomirs directed against particular miRNAs resulted in efficient and specific silencing of the given miRNAs [Nature (2005) 438:685–689].

[Lentivirus-mediated antagomir expression for the specific inhibition of miRNA function: Nucleic Acids Res (2007) 35: e149.]

Antagomirs have been used to study the role of microRNAs miR-27a and miR-451 in regulating the expression of *MDR*1/P-glycoprotein in ovarian cancer cells [Biochem Pharmacol (2008) 76(5):582–588], and in a study on the TAR miRNA of HIV-1 [Retrovirology (2009) 6:18].

anthrax toxin A toxin, produced by the Gram-positive pathogen *Bacillus anthracis*, which gives rise to the symptoms of anthrax. It comprises three protein components – each, alone, being unable to function as a toxin; these three proteins are encoded by the plasmid pXO1. (Another plasmid, pXO2, is needed for the pathogenicity of *B. anthracis;* this encodes an essential anti-phagocytic capsule which protects the organism from the host's immune system.)

One component of the toxin localizes in the cell membrane and permits internalization of the other two components – a zinc protease (which disrupts intracellular signaling) and an ADENYLATE CYCLASE (which e.g. promotes edema).

anti Abbreviation for ANTICLINAL.

antibiotic Any of an extensive range of natural, semi-synthetic and fully synthetic compounds which, in low concentrations, are able, selectively, to inhibit or kill specific types of micro-organism and, in some cases, other types of cell – e.g. tumor cells; an antibiotic acts at specific site(s) in a susceptible cell. (Compounds that are active against *viruses* are usually called 'antiviral agents' rather than antibiotics.)

Natural antibiotics (see e.g. BACTERIOCIN) have ecological roles. Some types of antibiotic have medical/veterinary uses in the prevention and/or treatment of infectious diseases, and some (e.g. nisin) are used as food preservatives.

(For examples of antibiotics see: AMINOGLYCOSIDE ANTIBIOTIC; CHLORAMPHENICOL; β-LACTAM ANTIBIOTICS; OXAZOLIDINONE ANTIBIOTICS, QUINOLONE ANTIBIOTICS, QUINOXALINE ANTIBIOTICS, RIFAMYCINS and VANCOMYCIN.)

In nucleic-acid-based technology, antibiotics are used in a variety of *selective* procedures. The presence of an antibiotic

ANTIBIOTIC: some antibiotics used in DNA technology (e.g. for marker selection)

Antibiotic	Group	Target organisms	Antibiotic action
Actinomycin D	–	Prokaryotic and eukaryotic	Intercalating agent; inhibits DNA-dependent RNA polymerase
Ampicillin	β-Lactams	Bacteria	Blocks synthesis of cell-wall polymer peptidoglycan
Blasticidin S	Nucleoside	Prokaryotic and eukaryotic	Inhibits protein synthesis by inhibiting the peptidyltransferase-mediated reaction at the ribosome
Carbenicillin	β-Lactams	Bacteria	Blocks synthesis of cell-wall polymer peptidoglycan
Cefotaxime	β-Lactams	Bacteria (mainly Gram-negative)	Blocks synthesis of cell-wall polymer peptidoglycan
Chloramphenicol (= chloromycetin)	–	Bacteria (broad spectrum), some fungi	Binds to prokaryotic and mitochondrial ribosomes; inhibits peptidyltransferase and (hence) protein synthesis
G418 sulfate	Related to gentamicin	Prokaryotes, yeasts, plants, mammalian cells	Inhibits protein synthesis
Gentamicin	Aminoglycosides	Bacteria	Binds to 30S subunit of bacterial ribosomes and inhibits protein synthesis
Hygromycin B	Aminoglycosides	Mammalian and plant cells	Inhibits protein synthesis
Kanamycin	Aminoglycosides	Bacteria	Binds to 30S subunit of bacterial ribosomes and inhibits protein synthesis
Kasugamycin	Aminoglycosides	Bacteria, some fungi	Inhibits polypeptide chain initiation and, hence, protein synthesis
Mycophenolic acid	–	Bacteria (also antitumor agent)	Inhibits synthesis of guanosine monophosphate, inhibiting synthesis of nucleic acids
Nalidixic acid	Quinolones	Bacteria (mainly Gram-negative)	Inhibits function of the A subunit of gyrase; inhibits DNA synthesis
Novobiocin	–	Bacteria	Inhibits binding of ATP to the B subunit of gyrase, inhibiting DNA synthesis
Penicillin G (= benzylpenicillin)	β-Lactams	Bacteria (mainly Gram-positive)	Blocks synthesis of cell-wall polymer peptidoglycan
Polymyxin B	Polymyxins	Bacteria (mainly Gram-negative)	Increases permeability of the cytoplasmic membrane and affects the integrity of the outer membrane
Puromycin	Nucleoside	Prokaryotic and eukaryotic	Inhibits protein synthesis by acting as an analog of part of an aminoacyl-tRNA
Rifampicin	Rifamycins	Bacteria (mainly Gram-positive)	Inhibits DNA-dependent RNA polymerase by binding to the β subunit of the enzyme
Streptomycin	Amino glycosides	Bacteria, some fungi	Interacts with ribosomes and inhibits protein synthesis
Tetracycline	Tetracyclines	Bacteria	Binds to ribosomes; inhibits protein synthesis by blocking the binding of aminoacyl-tRNAs to the A site
Zeocin™	Bleomycin/ phleomycin	Bacteria, yeast, and mammalian cells	Binds to, and cleaves, DNA

in a growth medium selects for those cells that are expressing a gene conferring resistance to that antibiotic; those cells that are susceptible to the antibiotic – i.e. which do not contain a relevant antibiotic-resistance gene – are growth-inhibited or killed by the antibiotic. An antibiotic-resistance gene may be included e.g. in a vector used for transfection of a population of cells; cells which internalize the vector (and which express the antibiotic-resistance gene) are then selected by growth on a medium containing the relevant antibiotic.

An antibiotic may be MICROBICIDAL or MICROBISTATIC; at lower concentrations, a microbicidal antibiotic may behave as a microbistatic antibiotic.

A mixture of antibiotics may behave synergistically or antagonistically (or may not display either effect).

Synergism is shown when different antibiotics, that are acting (simultaneously) on a given organism, produce an effect which is greater than the sum of their individual effects. For example, the antibiotics sulfamethoxazole and trimethoprim block different reactions in the same major metabolic pathway: sulfamethoxazole inhibits the formation of dihydrofolic acid (DHF), and trimethoprim inhibits the conversion of DHF to the important coenzyme tetrahydrofolate (THF); these two antibiotics act synergistically and they are used, together, in the therapeutic agent *cotrimoxazole*.

Antagonism, the converse of synergism, can occur in different ways. In one form, an antibiotic that inhibits growth (e.g. CHLORAMPHENICOL) antagonizes those antibiotics (such as the penicillins and other β-lactam antibiotics) whose activity depends on growth in the target cell. In a different form of antagonism, certain antibiotics stimulate cells to produce enzymes that inactivate *other* antibiotics; in one example, the β-lactam imipenem (or cefoxitin) induces the synthesis of β-lactamases – enzymes which can inactivate certain other β-lactam antibiotics.

Modes of action

To be effective at all, an antibiotic must be able to enter, or pass through, the cell envelope in order to reach the relevant target site(s). Moreover, an antibiotic can be effective against a given population of cells only if its concentration is above the appropriate minimum level for that agent under the given conditions.

Modes of action include:

- Interference with DNA gyrase (a topoisomerase), with consequent inhibition of DNA synthesis (e.g. novobiocin, quinolone antibiotics).
- Depletion of guanine nucleotides (by inhibiting synthesis of GMP), affecting the synthesis of nucleic acids (e.g. MYCOPHENOLIC ACID).
- Binding to ribosomes and inhibiting protein synthesis (e.g. aminoglycoside antibiotics, chloramphenicol, macrolide antibiotics (such as erythromycin), tetracyclines, viomycin).
- Binding to RNA polymerase, inhibiting transcription (e.g. rifamycins).
- Disruption of the bacterial cytoplasmic membrane, altered permeability of the membrane affecting the cell's integrity/survival (e.g. gramicidins, polymyxins).
- Inhibition of synthesis of the bacterial cell wall polymer peptidoglycan, leading to cell lysis (e.g. β-lactam antibiotics, vancomycin).
- Inhibition of the enzyme dihydrofolate reductase, thereby inhibiting tetrahydrofolate-dependent reactions (that include synthesis of deoxythymidine – and, hence, synthesis of DNA) (e.g. pyrimethamine, trimethoprim).
- Interference with DNA function by intercalating agents (e.g. actinomycin D, quinoxaline antibiotics).
- Interaction with sterols in the cytoplasmic membrane (e.g. in yeasts and other fungi), causing leakage (e.g. polyene antibiotics).
- Inhibition of the enzyme chitin synthase (in certain fungi), affecting cell wall synthesis (e.g. polyoxins).

Mechanisms of bacterial resistance to antibiotics

Resistance to a particular type of antibiotic is *constitutive* in cells which (i) lack the antibiotic's specific target, (ii) have a variant form of the target which is not susceptible to the antibiotic, and (iii) are impermeable to the antibiotic. Examples: (i) *Mycoplasma* is resistant to β-lactam antibiotics because it lacks a cell wall; (ii) strains of *Staphylococcus aureus* known as MRSA (methicillin-resistant *S. aureus*) generally contain a modified target PBP that is not susceptible to methicillin; (iii) typically, Gram-negative bacteria are insensitive to penicillin G (a β-lactam antibiotic) because this antibiotic is not able to penetrate the outer membrane of Gram-negative bacteria.

As well as constitutive resistance (see above) resistance can be acquired e.g. by mutation, or by the acquisition of plasmid (or transposon) gene(s) specifying resistance to one or more types of antibiotic.

Examples of resistance mechanisms include:

- Due to mutation, the target of a given antibiotic may be altered so that it fails to bind the antibiotic; consequently, the target (e.g. an enzyme) is not affected by otherwise inhibitory concentrations of that antibiotic. Thus e.g. a mutant form of the ribosomal protein L22 in *Staphylococcus aureus* confers resistance to quinupristin/dalfopristin (Synercid®), a streptogramin, and in *Mycobacterium tuberculosis* point mutations in the *rpoB* gene (encoding the β subunit of RNA polymerase) can confer resistance to rifamycins (such as rifampin) for which RNA polymerase is the target. A novel mechanism for antibiotic resistance has been reported in the Gram-positive pathogen *Streptococcus pneumoniae;* in this organism, resistance to linezolid (an oxazolidinone) is linked to mutation in a gene encoding a methyltransferase which, in *sensitive* strains, methylates a site in the 23S rRNA (the target for linezolid) [Genome Res (2009) 19:1214–1223].
- Transposon Tn*10* encodes an inducible efflux system that enables certain Gram-negative bacteria to externalize tetracycline via an 'efflux pump' located in the cell envelope.
- Mutant forms of certain envelope proteins are associated with decreased permeability. For example, alteration in outer membrane porins in *Enterobacter aerogenes* increases resistance to certain antibiotics, and in *Pseudomonas aeruginosa*

resistance to the aminoglycosides and other antibiotics can be determined through membrane permeability controlled by a TWO-COMPONENT REGULATORY SYSTEM.

- Degradation of antibiotics by plasmid-encoded or chromosome-encoded enzymes. Such enzymes include the inducible and constitutive β-LACTAMASES that cleave the β-lactam ring in, and inactivate, β-lactam antibiotics such as penicillins and cephalosporins. The enzyme chloramphenicol acetyltransferase, which degrades CHLORAMPHENICOL, is another example – as are acetyltransferases, adenylyltransferases and phosphotransferases that inactivate aminoglycoside antibiotics.
- Increased production of an affected metabolite. Thus, for example, synthesis of higher levels of *p*-aminobenzoic acid (PABA) may overcome the effect of competitive inhibition by sulfonamides.
- Gene amplification. At least some bacteria respond to antibiotics by a gene amplification mechanism which can e.g. upregulate enzymes that hydrolyze antibiotic(s). [Bacterial gene amplification: Nature Rev Microbiol (2009) 7:578–588.]

[Antibiotic resistance genes (database): Nucleic Acids Res (2009) 37(Database issue):D443–D447.]

Nucleic-acid-based tests for antibiotic resistance

See the entry ANTIBIOTIC RESISTANCE TESTING.

antibiotic resistance testing (nucleic-acid-based approaches for bacteria) The traditional, culture-based approaches for detecting antibiotic resistance in bacteria have been appropriate mainly for the species (e.g. *Escherichia coli*, *Salmonella* spp) that grow rapidly on laboratory media; for these species, sensitivity to a given antibiotic is shown by growth inhibition in the presence of the given antibiotic. Resistance to particular antibiotics in such organisms may be determined e.g. by growing the organisms on agar plates that contain antibiotic-impregnated paper disks (on the surface); antibiotics diffuse out from the disks, and organisms that are sensitive to any given antibiotic form a zone of growth-inhibition around the relevant disk.

Growth-based methods are less satisfactory for some slow-growing species (such as *Mycobacterium tuberculosis*) – and for pathogens such as *Chlamydia trachomatis* (which have a complex life cycle, and which fail to grow on ordinary laboratory media). Moreover, while culture-based methods can, in some cases, indicate the MIC (i.e. the minimum inhibitory concentration) of a given antibiotic (using e.g. the so-called E test) they give no information regarding the precise causation (i.e. mechanism) of resistance.

Nucleic-acid-based ('genotypic') methods for detecting antibiotic resistance (in various organisms) have developed over the last few decades. Below is a brief glimpse at some of the methods used for two important pathogens: *Staphylococcus aureus* and *Mycobacterium tuberculosis*.

Staphylococcus aureus

Among early studies, a BDNA ASSAY was used for detecting the MECA GENE of *S. aureus* in samples from blood-culture bottles (without the usual prior subculture to solid media) [J Clin Microbiol (1999) 37:4192–4193]; it was suggested that the results of this approach could help the clinician to choose an appropriate antibiotic (β-lactam or vancomycin) to match the actual requirement of an infection.

MULTIPLEX PCR has also been used for detecting the *mecA gene* (and the COA GENE) in *S. aureus*.

A pentaplex assay (targeting five genes) has been designed (a) to detect *S. aureus*, (b) to identify the *mecA* gene, and (c) to help distinguish between *community-acquired* MRSA and hospital-acquired MRSA (see the entry MRSA); for the latter purpose, one of the targeted genes was *lukS* – which encodes the PANTON–VALENTINE LEUCOCIDIN (q.v.) [BMC Microbiol (2009) 9:13].

Mycobacterium tuberculosis

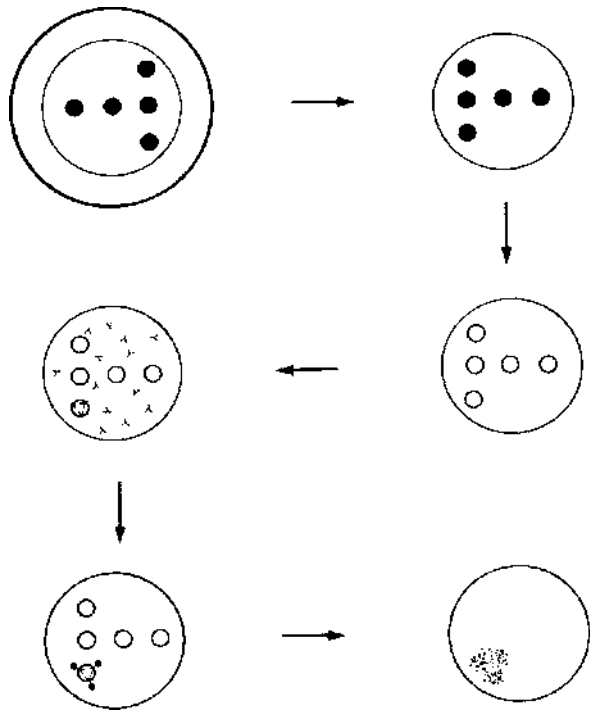
The resurgence of *M. tuberculosis* as a major threat, and the appearance of multidrug-resistant (MDR) strains, has given a strong impetus to the search for suitable nucleic-acid-based methods for detecting antibiotic resistance in this organism. Unlike resistance to β-lactam antibiotics in *S. aureus*, which is commonly due to specific gene products (e.g. PBP 2a or β-lactamase enzymes), the resistance of *M. tuberculosis* to anti-tuberculosis drugs usually arises through mutation(s); these mutations affect the genes whose products are the targets of anti-tuberculosis drugs. While resistance to some drugs (e.g. isoniazid, streptomycin) can be due to mutation in more than one gene, resistance to rifampin (see RIFAMYCINS) is usually due to mutation(s) in one gene, *rpoB*, which encodes the β-subunit of the RNA polymerase. Hence, detecting resistance to rifampin is technically simpler; moreover, strains that are resistant to rifampin are frequently resistant to various other anti-tuberculosis drugs, so that rifampin resistance may be a useful marker for multi-resistant strains.

Mutations in the *rpoB* gene are detected in various ways. In some methods the initial step is to use PCR to copy the *rpoB* gene – or a specific region of *rpoB* in which the resistance-associated mutations commonly occur. The PCR products are then examined e.g. by SSCP ANALYSIS or by a LINE PROBE ASSAY. Both methods are able to indicate *specific* mutations in the *rpoB* gene. (By identifying specific mutations, methods such as these are valuable e.g. for tracking resistance in the pathogen population and for epidemiological studies.)

In a different approach, *M. tuberculosis* was infected with mycophages carrying a fluorescent reporter gene (encoding either green fluorescent protein or ZsYellow); infection of these cells was carried out in the presence of either streptomycin or rifampin. The cells that were susceptible (sensitive) to the drug failed to exhibit fluorescence because (owing to inhibition of their RNA polymerase) the reporter gene was not transcribed; cells that were resistant to the drug were able to transcribe the reporter gene and (hence) exhibited fluorescence [PLoS ONE (2009) 4(3):e4870].

antibody-based library screening An alternative to screening with a nucleic acid probe (cf. COLONY HYBRIDIZATION). The method is shown diagrammatically in the figure.

antibody-labeling reagents See e.g. ZENON ANTIBODY LABELING REAGENTS.



ANTIBODY-BASED LIBRARY SCREENING: screening a cDNA expression library (diagrammatic). The cDNA library – consisting of protein-encoding cDNAs within expression vectors – was inserted into a population of bacteria; these bacteria were then inoculated onto agar plates in such a way that each bacterium gave rise to an individual colony. The object now is to determine *which* colony contains the cDNA that encodes (and expresses) the particular protein of interest. While each plate may contain >100 colonies, the plate considered here has only five colonies (for clarity).

The cDNA-containing colonies (*top*, *left*) are lightly overlaid with a nitrocellulose filter; when lifted off, the filter contains a mirror-image replica of the colonies (*top*, *right*). The cells on the filter are lysed, and the proteins they contain (including any encoded by the cDNAs) are bound to the filter (*center*, *right*). The filter is then treated with antibodies (Y) that are specific to the protein of interest (*center*, *left*); antibodies have bound to the given protein (present in *one* of the colonies). Unbound antibodies are washed away. The filter is then treated with *protein A* – a protein (derived from the bacterium *Staphylococcus aureus*) which binds to antibodies. Unbound protein A is washed away, leaving some protein A (•) bound to the specific antibody–protein complex (*bottom*, *left*). Before use, the protein A is labeled with a radioactive source so that its presence can be detected by *autoradiography;* this involves exposing a photographic film to the nitrocellulose filter for an appropriate time and then developing the film. When the film is developed (*bottom*, *right*) it identifies the particular colony on the original plate which contains the cDNA of interest. This colony can then be used as an inoculum to grow more of the cells which contain the specific cDNA.

This procedure clearly depends on efficient and specific protein–antibody binding. A protein synthesized in bacteria from a *eukaryotic* gene may lack the three-dimensional structure and/or post-translational modification that characterizes the native eukaryotic protein; it may therefore fail to bind antibodies raised against the native protein. (Moreover, even if the protein *is* synthesized with normal conformation, the process used to bind it to the filter may alter that conformation.) However, it is possible to produce antibodies that recognise, and bind to, a short, specific run of amino acid residues, even when the target protein lacks the conformation and/or post-translational modification of the native protein.

Finally, a colony containing the *particular* cDNA of interest may not be present on the plate being examined. Hence, it may be necessary to carry out the procedure on more than one plate.

Figure reproduced from *Bacteria in Biology, Biotechnology and Medicine*, 6th edition, Figure 8.10, page 222, Paul Singleton (2004) John Wiley & Sons Ltd, UK [ISBN 0-470-09027-8] with permission from the publisher.

anti-cancer therapy (*DNA technol.*) See CANCER THERAPY.

anticlinal (*anti*) Of a nucleotide: the conformation in which the oxygen atom within the sugar ring (–O–) is at the maximum distance from the 6-position of a purine (or the 2-position of a pyrimidine).

(cf. SYNCLINAL.)

anticoagulant (*DNA technol.*) A term that usually refers to an agent which inhibits the coagulation (i.e. clotting) of blood.

The anticoagulants include sodium citrate, sodium oxalate, heparin and SODIUM POLYANETHOLESULFONATE (SPS); the latter two agents can inhibit PCR.

anticodon Three consecutive bases in a tRNA molecule which are complementary to a CODON which specifies the particular amino acid carried by that tRNA.

An anticodon is written in the 5′-to-3′ direction – as is a codon.

(See also WOBBLE HYPOTHESIS.)

anti-downstream box (*or* antidownstream box) See DOWNSTREAM BOX.

antigenic drift (in viruses) See the entry GENETIC DRIFT.

antigenic variation Successive changes in cell-surface antigens which are exhibited by certain types of microorganism (e.g. *Trypanosoma* – see e.g. VSG).

At least two (distinct) mechanisms are known: (i) the alternative antigens are transcribed from specific, *pre-existing* genes (see PHASE VARIATION), and (ii) the alternative antigens arise by ongoing recombinational events – i.e. they are encoded by newly formed, rather than pre-existing, genes. An example of the second mechanism is the formation of new versions of a subunit in the fimbriae of *Neisseria gonorrhoeae;* variant forms of this subunit arise through repeated recombination between the chromosomal subunit gene, *pilE*, and another chromosomal gene, *pilS*, as well as between *pilE* and any homologous DNA received e.g. by transformation.

Some surface proteins of the fungus *Aspergillus fumigatus* may be hypervariable owing to recombination among internal mini- or microsatellite tandem repeat sequences in the genes encoding them [Eukaryotic Cell (2007) 6(8):1380–1391].

Antigenic variation apparently helps a pathogen to evade a host's immunologic defense mechanisms.

anti-miRNA oligonucleotide See e.g. the entry MICRORNAS.

antimutator gene Any gene whose activity reduces the rate of spontaneous mutation in a cell. For example, some strains of *Escherichia coli*, with a mutant form of DNA polymerase, have mutation rates below those of wild-type strains; in this case the antimutator activity presumably involves improved fidelity/proof-reading.

antiparallel (of strands in dsDNA) The (usual) arrangement in which a 5′-to-3′ strand is hybridized to a 3′-to-5′ strand, i.e. each end of a double-stranded DNA molecule has a 5′ terminal and a 3′ terminal.

(See also DNA.)

antiretroviral agents Agents with activity against retroviruses; some are useful e.g. in chemotherapy against AIDS – see e.g. the NON-NUCLEOSIDE REVERSE TRANSCRIPTASE INHIBITORS, NUCLEOSIDE REVERSE TRANSCRIPTASE INHIBITORS and PROTEASE INHIBITORS. Other agents that are useful against AIDS include the INTEGRASE INHIBITORS, THE FUSION INHIBITORS and the CHEMOKINE CORECEPTOR ANTAGONISTS.

(See also HAART, HIV-1 and RETROVIRUSES.)

The ability of certain indole derivatives to inhibit synthesis of a retroviral glycoprotein, protecting mice from a challenge with MLV (murine leukemia virus), has suggested a potential use for these compounds as antiretroviral therapeutic agents [PLoS ONE (2009) 4(2):e4533].

Specific HDAC inhibitors (see HDAC), as well as genome-specified agents, have been suggested as a means to address the problem of *latency* in HIV-1: see *Quiescent HIV-1* in the entry AIDS.

anti-reverse cap analog (ARCA) A chemically modified form of CAP ANALOG (Ambion, Austin TX) designed to maximize the efficiency of *in vitro* translation by ensuring that the cap analog is incorporated in the transcript in the correct orientation (a cap analog incorporated in the reverse orientation does not support translation).

The use of an anti-reverse cap analog enhanced translation by a factor of 1.5–1.8 in studies on translational efficiency of mRNA encoding the HIV-1 *gag* region [PLoS ONE (2008) 3(6):e2356].

antisense gene In a genetically engineered cell: any gene, inserted into that cell, whose presence is intended to inhibit or block the expression of another, endogenous gene.

antisense RNA Any natural or synthetic RNA whose sequence permits interaction with a given sense sequence – in RNA or in DNA – and which can affect the activity/expression of the given target molecule: see e.g. FINOP SYSTEM, MICRORNAS, MULTICOPY INHIBITION, POST-SEGREGATIONAL KILLING, the R1 PLASMID, RNA INTERFERENCE.

Other antisense systems

Engineered antisense systems have been devised using PNA or MORPHOLINO ANTISENSE OLIGOMERS.

(See also GENE SILENCING.)

antisense strand (of DNA) The non-CODING STRAND.

antisense transcriptome See TRANSCRIPTOME.

antiviral agent Any agent which has activity against viruses – *in vivo* and/or *in vitro*. (cf. ANTIBIOTIC.)

(See e.g. ANTIRETROVIRAL AGENTS; see also ACYCLOVIR, AMANTADINE, GANCICLOVIR, LAMIVUDINE, OSELTAMIVIR, RIBAVIRIN AND ZANAMIVIR.)

antizyme Any of a group of proteins associated with regulation of ornithine decarboxylase (ODC) – which is involved in biosynthesis of polyamines. In eukaryotes, functional antizyme is expressed in the presence of increased levels of polyamines – antizyme 1 promoting UBIQUITIN-dependent degradation of ODC via the 26S PROTEASOME. Antizymes are regulated by antizyme inhibitor.

[Antizyme in prokaryotes: BMC Biochem (2007) 8:1.]

AOX1 In the (methylotrophic) yeast *Pichia pastoris*: a highly regulated, inducible gene which encodes alcohol oxidase – a peroxisomal enzyme involved in metabolism of methanol.

(See also PPICZ VECTOR.)

AP ALKALINE PHOSPHATASE.

AP endonuclease Any enzyme with endonuclease activity that is involved in the excision of apurinic/apyrimidinic nucleotide residues (see e.g. BASE EXCISION REPAIR).

AP-PCR Arbitrarily primed PCR: any form of PCR which uses primers of arbitrary sequence and which amplifies random, but discrete, sequences of chromosomal DNA; AP-PCR has been used for TYPING bacteria.

PCR is initially carried out under low stringency, and the primers bind at various sites to each strand of heat-denatured chromosomal DNA; the binding of primers occurs at 'best-fit' sequences, and may include mismatches. In some cases two primers bind with relative efficiency, on opposite strands, at locations separated by a few hundred bases. If synthesis can occur normally from these two primers, a further round of cycling under low-stringency conditions, followed by many cycles under high-stringency conditions, may produce copies of an amplicon delimited by the two best-fit sequences. In the phase of high-stringency cycling not all the primers will bind to their best-fit sequences – so that only a proportion of the amplicons produced under low-stringency conditions will be amplified in the high-stringency phase.

The amplicons from a given sample are subjected to gel electrophoresis, and the stained bands of amplicons form the fingerprint. Strains are compared and classified on the basis of their fingerprints.

One advantage of this approach is that there is no need for prior knowledge of the genome sequence; there is no need to design specific primers, and any isolate is potentially typable.

Results are generally reproducible under standardized conditions in a given laboratory, but comparable results will not necessarily be obtained in other laboratories unless the procedures are *identical;* reproducibility of results depends not only on the primer sequence but also e.g. on the particular type of polymerase used and on the initial procedure used for preparing the sample DNA.

Some other named methods are based on the same principle – e.g. RAPD (random amplified polymorphic DNA) analysis and DAF (direct amplification fingerprinting). Such methods may differ e.g. in the length of primers used, annealing temperature for the primers, and the type of gel used for electrophoresis. The original AP-PCR employed primers of 20 to 50 nucleotides, an annealing temperature of ~40°C, and agarose gel. In RAPD the primers are often 10–20 nt, with annealing at ~36°C, and products separated in an agarose gel. DAF uses short primers (5–8 nt) at an annealing temperature of ~30°C; as there are many more (smaller) products, electrophoresis is carried out in a polyacrylamide gel and silver staining is used to detect bands in the fingerprint.

AP site (abasic site) In a nucleotide sequence: a site at which the base (purine or pyrimidine) is missing – the remainder of the nucleotide (sugar, phosphate) being present. (cf. GAP.)

APES See the entry AAS.

aphidicolin A tetracyclic diterpenoid, isolated from a fungus, which strongly inhibits the eukaryotic α DNA polymerase. Bacterial DNA polymerases are unaffected, but aphidicolin was reported to inhibit DNA synthesis in at least some members of the domain ARCHAEA (e.g. some methanogens).

[Example of use: Proc Natl Acad Sci USA (2009) 106(3): 803–807.]

Aphthovirus A genus of PICORNAVIRUSES which includes the causal agent of foot-and-mouth disease.

[Inhibition of replication of *Aphthovirus* by siRNA: Virol J (2008) 5:86. Diagnosis of foot-and-mouth disease in Brazil by RT-PCR: BMC Vet Res (2008) 4:53.]

APO-1 (CD95) The cell-surface Fas receptor: see APOPTOSIS.

APOBEC-1 See CYTIDINE DEAMINASE and RNA EDITING.

apolipoprotein B See RNA EDITING.

apoptosis In various types of eukaryotic cell: death that occurs in a regulated ('programed') manner – an organized process in which fragmentation of the genome is a characteristic feature.

DNA fragmentation can be assessed by the TUNEL ASSAY.

In the vertebrates, apoptosis occurs naturally, for example, during embryogenesis. It occurs during metamorphosis in the invertebrates.

Apoptosis can be a response to damaged DNA in the cell: see e.g. the entry P53.

Apoptosis can be induced by physical agents, such as heat and radiation, and by certain chemical agents; for example, in osteoblasts, it may be stimulated by glucocorticoids (leading to osteoporosis).

Major players in apoptosis are certain intracellular cysteine proteases (*caspases*) that are synthesized in an inactive form (zymogen) and activated specifically during apoptosis; these enzymes cleave specific protein substrates at a site next to an aspartic acid residue. Cleavage by a caspase may inactivate or activate the substrate – e.g. caspase-mediated activation of the nuclease *caspase-activated DNase* (CAD) results in DNA fragmentation. (CAD is normally present in an inactive form, complexed with an inhibitory partner known as ICAD.)

Caspase activation apparently occurs primarily by cleavage of the zymogen. In at least some cases, one caspase may be activated by another, 'upstream' caspase in a caspase cascade.

In macrophages that have taken up the bacterium *Shigella flexneri* (by phagocytosis), apoptosis in the macrophage can be promoted by a plasmid-encoded protein, IpaB, secreted by *S. flexneri;* IpaB activates the host's IL-1β-converting enzyme (ICE, a cysteine protease) which then initiates apoptosis.

Other factors that may promote apoptosis include binding of certain cytokines (e.g. TNFα) and cytotoxic ($CD8^+$) T cells to specific cell-surface receptors; in the latter case, Fas ligand (on the T cell) binds to the target cell's Fas receptor (which may also be referred to as CD95 or APO-1).

Inhibition of apoptosis is seen, for example, in B lymphocytes latently infected with EPSTEIN–BARR VIRUS; in these B cells apoptosis is inhibited by small EBV-encoded RNAs: see EBER.

Fluorophore-labeled probes, which label *active* caspases *in vivo* ('activity-based probes'), allowed researchers to monitor

the kinetics of apoptosis in live mice [Nature Med (2009) 15: 967–973].

Ongoing study of apoptosis has enabled the development of pro-apoptotic receptor agonists that may be used to promote apoptosis in cancer cells [Nature Rev Drug Discovery (2008) 7:1001–1012].

(See also ALTERNATIVE SPLICING (gene *bcl-x*), BARNASE, BINASE, CISPLATIN, MDA-7/IL-24, MYB, MYC and SURVIVIN.)

APSR (amplicon primer site restriction) In PCR, a method for preventing the contamination of a reaction mixture with amplicons from previous assays. In APSR, *all* the assays are conducted with primers whose 5′ ends carry a recognition site for a (type IIS) RESTRICTION ENDONUCLEASE which cleaves both strands 3′ of its binding site. In a reaction mixture, the (added) restriction enzyme will cut any carry-over amplicons but will not cleave the *template* DNA (unless, by chance, it contains the given recognition site); the restriction enzyme is inactivated during PCR temperature cycling.

(See also AMPLICON INACTIVATION.)

APT paper See SOUTHERN BLOTTING.

aptabody A construct, made from multiple RNA APTAMERS, which can function as a protein. The prototype aptabody was a tetravalent construct that was able to mimic an antibody by simultaneously binding two molecules of the *Drosophila* B52 protein and two molecules of streptavidin. The performance of the aptabody was reported to rival that of a monoclonal antibody against the B52 protein [Nucleic Acids Res (2009) doi: 10.1093/nar/gkp243].

aptamer (1) Any of a large number of synthetic oligonucleotides that can adopt (at least one) three-dimensional structure and bind, with high specificity, to a given ligand; both RNA and DNA aptamers can be synthesized.

(See also APTABODY and INTRAMER.)

Selection of an RNA aptamer for a given target molecule can be achieved by SELEX (systematic evolution of ligands by exponential enrichment). Briefly, in SELEX, the immobilized target ligand (for example, a protein) is exposed to a large and diverse population of oligonucleotides synthesized with random sequences. After removal of unbound oligos, the relatively few bound oligos are eluted and then converted to cDNAs. The cDNAs are amplified by PCR using primers that incorporate a promoter; the amplicons are transcribed, and the transcripts are used in another round of selection with the target molecule. This cycle is repeated, target–RNA binding becoming more specific at each round; the range and affinity of the selected aptamers can be determined e.g. by regulating buffer conditions at the binding stage. (A principle similar to this is found in PHAGE DISPLAY.)

In general, SELEX is useful for analyzing protein–nucleic acid binding and e.g. interactions between RNA and various low-molecular-weight molecules.

[Combining SELEX and yeast 3-hybrid system for *in vivo* selection and classification of aptamers: RNA (2007) 13(4): 614–622.]

Uses/applications of aptamers

The high-level binding specificity of the aptamers, and their ability to be linked to other molecules, has suggested various uses.

An aptamer–shRNA fusion transcript was used to regulate gene expression within mammalian cells. The activity of the shRNA moiety was controlled by an interaction between the aptamer and its ligand, theophylline (1,3-dimethylxanthine), the latter being supplied exogenously.

An aptamer – known to bind to prostate tumor cells – was conjugated to an siRNA via a streptavidin bridge; on addition to cells, this conjugate was internalized, and the siRNA was able to inhibit gene expression as efficiently as when it was transfected by a lipid-based procedure.

[Magnetically mediated selection of DNA aptamers: BioTechniques (2007) 43(3):344–353.]

[Cellulose-binding aptamers: Nucleic Acids Res (2007) 35 (19):6378–6388.]

[An *in silico* directed evolution study of DNA aptamers that bind allophycocyanin: Nucleic Acids Res (2009) 37(1):e6.]

[*In silico* selection of (RNA) aptamers: Nucleic Acids Res (2009) doi: 10.1093/nar/gkp408.]

A 58-nt aptamer sequence (which was developed by the use of SELEX) can – following transcription – adopt *two* distinct structures with different functions; both of the structures are required (jointly) to inhibit a particular receptor. These two structures cannot be interconverted (by unfolding and then re-folding) apparently owing to their inherent structural stability [Nucleic Acids Res (2009) doi:10.1093/nar/gkp284].

Using a SELEX-based method, five aptamers were selected for their high specificity towards ligands at the surface of the (pathogenic) Gram-positive bacterium *Staphylococcus aureus*– each of the five aptamers recognizing a different molecular target. When used in combination, this panel of five aptamers constitutes a system for the detection and identification of *S. aureus* [Nucleic Acids Res (2009) doi: 10.1093/nar/gkp489].

(See also APTAZYME and PROXIMITY LIGATION ASSAY.)

(2) A *natural* sequence within a RIBOSWITCH.

aptazyme (*DNA technol.*) An APTAMER–RIBOZYME construct in which the aptamer – on binding a certain ligand – typically undergoes a conformational change resulting in modification/regulation of the ribozyme.

In one application, ligand-binding by the aptamer promotes the ability of the ribozyme moiety to polymerize ribonucleotides – using another copy of the ribozyme as template. Thus, in the reaction mixture, the ribozymes synthesize copies of each other, and exponential amplification of the RNA occurs in the presence (although not in the absence) of the particular ligand. As the rate of increase in copies of RNA is governed by the ligand's *concentration*, the concentration of the ligand in a given sample can be assessed from the increase in RNA. Thus, systems can be set up in which an aptamer, in responding to a particular ligand, provides a sensitive assay for that

ligand. The ligand may be any of a variety of small molecules or proteins – e.g. those relevant in medical or environmental investigations. [Nature Biotechnol (2009) 27:288–292.]

One study has examined the performance of an engineered drug-sensing aptazyme made from a theophylline-responsive aptamer fused to a HAMMERHEAD RIBOZYME [RNA (2009) 15(1):76–84].

apurinic Lacking a purine residue: see AP SITE.

apyrase A nucleotide-degrading enzyme (EC 3.6.1.5) with various applications in technology (see e.g. PYROSEQUENCING).

apyrimidinic Lacking a pyrimidine residue: see AP SITE.

***araBAD* operon** See OPERON.

Arabidopsis thaliana A small cruciferous plant commonly used in plant genetics because of its simple genome and its short generation time.

[Genome of *A. thaliana*: Nature (2000) 408:791–826.]

(See also FLORAL DIP METHOD.)

araC See OPERON.

Aranesp® See BIOPHARMACEUTICAL (table).

arbitrarily primed PCR See AP-PCR.

arbitrary degenerate primer (AD primer) See TAIL-PCR.

arboviruses Arthropod-borne viruses: a (non-taxonomic) group which includes those viruses that are able to replicate within a vertebrate host *and* in arthropods (such as mosquitoes and ticks). Many of the arboviruses can cause disease in humans or animals; they include bluetongue virus (genus *Orbivirus*), Lassa fever virus (genus *Arenavirus*), West Nile virus (genus *Flavivirus*) and yellow fever virus (genus *Flavivirus*).

Arboviruses that infect mosquitoes can cause damage to the *mosquito* host – damage which may be limited by the insect's antiviral response: RNA INTERFERENCE (q.v.). An experiment to study the role of RNAi in protecting the mosquito used a recombinant SINDBIS VIRUS that expressed a protein (the B2 protein of Flock House virus) which inhibits RNAi. In this experiment it was found that expression of the B2 protein in mosquito cells inhibits RNAi. The recombinant Sindbis virus, inoculated intrathoracically, was lethal in different mosquito species, and the recombinant virus caused mortality in *Aedes aegypti* in a dose-dependent manner [BMC Microbiol (2009) 9:49].

ARCA See ANTI-REVERSE CAP ANALOG.

Archaea One of the two domains of prokaryotic organisms, the other being BACTERIA. Organisms in the two domains differ e.g. in 16S rRNA sequences, composition of cell wall macromolecules, composition of cytoplasmic membrane lipids and flagellar structure. The general features of gene expression in the two domains are also dissimilar.

(See also PROKARYOTE.)

archaean (*syn*. archaeon) An organism within the (prokaryotic) domain ARCHAEA.

Archaebacteria A now-obsolete kingdom of prokaryotes; the organisms formerly placed in this taxon are currently classified in the domain ARCHAEA.

ArchaeMaxx™ A polymerase-enhancing factor, marketed by Stratagene (La Jolla CA), which was designed to overcome the so-called DUTP POISONING effect.

This factor is used e.g. in association with the *PfuTurbo*® and Herculase® DNA polymerases (also marketed by Stratagene).

[Example of use: PLoS Biol (2006) 4(3):e73.]

archaeon See the entry ARCHAEAN.

ARES™ See PROBE LABELING.

Argonaute Any of a family of proteins which occur in higher eukaryotes and which are involved e.g. as effector molecules in the gene-silencing activities of certain small RNAs (see e.g. MICRORNAS and RNA INTERFERENCE).

Two subfamilies of Argonaute proteins are recognized. The proteins of the Ago subfamily are ubiquitous, being involved e.g. in post-transcriptional gene silencing (by miRNAs and siRNAs) in which mRNAs may be either physically degraded or functionally repressed. (See also BIM.) Proteins of the Piwi subfamily (see PIWI PROTEIN) apparently occur primarily in germ line cells – in which they act in conjunction with small RNAs (piRNAs) e.g. to silence TRANSPOSABLE ELEMENTS.

Various species are known to have several (or many) types of Argonaute protein, and, in a given species, these proteins may carry out different functions. Some Argonaute proteins have endonucleolytic activity, and these, in association with miRNAs or siRNAs, are able to cleave specific mRNAs; this cleavage is reported to occur when there is perfect complementarity between the small RNA and its target sequence on an mRNA. In human cells, only the Ago2 was reported to have nucleolytic capability [Genome Biol (2008) 9(2):210]. Both Ago1 and Ago2 apparently have nucleolytic capability in the cells of *Drosophila* – in which miRNA-directed cleavage of mRNAs appears to involve Ago 1, while Ago 2 works mainly in association with siRNAs.

Other types of Argonaute protein may be involved in the repression of translation when miRNAs bind to imperfectly complementary target sites.

Ago proteins may also be involved in *transcriptional gene silencing* and *transcriptional gene activation* (see SIRNA).

(See also ARNA (sense 2).)

ARMS Amplification-refractory mutation system: a procedure used e.g. for demonstrating or detecting a point mutation *at a specific site* in DNA whose wild-type (non-mutant) sequence is known.

Essentially, use is made of a primer in which the 3′-terminal nucleotide is complementary to the given *mutant* base at the specific site. After hybridization, extension of the primer by a polymerase signals the presence of the mutation at that site, while the absence of extension indicates the presence of a wild-type (or other) nucleotide.

armyworm The insect *Spodoptera frugiperda*. Cell cultures of this organism are used e.g. for the synthesis of recombinant proteins in BACULOVIRUS EXPRESSION SYSTEMS.

(See also SF9 CELLS.)

aRNA (1) Antisense RNA – see the entry MESSAGEAMP ARNA AMPLIFICATION KIT.

(2) Aberrant RNA: a type of RNA which is produced in cells (under certain conditions) and which can be converted to an effector molecule by modification involving the activity of an RNA-dependent RNA polymerase.

In the fungus *Neurospora crassa*, an aRNA that is induced by damage to DNA acts as a precursor to a small interfering RNA (∼20 nt long) referred to as qiRNA. The production of qiRNA (so named because of its interaction with Argonaute protein QDE-2) depends on the activity of an RNA-dependent RNA polymerase – and also requires a helicase and the Dicer enzyme [Nature (2009) 459:274–277].

Note. aDNA is used to refer to *ancient* DNA.

array A shortened version of MICROARRAY – but also used to refer to other oligonucleotide- or tissue-based arrangements etc. with analogous or distinct uses.

ARS Autonomously replicating sequence: a genomic sequence which, if isolated and linked to a non-replicative fragment of DNA, promotes independent (extrachromosomal) replication of that fragment within the cell.

ARSs were first identified in *Saccharomyces cerevisiae*, an organism which contains, on average, one ARS in every ∼40 kb of genomic DNA. ARSs also occur in at least some yeast plasmids.

It appears that some ARSs are active chromosomal origins while others are *silent origins;* some of the silent origins (and some active ones) may function as transcription silencers.

[Genome-wide hierarchy of replication origin usage in the yeast *Saccharomyces cerevisiae*: PLoS Genetics (2006) 2(9): e141.]

Factors reported to contribute to the efficient replication of ARS-containing plasmids in yeast cells include (i) the CEN (centromere) element and (ii) minichromosome maintenance protein 1 (Mcm1).

The circular dsDNA genome of human papillomavirus type 16 can replicate stably in *S. cerevisiae* independently of ARS or CEN; sequences in the viral DNA apparently substitute for both ARS and CEN [J Virol (2005) 79(10):5933–5942].

ARSs were also reported in species of the ARCHAEA.

(See also YEAST ARTIFICIAL CHROMOSOME.)

Artemis A DNA sequence viewer and annotation tool (Sanger Institute) used for displaying features of a given sequence of nucleotides and the results of analyses of that sequence (and of its six-reading-frame translation).

Artemis nuclease A multifunctional enzyme involved e.g. in a DNA repair mechanism – *non-homologous DNA end-joining*, NHEJ – used e.g. for repair of double-stranded breaks caused by ionizing radiation; it trims the ends of a double-stranded break, making them suitable for subsequent ligation by other components of the NHEJ system [see e.g. Nucleic Acids Res (2008) 36(10):3354–3365].

arthropod-borne viruses See the entry ARBOVIRUSES.

artificial *trans*-encoded sRNAs See the entry SRNAS.

ascospores See SACCHAROMYCES.

ascus See SACCHAROMYCES.

aseptic technique Measures which avoid the contamination of cultures, sterile media etc. – and/or contamination of persons, animals or plants – by microorganisms which are present in the environment (e.g. in the air) or which may be associated with particular source(s).

In this approach, the vessels used for media etc. must be sterile before use (e.g. pre-sterilized Petri dishes), and sterile material should not be exposed to any non-sterile conditions before use (see also STERILIZATION).

The working surfaces of forceps and other types of metal instrument (such as bacteriological loops etc.) are sterilized by 'flaming' before use, and the rims of bottles etc. used for dispensing sterile (non-flammable) materials are also flamed.

Benches are regularly treated with disinfectants and/or with ULTRAVIOLET RADIATION (UVR). The so-called 'germicidal' lamps, which may emit UVR at ∼254 nm, are used e.g. for the disinfection of air and exposed surfaces in enclosed areas. In general, UVR has rather poor powers of penetration, and its effects on microorganisms may be reversible by certain DNA repair processes (see e.g. UVRABC-MEDIATED REPAIR).

Some procedures, e.g. handling specimens likely to contain certain pathogens (such as *Mycobacterium tuberculosis*, or certain highly hazardous viruses such as Ebola virus or Lassa fever virus), are carried out in a SAFETY CABINET.

Asian flu See the entry INFLUENZAVIRUS.

ASLV vectors Avian sarcoma and leukosis virus-based vectors – see e.g. GENE THERAPY (*Viral vectors*).

ASP APOBEC-1-stimulating protein: see RNA EDITING.

aspart See INSULIN ASPART.

Aspergillus A genus of (mycelial) fungi (class Hyphomycetes) which are common in nature. *A. fumigatus* is pathogenic (see also AFLATOXINS), while many species are saprotrophic.

[*Aspergillus* genome database: Nucleic Acids Res (2009) doi: 10.1093/nar/gkp751.]

(See also INSERTIONAL MUTAGENESIS.)

ASR/GMP oligonucleotides Primers that comply with specifications of the FDA (Food and Drug Administration) in the USA and which are used (in restricted circumstances, and under stated conditions) for specific laboratory tests designed to provide analytical results.

assembly of DNA (*in vitro*) See the entry DNA CUTTING AND ASSEMBLY.

assisted reproductive technology See e.g. SCNT (for cloning of domestic animals) and IVF (in humans).

association studies See e.g. COPY NUMBER VARIANT and SNP GENOTYPING.

asymmetric PCR A form of PCR in which the concentration of *one* of the primers is much lower than that of the other (e.g. a ratio of 1:50); during temperature cycling, this primer will be quickly used up – so that only one strand of the target sequence will be significantly amplified.

Uses of asymmetric PCR include the preparation of probes and the preparation of single-stranded DNA for sequencing.

ssDNA products from PCR can also be obtained in a different way. One of the two types of primer can be labeled with BIOTIN and the reaction carried out with both primers in their

normal concentrations. When the reaction finishes, STREPTAVIDIN is added; this binds only to the biotin-labeled strands. Subsequent gel electrophoresis (in a denaturing gel) separates the two types of strand: the mobility of a streptavidin-bound strand is much lower. In this approach the biotinylated primer gives rise to the strand which is *not* required.

AT type See the entry BASE RATIO.

ATMS *p*-Aminophenyltrimethoxysilane: a reagent used for covalently binding DNA probes to a solid support when preparing a MICROARRAY. (In an earlier procedure, DNA was bound *non*-covalently to glass slides by the reagent poly-L-lysine.) [Method: Nucleic Acids Res (2001) 29:e107.]

(See also DENDRICHIP.)

atomic force microscopy (AFM; or scanning force microscopy) A method for imaging *surfaces*, including those of molecules and of (living) cells, in e.g. air or liquid, at nanometer-scale resolution. Essentially, the object's surface is scanned in a raster pattern with a fine probe located underneath a traveling cantilever; a laser, reflected from the cantilever – and thus providing information on the movements of the cantilever – is detected by a photodiode system, and the incoming signals are converted, by computer, into a surface profile.

Atomic force microscopy was used e.g. to study chromatin structure and nucleosome remodeling [Methods (2007) 41(3): 333–341]; to analyze repetitive α-satellite DNA [Eur Biophys J (2007) 37(1):81–93]; and for the characterization of an antibody scFv [Nanomedicine (2008) 4(1):1–7].

Atomic force microscopy has been used for direct visualization of G-quadruplexes in DNA [Nucleic Acids Res (2009) doi: 10.1093/nar/gkp679].

atsRNAs Artificial *trans*-encoded sRNAs: see the entry SRNAS.

***att* sites** Sites involved e.g. in SITE-SPECIFIC RECOMBINATION that occurs when the PHAGE LAMBDA genome integrates into a bacterial chromosome.

Lambda *att* sites are used in commercial DNA technology systems: see e.g. the GATEWAY SITE-SPECIFIC RECOMBINATION SYSTEM, MULTISITE GATEWAY TECHNOLOGY, BP CLONASE and LR CLONASE.

(See also *att*Tn*7* in the entry TN7.)

attaching and effacing lesion See PATHOGENICITY ISLAND.

***att*B, *att*P** See the entry PHAGE LAMBDA. (See also ATT SITES and GATEWAY SITE-SPECIFIC RECOMBINATION SYSTEM.)

attenuator control See OPERON.

***att*L, *att*R** See the entry PHAGE LAMBDA. (See also ATT SITES AND GATEWAY SITE-SPECIFIC RECOMBINATION SYSTEM.)

atto- A prefix meaning 10^{-18}.

AttoPhos™ A reagent (Promega, Madison WI) which can be cleaved (by ALKALINE PHOSPHATASE) to yield a fluorophore.

Attophos™ has been used e.g. in studies on the staging of trypanosomiasis [PLoS Negl Trop Dis (2009) 3(6):e459] and in immunoblotting in studies on prion proteins [Mol Biol Cell (2009) 20(1):233–244].

att*Tn*7 See the entry TN7.

Aubergine A member of the Piwi subfamily of ARGONAUTE proteins which is found in *Drosophila*. It is reported to be essential e.g. for germ cell formation.

(See also PIWI PROTEIN.)

Augmentin A combined formulation of the β-lactam antibiotics clavulanic acid (see β-LACTAMASES) and amoxycillin; it is used e.g. for the treatment of urinary tract infections.

autoactivation (*syn*. self-activation) (two-hybrid systems) Activation of a reporter system without prior interaction between the bait and prey proteins.

autocatalytic aptazyme See APTAZYME.

autocatalytic splicing See SPLICING.

autoclave An apparatus within which objects and/or materials are sterilized by saturated (air-free) steam under pressure; the conditions in a working autoclave are commonly within the range 115°C (∼69 kPa; 10 lb/inch^2) to 134°C (∼207 kPa; 30 lb/inch^2).

STERILIZATION in an autoclave is carried out e.g. when preparing certain types of media. Heat-labile constituents of a medium (e.g. a solution of an antibiotic) may be membrane-filtered before being added to a sterile (autoclaved) medium.

Some steam-impermeable items that cannot be sterilized by autoclaving may be sterilized in a hot-air oven at 160–170°C for ∼1 hour.

autoclave tape A paper strip (usually self-adhesive) which is included with the objects being sterilized in an autoclave; it exhibits a visible change (e.g. in color) when it is subjected to appropriate sterilizing conditions, and can therefore act as a check on the correct operation of the autoclave.

***Autographa californica* NPV** See NUCLEAR POLYHEDROSIS VIRUSES.

autoinducer (in quorum sensing) See QUORUM SENSING.

automated sequencing (of DNA) A method used for rapidly sequencing DNA fragments of up to ∼800 nt. Essentially, the process involves conventional chain-termination (i.e. Sanger) sequencing (see DIDEOXY METHOD) with fluorophore-labeled ddNTPs – each type of ddNTP (A, G, C, T) being labeled with a fluorophor that emits a distinctive color on excitation. The sequencing products are separated by polyacrylamide gel electrophoresis and the bands of products are scanned by a laser; the positions of individual nucleotides, identified by the color of the fluorescence, are recorded automatically.

(See also DNA SEQUENCING AND PYROSEQUENCING.)

autonomously replicating sequence See ARS.

autoplast A PROTOPLAST OR SPHEROPLAST which develops as a result of activity of an organism's own autolytic enzymes.

autoradiography A procedure in which a radioactive source is detected or quantitated by its effect on a photographic film; a film is exposed to the radioactive source, in the dark, and for an appropriate period of time, and is subsequently processed.

Autoradiography is used e.g. for investigating intracellular processes (radioactive isotopes being incorporated into biomolecules), and is also used for detecting bands of products (fragments of DNA), following gel electrophoresis, by means of radioactively labeled probes (see PROBE LABELING).

In general, optimal resolution may require the use of those isotopes which have relatively low-energy emission (such as

tritium, ^{3}H) rather than those (such as ^{32}P) which have high-energy emission.

autosomal dominant disorder Refers to a genetic disorder in which the phenotypic manifestation arising from expression of an abnormal autosomic allele occurs in the presence of the corresponding normal allele – that is, the influence of the abnormal allele overrides that of the normal allele. This type of disorder tends to exhibit a so-called *vertical* pattern of transmission from one generation to the next; in such cases, an abnormal trait is more likely (than in an autosomal recessive condition) to affect each successive generation.

Both males and females can be affected – and, unlike the situation in X-linked dominant disorders, father-to-son transmission can occur.

Examples of this disorder: AXENFELD–RIEGER SYNDROME and PEUTZ–JEGHERS SYNDROME (and see also CHARCOT–MARIE–TOOTH DISEASE).

(cf. AUTOSOMAL RECESSIVE DISORDER and X-LINKED DISORDER.)

autosomal recessive disorder Refers to a genetic disorder in which manifestation of the abnormal phenotype is exhibited when an abnormal allele is not accompanied by the presence of the corresponding normal, wild-type allele; heterozygous individuals with one normal allele do not usually exhibit the abnormal phenotype.

Both males and females can be affected.

Only when mating occurs between two (homozygously) affected individuals will the trait necessarily appear in all the offspring; mating between two heterozygous individuals, or between one heterozygous and one normal individual, tends to spare some of the offspring, so that an autosomal recessive disorder may miss generation(s) and is said to exhibit a *horizontal* mode of transmission.

(cf. AUTOSOMAL DOMINANT DISORDER and X-LINKED DISORDER.)

autosome Any chromosome other than a HETEROSOME.

autotransporter See the entry OMPT GENE.

auxins Phytohormones (plant hormones) which promote stem elongation and other aspects of plant development; the auxins are derivatives of tryptophan. Indole 3-acetic acid (IAA; also called 'auxin' or 'heteroauxin') is a major auxin; it is synthesized from the precursor indole 3-acetonitrile (IAN).

Abnormally high levels of auxins (*hyperauxiny*) are found in some plant diseases (e.g. CROWN GALL).

auxotrophic mutant Any microorganism which, as a result of a mutation, is unable to synthesize an essential nutrient and which therefore can grow only if provided with an exogenous source of that nutrient. (An organism which does *not* contain such a mutation, and which can synthesize all of its essential nutrients, is called a *prototroph*.)

One example of an auxotrophic mutant is mentioned in the entry AMES TEST.

Isolation of auxotrophic bacteria

The usual selective procedures are not suitable for isolating auxotrophic bacteria from a mixture of prototrophs and auxotrophs: a medium that supports the growth of auxotrophs will also permit the growth of the corresponding prototrophs.

The *limited enrichment* method uses an agar-based *minimal medium* enriched with *small* amounts of nutrients. A minimal medium is one which supports the growth of prototrophs but – because it lacks one or more essential nutrients – does not support the growth of auxotrophs. Any colony of auxotrophic cells will quickly exhaust the small amount of nutrients in its vicinity – so that the colony will remain small; however, the unrestricted growth of prototrophs on this medium means that colonies of prototrophs will be larger than those of the auxotrophs. Hence, the small colonies indicate presumptive auxotrophs.

The *delayed enrichment* technique employs an agar-based minimal medium for initial growth, so that prototrophs (only) form colonies in the initial incubation. Complete medium is then poured onto the plate and allowed to set; the nutrients in this medium diffuse into the minimal medium below, allowing the growth of auxotrophs. Again, small colonies indicate presumptive auxotrophs.

For (penicillin-sensitive) bacteria, auxotrophic mutants can be isolated by virtue of their inability to grow in a (penicillin-containing) minimal medium; penicillin is an antibiotic that acts only on *growing* cells. In this technique, a well-washed population of bacteria (that includes auxotrophs) is exposed to penicillin in a minimal medium; the prototrophs (which are able to grow) are killed by the penicillin. The remaining cells are washed and re-plated on a complete medium in order to recover any auxotrophs that may be present. It's important to note that, if the auxotrophs had developed through an *in vitro* process of mutagenization, the cells must be allowed to grow for several generations in complete medium prior to exposure to penicillin; this is because newly mutated cells will contain a full complement of (prototrophic) enzymes – auxotrophy developing only after several rounds of cell division (during which the prototrophic enzymes are 'diluted out'). A failure to observe this requirement would cause the death of any newly mutagenized auxotrophs on exposure to penicillin. A further requirement in this method is that only a low concentration of cells be used; the reason for this is that auxotrophs should not be allowed to grow on nutrients released by lysed prototrophs as this would render them susceptible to lysis by penicillin. For this reason, STREPTOZOTOCIN (q.v.) may be employed in place of penicillin.

Auxotrophs may also be isolated by REPLICA PLATING.

avian erythroblastosis virus See the entry ERB.

avirulence gene See GENE-FOR-GENE CONCEPT.

Axenfeld–Rieger syndrome An autosomal dominant disorder involving eye defects and certain systemic abnormalities. The syndrome has been associated with mutations in the *PITX2* gene – which encodes a transcription factor – or mutation in other genes (e.g. *FOXC1*, *PAX6*). [A novel *PITX2* mutation: Mol Vision (2008) 14:2205–2210.]

In some patients the disorder is reported to involve aberrant splicing of pre-mRNA; it was suggested that variability in the

extent of the splicing fault may be reflected in the variability of phenotypic manifestations [BMC Med Genet (2006) 7:59].

5-aza-2′-deoxycytidine (decitabine) A nucleoside analog used for studying DEMETHYLATION and as a therapeutic agent in the treatment of e.g. myelodysplastic syndrome and myeloid leukemia. In cells, 5-aza-2′-deoxycytidine is phosphorylated and incorporated into DNA; it then inhibits methyltransferase activity – for example, it has been shown to reactivate tumor suppressor genes silenced by methylation of promoter DNA. While effective against leukemias, this agent is generally less effective against solid tumors. Studies on MELANOMA cells suggest that the activity against this type of cancer could be made more effective given a better knowledge of the epigenetic and genetic background of each individual tumor [PLoS ONE (2009) 4(2):e4563]. (These studies have also suggested that HDAC and proteasomal inhibitors might act synergistically with the methyltransferase inhibitor in some patients.)

Studies to determine optimal therapeutic dosing of 5-aza-2′-deoxycytidine have been carried out in cultured cells [BMC Cancer (2008) 8:128], and 5-aza-2′-deoxycytidine (combined with various HDACs) has been studied in Ewing's sarcoma cells [Cancer Cell Int (2008) 8:16]. The agent has also been used in studies on promoter methylation in breast tumor cell lines [BMC Cancer (2009) 9:80] and for the identification of hypermethylated genes [BMC Med Genomics (2009) 2:11].

5-aza-2dC The nucleoside analog 5-AZA-2′-DEOXYCYTIDINE.

5-aza-CdR The nucleoside analog 5-AZA-2′-DEOXYCYTIDINE.

5-azacytidine A nucleoside analog that is used e.g. for inhibiting DNA methyltransferases; its activity is similar to that of 5-AZA-2′-DEOXYCYTIDINE (q.v.).

azaserine (*O*-diazoacetyl-L-serine) An agent with antimicrobial and antitumor activity produced by *Streptomyces* sp (a Gram-positive bacterium). Azaserine inhibits the activity of certain enzymes, including phosphoribosylformylglycinamidine synthetase – thus inhibiting biosynthesis of purines and, hence, nucleotides.

(cf. DON; see also HADACIDIN.)

AZT See ZIDOVUDINE.