Multidrug resistance: An emerging threat

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Multidrug resistance (MDR) has been the main cause of failure of cancer chemotherapy where it is defined as the tendency of tumour cells to exhibit simultaneous resistance to unrelated chemotherapeutic agents. MDR has been mainly associated with the overexpression of an ATP binding cassette (ABC) protein, P-glycoprotein. Research in the past decade has revealed that the MDR phenomenon is not restricted to mammalian cells but rather occurs throughout the evolutionary scale. Thus over hundred ABC proteins have been characterized in mammals, bacteria and yeast. This review briefly describes the advancement in this field and identifies the problems which have emerged due to MDR.

MULTIDRUG resistance, which is a major problem in medical and agricultural developments, is an emerging phenomenon observed in various organisms throughout the evolutionary scale. In agriculture, the control of resistance of plant pathogens towards natural plant defence toxins and other common fungicides, as well as the emergence of parasite-toxin resistant crops, are of major economic importance. In medicine, the problem of cancer is compounded by the acquisition of multidrug resistance (MDR) by human malignancies. MDR has been one of the principle causes of failure of cancer chemotherapy where it can be defined as the tendency of tumour cells in patients and cultured cells to exhibit simultaneous resistance to multiple chemically unrelated chemotherapeutic agents[1,2]. The elucidation of the mechanism by which tumour cells develop resistance to toxic effects of potent chemotherapeutic agents has revealed a great deal about the process of drug uptake, metabolism and extrusion. This has also provided basic insights into cellular process such as regulation of gene expression and gene amplification[1-3]. It has been shown that overexpression of certain ATP-binding cassette (ABC)-proteins in prokaryotes and eukaryotes is linked to drug resistance phenomenon[4]. The well characterized mammalian protein MDR1 (P-glycoprotein) is associated with the development of a drug-induced multidrug resistance phenotype in tumour cells[1-5]. Further, overexpression of *Ldpgp A* from *Leishmania* is responsible for methotrexate and heavy metal resistance, and *Plasmodium Pfmdr* has been implicated in chloroquine resistance in the malarial parasite[6-10]. Likewise, bacterial erythromycin resistance in *Staphylococcus* is caused by *MsrA* overexpression, and the ABC-protein *DrrAB* of *Streptomyces* appears to be daunomycin resistance determinant[11,12].

Drug resistance

In mammalian cells

Selective passage of specific molecules across membrane is the key to cell's survival which is achieved by specific membrane transporters. The importance of membrane transport is becoming even more apparent from genome sequencing projects where a majority (20–30%) of genes have been found to encode for membrane and particularly transport proteins[13]. It has been shown that there exists a limited number of transporter families where member proteins of a family are related to each other in sequence and in molecular mechanism and probably have a common evolutionary origin[11,12,14].

It is now clear that a major mechanism of MDR in mammalian cells involves the overproduction of a 170 kDa plasma membrane glycoprotein, P-glycoprotein[1,5,15]. This protein appears to cause MDR via an ATP-dependent drug efflux mechanism, which prevents the intracellular accumulation of drugs to an effective cytotoxic concentration[1-3]. P-glycoprotein is a member of super gene family of bacterial and eukaryotic transporter proteins (Table 1). The mammalian P-glycoproteins are encoded by small families of linked genes, two in humans, three in rodents. The human *MDR1* gene, the *mdr1* and *mdr3* genes of mice and the *pgp1* and *pgp2* of hamster encode related proteins which transport hydrophobic drugs[1,3,16].

Cloning and sequencing was a major step towards understanding the structure and function of P-glycoprotein. The sequence encoding P-glycoprotein revealed that it is a tandemly repeated molecule of about 1280 amino acids (~ 170 kD). Each half consisting of a large hydrophobic domain containing three pairs of putative membrane-spanning α-helices and a conserved hydrophilic cytoplasmic domain containing an ATP-binding site[1,17-20]. It has been proposed that the 12 transmembrane domains associate to form a pore or channel through which P-glycoprotein actively effluxes drugs[1,5]. *In vitro* mutagenesis of the putative ATP-binding sites suggests that both sites are required and these may functionally interact to affect drug efflux[1,21].

Although the mechanism of drug transport has not

been defined, it is thought that direct binding of the drug to P-glycoprotein could be one of the essential steps[1]. Extensive genetic manipulation involving deletion and insertion analyses of human MDRs has revealed that there are several coding regions which appear to have no effect on drug binding and specificity. However, there are several point mutations scattered throughout the gene which selectively alter drug specificity of the P-glycoprotein[21]. The drug specificity of MDR is a complex phenomenon which either requires a highly ordered structure or is affected by multiple independent parts of the protein molecule[22,23].

The ability of drugs and reversing agents to inhibit each other's binding to P-glycoprotein suggests that they compete for common binding site(s)[1] (Table 2). Thus, one mechanism of MDR reversal by chemosensitizers and non-toxic drug analogues may be explained on the basis of competition for drug binding, which results in a decrease in efflux rate and a higher intracellular level of toxic drugs in MDR cells[21]. The P-glycoprotein recognizes a diverse group of substrates and shows different cross reactivity profiles[1-3]. It is believed that a spontaneous mutation in P-glycoprotein gene, leading to altered drug specificity, may change the overall MDR profile[1,21-25].

That other mechanisms may also generate diversity in MDR phenotype has not been completely ruled out. In rodents, two different P-glycoproteins confer MDR

Table 1. Multidrug resistance pumps identified from microbes to man*

Organism	Proteins	Family	Function/substrate	Topology®
Prokaryotes				
E. coli	EmrE/MvrC	Major facilitator	Drug/H + transporters	
Staphylococcus	QacA	Major facilitator	Drug/H + transporters	12–14 TM helices
Staphylococcus	MsrA	Major facilitator	Drug/Antibiotics transporter	
B. subtilis	Bmr	Major facilitator	Tetra phenyl phosphonium	12–14 TM helices
Yeast (See Table 3)				
Protozoa				
P. falciparum	Pfmdr	ABC protein	Chloroquinine	12 TM helices
L. donovani	Ldmdr		Arsenite (?)	
Moulds				
C. elegans	Cepgp Ag	ABC protein	?	12 TM helices
Insects				
Drosophila	Mdr 49/50	ABC protein	?	12 TM helices
Plants				
Arabidopsis	Atpgp	ABC protein	?	12 TM helices
Mammals				
Hamster	Pgp1	ABC protein	Lipophilic drugs	12 TM helices
Mouse	Mdr1	ABC protein	Lipophilic drugs	12 TM helices
Man	Mdr1	ABC protein	Anticancer/lipophilic drugs	12 TM helices
Man	CFTR	ABC protein	Chloride channel	12 TM helices

*The table is compiled from refs 3, 11, 12, 53.
®Deduced from hydropathy analyses.

Table 2. Compounds which can interact with the multidrug resistance pump

Anticancer drugs	Other cytotoxic drugs	MDR-reversing agents	Cyclic and linear peptides
Daunorubicin	Colchicine	Verapamil	Gramicidin D
Doxorubicin	Emetine	Quinidine	Valinomycin
Mitoxanthrone	Ethidium bromide	Quinine	Yeast a-factor pheromone
Etopside	Puromycin	Cyclosporin A	*N*-acetyl–leucyl–leucyl–norleucine
Teniposide	Podophyllotoxin	Forskolin	
Vinblastine		Azidopine	
Vincristine			
Actinomycin D			
Mitomycin C			
Taxol			
Topotecan			
Many others			

and differential expression of these genes probably could alter the stoichiometry of the individual isoform in the cell membrane, resulting in differences in profile of transported drugs[26]. Furthermore MDR is a result of overexpression of P-glycoprotein gene which may be accompanied by the coexpression of very large stretches of flanking DNA. In Chinese hamster cell line, P-glycoprotein amplification has been shown to be over one mega base pair in size and at least six classes of genes have been found to be coamplified and overexpressed[3,27,28]. It is, therefore, possible that overexpression of such linked gene may modify the drug resistance profile. Differences in drug resistance profile may also be the result of differences in post-translational modification of P-glycoprotein molecules itself[3,26]. It has recently been found that P-glycoprotein is phosphorylated at both serine and threonine residues[5,26]. It has been speculated that the extent of change in phosphorylation may modulate P-glycoprotein mediated drug transport mechanism. However, this remains to be confirmed. Study of P-glycoprotein glycosylation suggests that carbohydrate molecules do not affect drug resistance[5]. However, their role as modulators of P-glycoprotein function cannot be precluded.

In bacterial cells

When antibiotics like penicillin were discovered, some fifty years ago, they were treated as miracle drugs of the century. This scene has suddenly changed. We are now confronted with new resistant types of bacteria. Once bacteria have learnt a particular strategy to circumvent the toxic effect of an antibiotic, they exchange the genetic information, without any species specificity, with other bacteria. As a result, now with every possible bacterial infection, resistance to antibiotic treatment is a common phenomenon. The cause of resistance is attributed to the amplification of bacterial MDR genes.

Most bacterial MDR come under major facilitators families (MFS) which include arabinose/H^+ symporter of *Escherichia coli* and glucose facilitator of eukaryotes[11,12]. The proteins of this family are similar to P-glycoprotein of eukaryotic cells but lack ATP binding domains and thus are not classified as ABC proteins. MFS have 12 transmembrane α-helical domains and use proton motive force as a source of energy. QacA is one of the first MDR proteins identified in bacteria. *Staphylococcus* acquires resistance to the quaternary ammonium compounds (*QacA*) used in antiseptics. QacA is a membrane pump which effluxes out several drugs in a proton motive force dependent manner[11,29]. *emrA* and *emrB* are the two genes coded by *E. coli* which confer resistance to uncouplers (CCCP) and other antimicrobial agents[11,30]. Interestingly, EmrB protein is homologous to QacA. EmrA, on the other hand, is homologous to proteins participating in the efflux of bacterial toxins and proteases. EmrA is homologous to HlyD (a component of *E. coli* hemolysin efflux pump) albeit to a lesser degree. The function of these proteins is to form a channel between the inner and outer membrane (Figure 1). In case of hemolysin pump, HlyB is the actual pump while HlyD and a porin (TolC) are needed to form a channel to allow the passage of the peptide outside the cell. Thus, the topological design of EmrA–EmrB could be the same as that of hemolysin pump[12] (Figure 1).

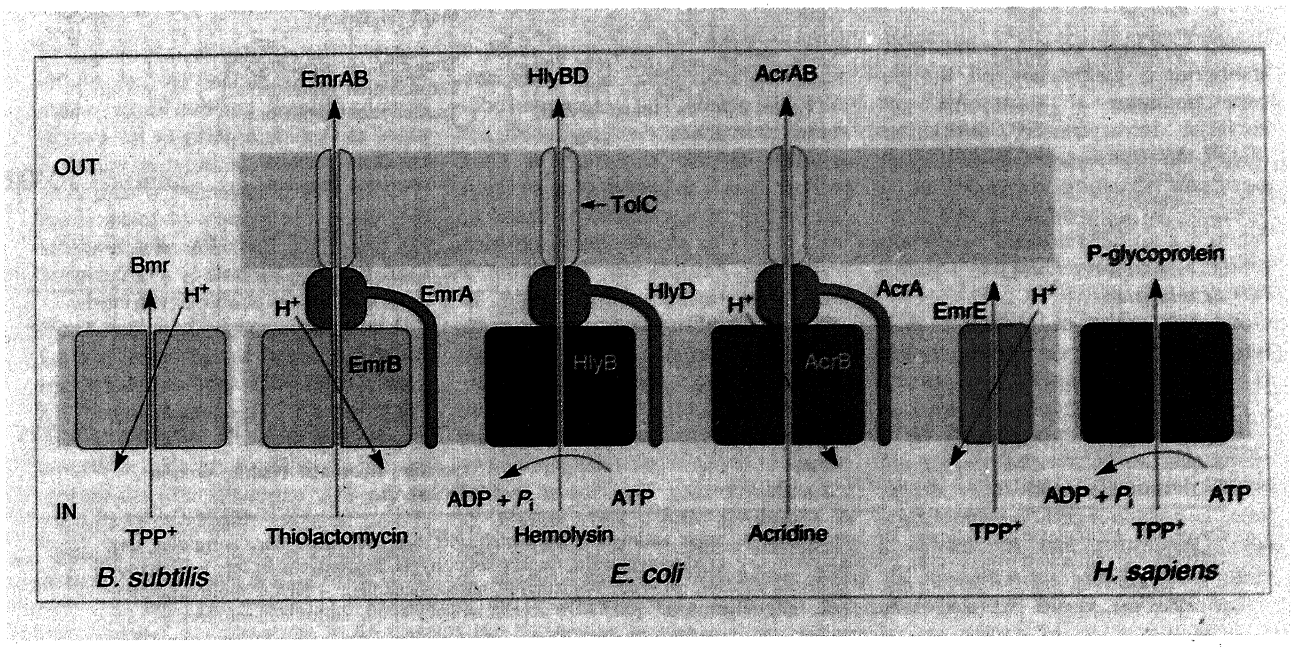

Figure 1. Topology of bacterial and human multidrug resistance. For comparison, the use of same colour indicates the homologous proteins in different MDR complexes. Reproduced from ref. 2 with permission.

In yeast cells

Multidrug resistance phenomenon is not restricted to mammalian or microbial cells. Host of genes homologous to MDR have been identified in yeasts during the past three decades. Yeast shares similarity in structural and functional organization with higher eukaryotes and is amenable to genetic manipulations and thus, serves as an excellent model for unravelling eukaryotic pathways of MDR. The studies involving MDR in yeasts have got further impetus since some yeast species are also pathogenic to plants and humans. Already about 25 genetic determinants associated with multidrug resistance (pleiotropic drug resistance, PDR in yeasts) have been characterized in *Saccharomyces cerevisiae*, *Schizosaccharomyces pombe* and *Candida albicans*[31,32]. The gene products encoded by these yeasts fall into three classes of proteins: ABC, MFS and transcription regulators (Table 3).

The *PDR5* gene was cloned as a multicopy plasmid borne DNA fragment capable of conferring pleiotropic drug resistance (PDR)[33,34]. The gene codes for a polypeptide of 1511 amino acid residues with calculated mol. wt of 170.4 kD. PDR5 protein is predicted to contain twelve 'integral' transmembrane spans gathered in two groups of six contiguous membrane spans. Each hydro-

Table 3. Yeast proteins of multidrug resistance family

Yeast	Protein	Substrates	Membrane topology/function
S. cerevisiae	PDR5/STS1/YDR1	cyh, chl, ery, amy, sts, flu, smm.	ABC membrane protein. (NBD-TM)2
S. cerevisiae	SNQ2	4-NQO, MNNG, flu, sts, tri.	ABC membrane protein. (NBD-TM)2
S. cerevisiae	STE6	Val	ABC membrane protein. (TM-NBD)2
S. cerevisiae	YCF1	Cd	ABC membrane protein. (TM-NBD)2
S. pombe	pmd1	lep, cyh, val	ABC membrane protein. (TM-NBD)2
C. albicans	CDR1	cyh, chl, mic, amy	ABC membrane protein. (NBD-TM)2
S. cerevisiae	ADP1	–	ABC membrane protein. (NBD-TM)
S. cerevisiae	YKL741	–	ABC membrane protein. (TM-NBD)
S. cerevisiae	MDL1	–	ABC membrane protein. (TM-NBD)
S. cerevisiae	MDL2	–	ABC membrane protein. (TM-NBD)
S. cerevisiae	Ssh1	–	ABC membrane protein. (TM-NBD)
S. cerevisiae	Ssh2	–	ABC membrane protein. (TM-NBD)
S. pombe	HMT1	heavy metals (Cd)	Vacuolar. (TM-NBD)
S. cerevisiae	ATM1	–	ABC membrane protein. (TM-NBD)
S. cerevisiae	ATR1/SNQ1	atr, 4-NQO	Major facilitator
S. cerevisiae	YCL069w	–	Major facilitator
S. cerevisiae	YCL023c		Major facilitator
S. cerevisiae	YCL070c		Major facilitator
S. cerevisiae	YKR105c		Major facilitator
S. cerevisiae	YKR106w		Major facilitator
C. albicans	Ben^r	ben, met	Major facilitator
C. maltosa	Cyh^r	cyh	Major facilitator
S. pombe	car1	aml	–
S. cerevisiae	PDR1	cyh, chl, oli, nys, ner, muc etc.	Transcription regulator
S. cerevisiae	PDR3	muc, chl, cyh, oli, tet, ner.	Transcription regulator
S. cerevisiae	yAP1/PDR4 SNQ3/PAR1	Cd, Zn, cyh, tre, smm, 4-NQO, phe, MNNG, nin	Transcription regulator
S. cerevisiae	CAD1/YAP2	Cd, Zn, phe	Transcription regulator
S. pombe	pap1	sts	Transcription regulator
S. cerevisiae	PDR7	cyh, smm	–
S. cerevisiae	PDR9	cyh, smm	Transcription regulator
S. cerevisiae	RPD1	cyh	Transcription regulator
S. cerevisiae	RPD3	cyh	Transcription regulator
S. cerevisiae	YGL022	cyh, smm	– –
S. cerevisiae	PDR6	cyh, bor, hygB	–
S. cerevisiae	PDR8	oli, smm	–
S. pombe	sts1	cyh, sts, caf, chl, divalent cation	–
S. cerevisiae	cpr	van	Soluble
S. cerevisiae	HOM3	bor	Soluble
S. cerevisiae	AMY1	amy	–
S. pombe	RIM-C	cyh	Soluble, ribosomal binding protein
S. cerevisiae	ZRC1	Zn, Cd	Transporter

Drugs are abbreviated as follows: atr, aminotriazole; amy, antimycin; aml, amiloride; ben, benomyl; bor, borrelidin; caf, caffeine; chl, chloramphenicol; cyh, cycloheximide; ery, erythromycin; flu, fluphenazine; hygB, hygromycin B; lep, leptomycin; mic, miconazole; muc, mucidin; nin, 1-nitroso-2-naphtol; MNNG, *N*-methyl-*N'*-nitrosoguanidine; 4-NQO, 4-nitroquinoline *N*-oxide; ner, neutral red; met, methotrexate; oli, oligomycin; phe, 1-10-phenanthroline; smm, sulfomethuron methyl; sts, staurosporine; tet, tetracycline; val, valinomycin; van, vanadate; tri, triaziquone; tre, trenimon. Other abbreviations are: NBD, nucleotide binding domain; TM, transmembrane region; ABC, ATP-binding cassette; (NBD-TM)2, NBD precedes TM and vice versa and has 2 halves. The table is compiled from refs 31, 41, 42.

phobic domain follows a hydrophilic region including a predicted ATP-binding cassette (ABC). Thus, PDR5 seems to have duplicated structure, consisting of two halves each composed of one hydrophilic and a hydrophobic domain[34,35] (Figure 2). The two similar ABC domains of PDR5 are conserved within a large super family of transport proteins[35].

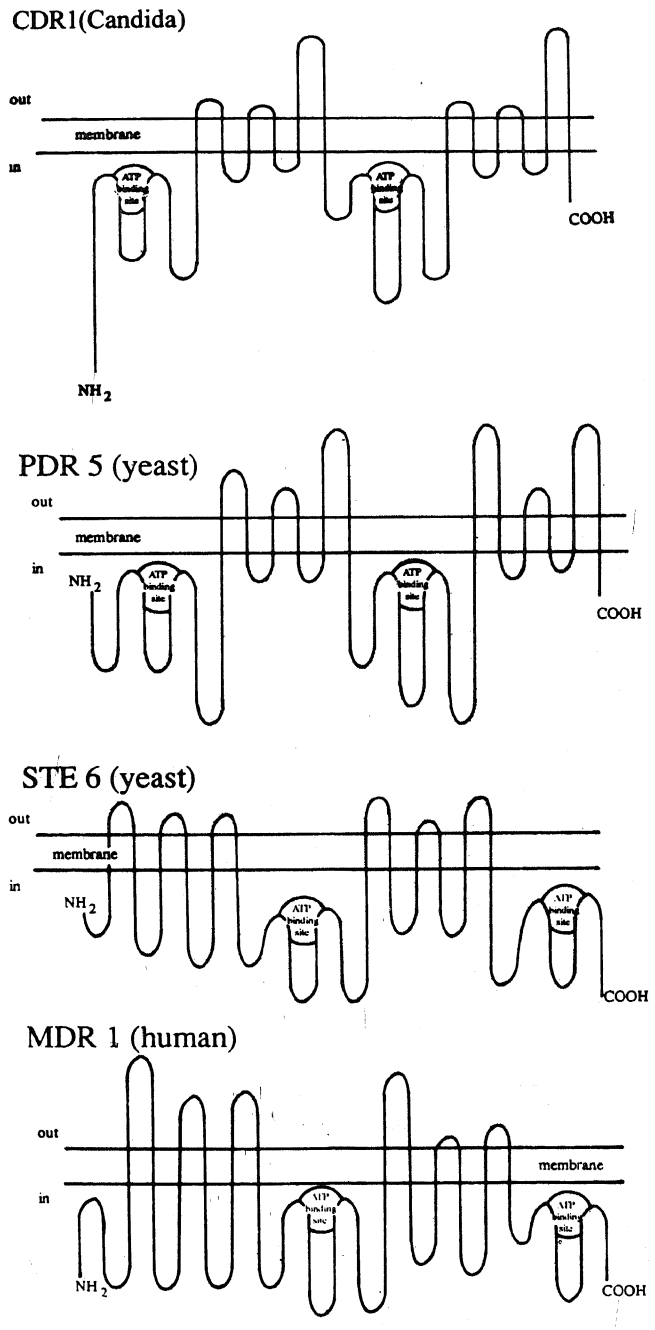

Figure 2. Predicted structure of the CDR1, PDR5, STE6 and MDR1 proteins. The CDR1 and PDR5 proteins are predicted to be composed of two repeated halves, each comprising of one hydrophilic domain followed by a hydrophobic domain. Two hydrophilic domains are cytoplasmic (IN) and each contains one ATP-binding site. The two hydrophobic domains are considered to be spanning the membrane. The sequence of domain inversion between CDR1 and STE6 can be seen.

A sequence alignment of entire protein in databanks revealed homology between PDR5 and other members of the ABC-transporters superfamily. The best comparison was obtained with yeast ADP1, pheromone transporter STE6, *Drosophila* white and brown eye pigment transporter, bacteria hemolysin secretion protein B, mouse MDR1, rat major histocompatibility complex Mtp1 and most importantly with cystic fibrosis protein CFTR in humans. The region of homology is mainly localized on ABC cassette[34] (Figure 2). Recent evidences for the modular structure of a four-domain ABC transporter have been provided by domain dissection analysis of the yeast STE6 transporter[36,37]. The two halves of the molecule were shown to be able to support, jointly, '**a**' factor transport activity[38].

A *PDR5* homologue *CDR1* has recently been cloned and characterized in a pathogenic yeast, *Candida albicans*, by functional complementation of a *PDR5* null mutant of *S. cerevisiae*. The nucleotide sequence of *CDR1* revealed that, like *PDR5*, it encodes a putative membrane pump belonging to ABC superfamily[39] (Figure 2). Fresh evidences from our group suggest that there are several homologues other than *CDR1* in *C. albicans* which display cross resistance pattern different from *CDR1* and *PDR5*[39,40]. Benomyl resistant (*Ben*^{r}) gene of *C. albicans* has also been shown to encode a putative membrane pump which belongs to MFS family[41]. The characterization of *CDR1*, *Ben*^{r} and identification of several other multidrug resistance genes from a pathogenic yeast could pave the way for tackling drug resistance in *Candida* and for the development of effective anti-*Candida* drugs. Recently, the field of drug resistance in pathogenic fungi has generated considerable interest because of spread of AIDS where *Candida* infections are most predominant.

A few ABC proteins have also been characterized in a fission yeast, *S. pombe*. HMT1 is a duplicated ABC protein associated with the vacuolar membrane and most similar to mammalian glycoprotein. Overexpression of the *HMT1* was correlated to enhanced heavy metal tolerance[42]. The *PMD1* encodes a half ABC protein (comprising of six transmembrane segments) homologous to *MDR1* and *STE6*. Overexpression of *PMD1* confers resistance to leptomycin B, cycloheximide and valinomycin[31]. HBA2, another ABC protein that confers resistance to brefeldin A and other drugs, has recently been identified in *S. pombe*[43].

The two pleiotropic drug resistance loci, *PDR1* and *PDR3*, were found to encode homologous transcription factors belonging to the family containing a 'Zinc 2 Cysteine 6' co-ordination complex in the DNA binding domain[32]. The *PDR1* gene product was shown to modulate

the expression of multidrug resistance genes, such as *PDR5* and *STE6*, and also affect the estradiol levels[32,44]. The fact that estrogen molecules are also substrates in the yeast *PDR* pathway, may provide a link between drug resistance and hormone tolerance[32]. The uncovering of regulatory elements, like *PDR1*, *PDR3*, etc. in yeast[45,46], might provide the basis for unravelling related circuits of control in human multidrug resistance.

Physiological role of P-glycoprotein

The availability of various sequences of P-glycoprotein genes of different species has allowed a comparison between different genes, both within a species and among different species. The comparison has shed some light on the evolution of P-glycoprotein and the organization of its gene. The similar organization of coding sequences and intervening sequences in different genes from the same species indicate that the internal duplication of the ancestral gene occurred prior to the formation of multigene family. The organization of homologous members of the multigene family in different mammalian species suggests that the formation of a multigene family preceded the divergence of species[3]. The evolutionary relation and conserved structure of P-glycoprotein leads to questions like: what is the physiological function of such proteins?

Expression of P-glycoprotein is cell and tissue-specific[2,47,48]. Therefore, the tissue distribution may help to identify the physiological role of P-glycoprotein as a transporter. But given the specific and complex pattern of expression, it has been difficult to imagine a single class of physiological substrate[2]. Therefore, it has been suggested that P-glycoprotein plays a diverse role in transport[23]. However, as of now there are only a few examples from higher eukaryotes where the physiological role of MDR proteins has been identified (Table 4).

Mechanisms of MDR

Multidrug transporter (ABC type) does not function as a simple transmembrane transport system which effluxes out drugs from cytoplasm to extracellular space. Most of the substrates of pump are hydrophobic and thus tend to partition in nonpolar environment in preference to aqueous phase. Indeed, the data also suggest that anticancer anthracyclines, rhodamine-123 are predominantly localized in the plasma membrane and intracellular membranous structures in addition to their targets. The spectrum of drugs handled by the transporter suggests that a simple model of substrate recognition may not be correct. At least one transporter of ABC family CFTR (cystic fibrosis transmembrane regulator) is also a Cl^- channel[49–51]. Therefore, a need to have a model of P-glycoprotein transporter which encompasses all the conflicting observations has been realized[52].

According to the most accepted model, drugs are removed by the transporter directly from the plasma membrane (lipid bilayer), thus, drugs are thrown out and are unable to reach the cytoplasm[2]. Conceptually, the multidrug transporter works as a 'hydrophobic vacuum cleaner' which removes drugs from the membrane. The mechanism of energy transduction during drug transport is, of course, not clear. The nucleotide-binding domains of P-glycoprotein and constitutive ATPase activity therein do suggest a role of nucleotide hydrolysis. Some evidences also suggest that the drug transporter could be an enzyme 'flippase' which would bind the drug from the inner leaflet and flips it to the outer leaflet from where the drug diffuses out to extracellular space or the pump could behave like a moving 'waterwheel' or 'escalator', which expels all membrane constituents of approximately similar size (molecular weight) and shape, with little substrate specificity. There is still no unifying model which could include all conflicting observations of drug transport[1,2,26] (Figure 3). But the recent suggestion of chloride channel activity associated with the multidrug transporter is consistent with the idea that the net positive charge (proton) accompanies drugs out of the cell and this may require an anion channel to maintain electric neutrality[2].

Future perspective

In the beginning a specific mechanism of antibiotic resistance was thought to be more important. Thus attempts were made to produce more effective antibiotics by modification of specific groups of antibiotic molecules in order to make them inert as potential substitute for commonly occurring antibiotic inactivating enzymes. However, the presence of more generalized mechanism of multidrug resistance has compelled the scientists to evaluate this strategy. As a result, several new drugs with new targets are in the pipeline and may hit the market in couple of years' time. Since these new drugs hit new targets it is hoped that bacteria will take still longer to learn to destroy them. There is also a need to obtain more knowledge about the substrate-binding process of these transporters. A possible approach would be to increase the spontaneous influx of drugs by making them sufficiently lipophilic so that efflux can be counter balanced by rapid influx. Indeed, it will be a major challenge for the pharmaceutical industry because some of the multidrug efflux systems seem to pump out almost any amphiphilic compound.

In plant pathogens, P-glycoprotein may be responsible for the secretion of fungal pathogenecity factors or toxic plant defence products playing a crucial role in plant–pathogen interaction. Understanding of the role of P-

glycoprotein in these processes would open new ways for indirect control of plant pathogens by interference with the plant–pathogen interaction. This particular area is still at its infancy. In this regard, recent cloning of a MDR homologue in *Arabidopsis thaliana* and identification of efflux pumps in pathogenic fungi of plants are interesting developments[53–55].

In mammalian cells, where numerous approaches to reverse or modify MDR are currently being investigated, two important problems need to be re-emphasized:

Table 4. Some of the ABC-proteins with known substrates

Species	Protein	Substrate	Function
Bacteria			
Salmonella typhimurium	Opp ABCDF	Oligopeptides	Import
Streptococcus pneumoniae	Ami ABCDEF	Oligopeptides	Import
Bacillus subtilis	Opp (Spo K)	Oligopeptides	Import
E. coli	Dpp	Dipeptides	Import
Bacillus subtilis	Dci A	Dipeptides	Import
S. typhimurium	His JQMP	Histidine	Import
E. coli	His JQMP	Histidine	Import
E. coli	Mal EFGK	Maltose	Import
S. typhimurium	Mal EFGK	Maltose	Import
Enterobacter aerogenes	Mal EFGK	Maltose	Import
E. coli	Ugp ABCE	Gly-3-Phosphate	Import
E. coli	Ara FGH	Arabinose	Import
E. coli	Rbs ACD	Ribose	Import
E. coli	Gln HPQ	Glutamine	Import
S. typhimurium	Pro U (VWX)	Glycine-betaine	Import
E. coli	Pro U (VWX)	Glycine-betaine	Import
E. coli	Liv HMGF (JK)	Leu-Ile-Val	Import
E. coli	Pst ABC	Phosphate	Import
Pseudomonas stutzeri	Nos DYF	Copper	Import
E. coli	Chl JD	Molybdenum	Import
E. coli	Cys PTWAM	Sulphate-thiosulphate	Import
E. coli	Btu CDE	Vit. B_{12}	Import
E. coli	Fhu BCD	Fe^{3+}-ferrichrome	Import
E. coli	Fec BCDE	Fe^{3+}-dicitrate	Import
S. marcens	Sfu ABC	Fe^{3+}	Import
Streptomyces fradiae	Tlr C	Tylosin	Export
Agrobacterium tumefaciens	Occ JQMP	Octopine	Import
E. coli	Hly B	Hemolysin	Export
Pasturella	Ltk B	Leukotoxin	Export
E. coli	Cva B	Colicin V	Export
Erwinia chrysanthemi	Prt D	Proteases	Export
Bordetella pertussis	Cya B	Cyclolysin	Export
Streptococcus	Com A	Competence factor	Export?
Haemophilus influenzae	Bex AB	Capsule polysaccharide	Export
E. coli	Uvr A	–	DNA repair
Rhizobium leguminosarum	Nod I	–	Nodulation
Cyanobacterium			
Anabaena	Het A	–	Differentiation
Synechococcus	Cys A	Sulphate	Import
Yeast			
S. cerevisiae	STE 6	α-mating factor	Export
S. cerevisiae	EF-3	–	Translation
Protozoa			
Leishmania	Idpgp A	Heavy metals	Export
Insects			
Drosophila	white-brown	Eye pigments	Transport
Plants			
Liverwort chloroplasts	Mbp X	?	Transport?
Mammals			
Mouse	CFTR	Chloride	Channel
Man	CFTR	Chloride	Channel
Man	mdr 3	?	Flippase?

*Modified from ref. 11 with permission.

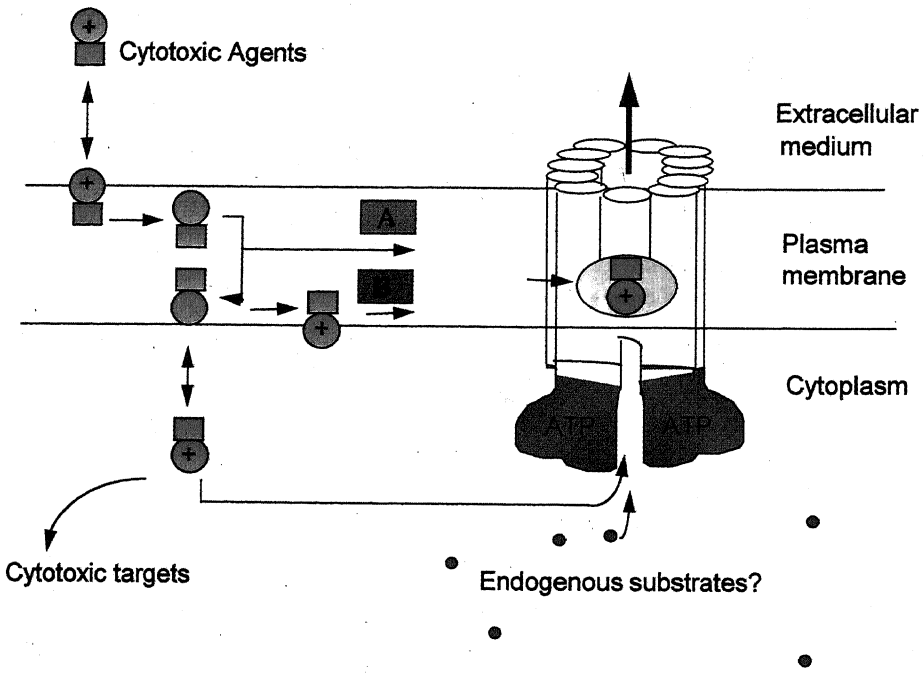

Figure 3. A model of MDR transporter. The drug is probably detected by the transporter within the lipid bilayer. Both uncharged (A) and charged (B) species of drugs could be the substrate for the transporter. The blue-coloured domains of protein indicate the ATP-binding sites. The red molecules are drugs and green molecules are putative physiological substrates for the transporter.

(i) MDR is unlikely, if ever, to be solely due to P-glycoprotein-mediated resistance, and (ii) P-glycoprotein is expressed by a very wide range of normal, noncancerous tissues as well. In the first case, therefore, prospective clinical protocol aimed at circumventing MDR may have to encompass more than just anti P-glycoprotein therapy. In the second case, successful anti-MDR therapy will probably have to be restricted to P-glycoprotein expressing tumour cells, to prevent unknown potentially deleterious consequences of inhibiting P-glycoprotein action in the normal healthy tissues.

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