Structure and Function of ABC Transporters in Health and Disease

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Clinical Pharmacology, January 13, 2011

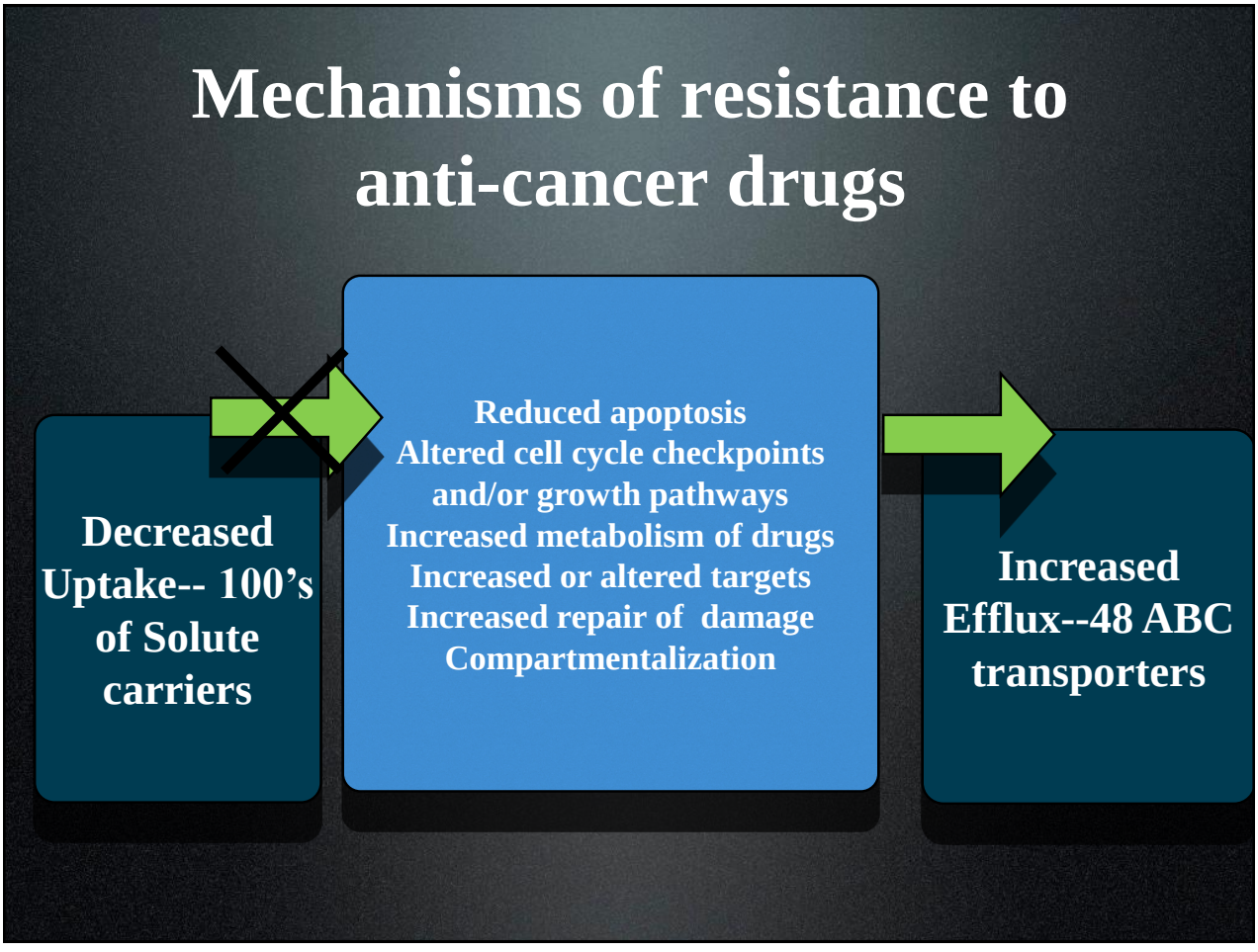
Drug Resistance in Cancer

- May affect multiple drugs used simultaneously: known as multidrug resistance (MDR)
- Affects all classes of drugs, including newly designed targeted drugs
- Just as oncogene targets have been catalogued, we need to enumerate all mechanisms of drug resistance in cancer to solve this problem and circumvent resistance

Ultimate Goals

1. Molecular analysis of human cancers to predict response to therapy
2. Use this information to develop novel drugs to treat cancer and new imaging modalities for cancer
3. To learn more about cellular pharmacology and pharmacokinetics of drugs

Mechanisms of resistance to anti-cancer drugs



Why study multidrug transporters?

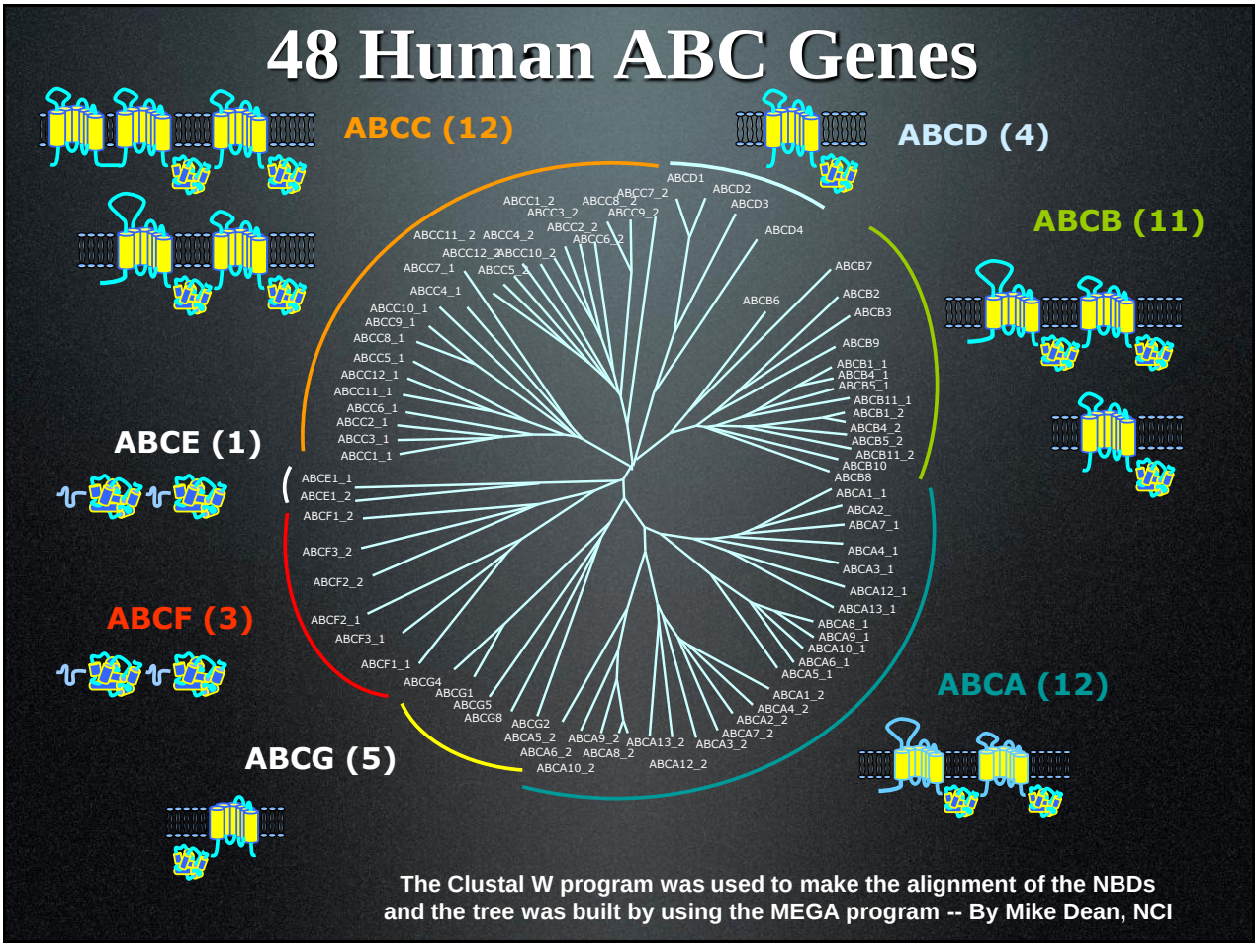
- Important role in multidrug resistance in cancer and in pathogens
- Important role in drug pharmacokinetics (uptake, distribution, and excretion)
- Important role in drug toxicity
- Key role in development (stem cells, morphogenesis)
- To learn about the biology of all transport systems

ATP-Binding Cassette (ABC) Transporter Superfamily

- One of the largest family of transport proteins known. Currently, more than 2000 members have been identified.
- Transport substrates include-- ions, sugars, glycans, phospholipids, cholesterol, peptides, proteins, toxins, antibiotics, and hydrophobic natural product anticancer drugs.
- Structurally, consist of various combinations of ATP-binding cassettes and segments with 6 trans-membrane domains.

The Eukaryotic ABCome
57 ABC-family genes

From M. Dean


48 Human ABC Genes
ABCC (12)
ABCD (4)
ABCB (11)
ABCE (1)
ABCF (3)
ABCG (5)
ABCA (12)
ABCD1
ABCD2
ABCD3
ABCD4
ABCC7_2
ABCC1_2
ABCC3_2
ABCC9_2
ABCC2_2
ABCC6_2
ABCC11_2
ABCC4_2
ABCC12_2
ABCC10_2
ABCC7_1
ABCC5_2
ABCC4_1
ABCC10_1
ABCC9_1
ABCC8_1
ABCC5_1
ABCC12_1
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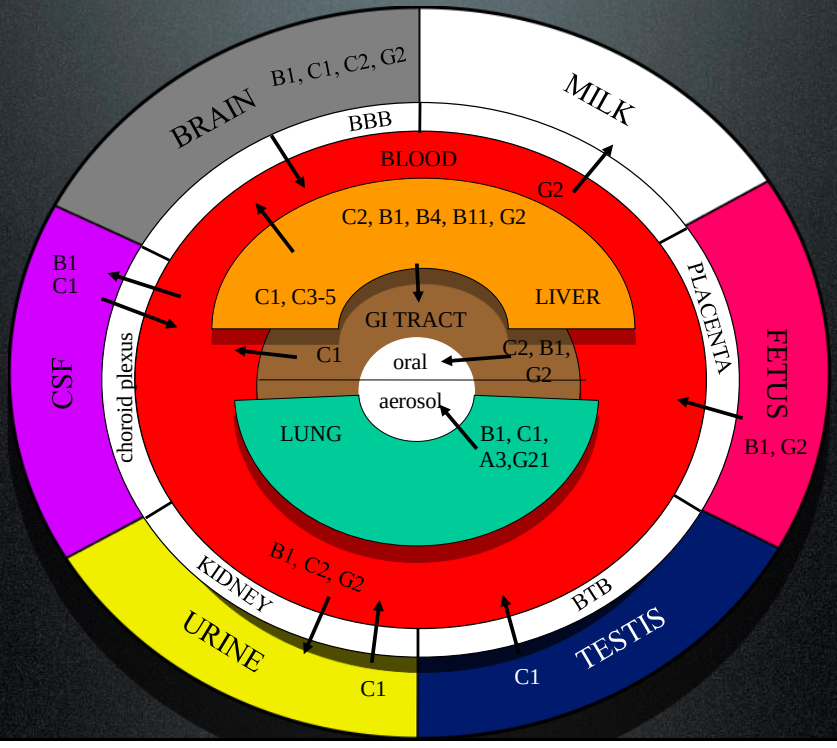
ABCC6_1
ABCC2_1
ABCC3_1
ABCC1_1
ABCB7
ABCB2
ABCB3
ABCB6
ABCB9
ABCB1_1
ABCB4_1
ABCB5_1
ABCB11_1
ABCB1_2
ABCB4_2
ABCB5_2
ABCB11_2
ABCB10
ABCB8
ABCE1_1
ABCE1_2
ABCF1_2
ABCF3_2
ABCF2_2
ABCF2_1
ABCF3_1
ABCF1_1
ABCG4
ABCG1
ABCG5
ABCG8
ABCG2
ABCA5_2
ABCA9_2
ABCA6_2
ABCA8_2
ABCA10_2
ABCA13_2
ABCA12_2
ABCA3_2
ABCA7_2
ABCA2_2
ABCA4_2
ABCA1_2
ABCA5_1
ABCA6_1
ABCA10_1
ABCA9_1
ABCA8_1
ABCA13_1
ABCA12_1
ABCA3_1
ABCA4_1
ABCA7_1
ABCA2_
ABCA1_1
The Clustal W program was used to make the alignment of the NBDs
and the tree was built by using the MEGA program -- By Mike Dean, NCI

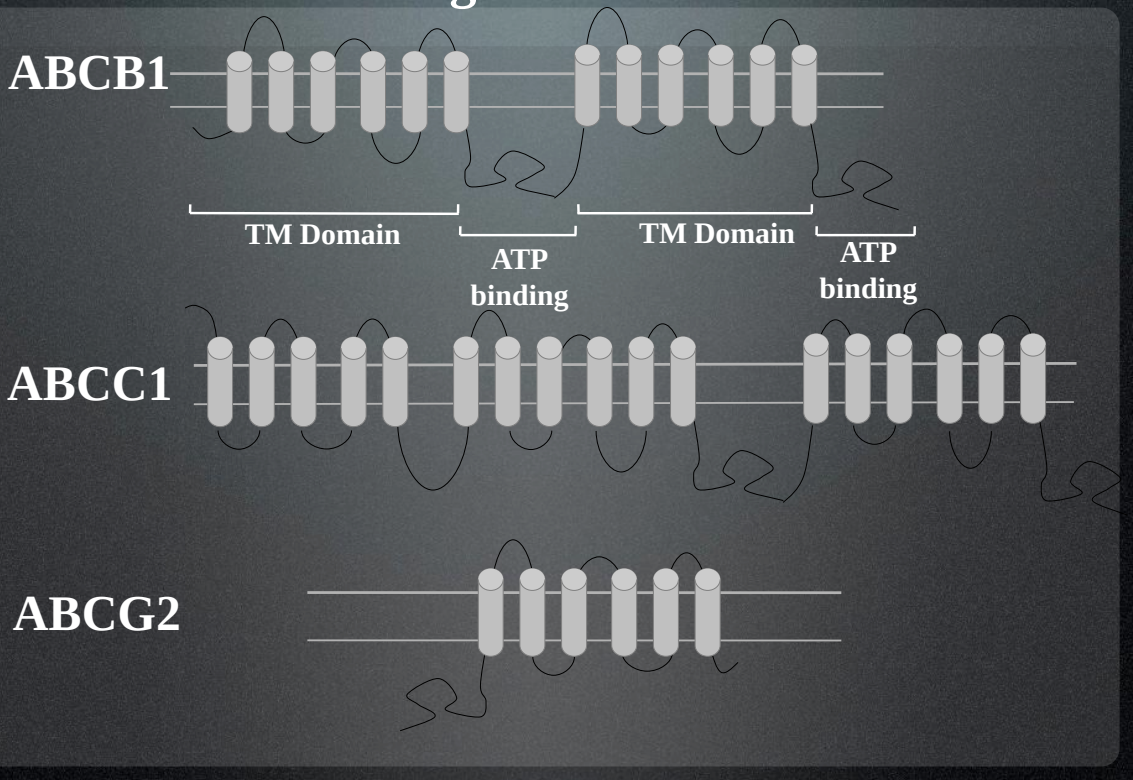
ABC transporters determine oral bioavailability, excretion, penetration and protect the organism against airborne xenobiotics



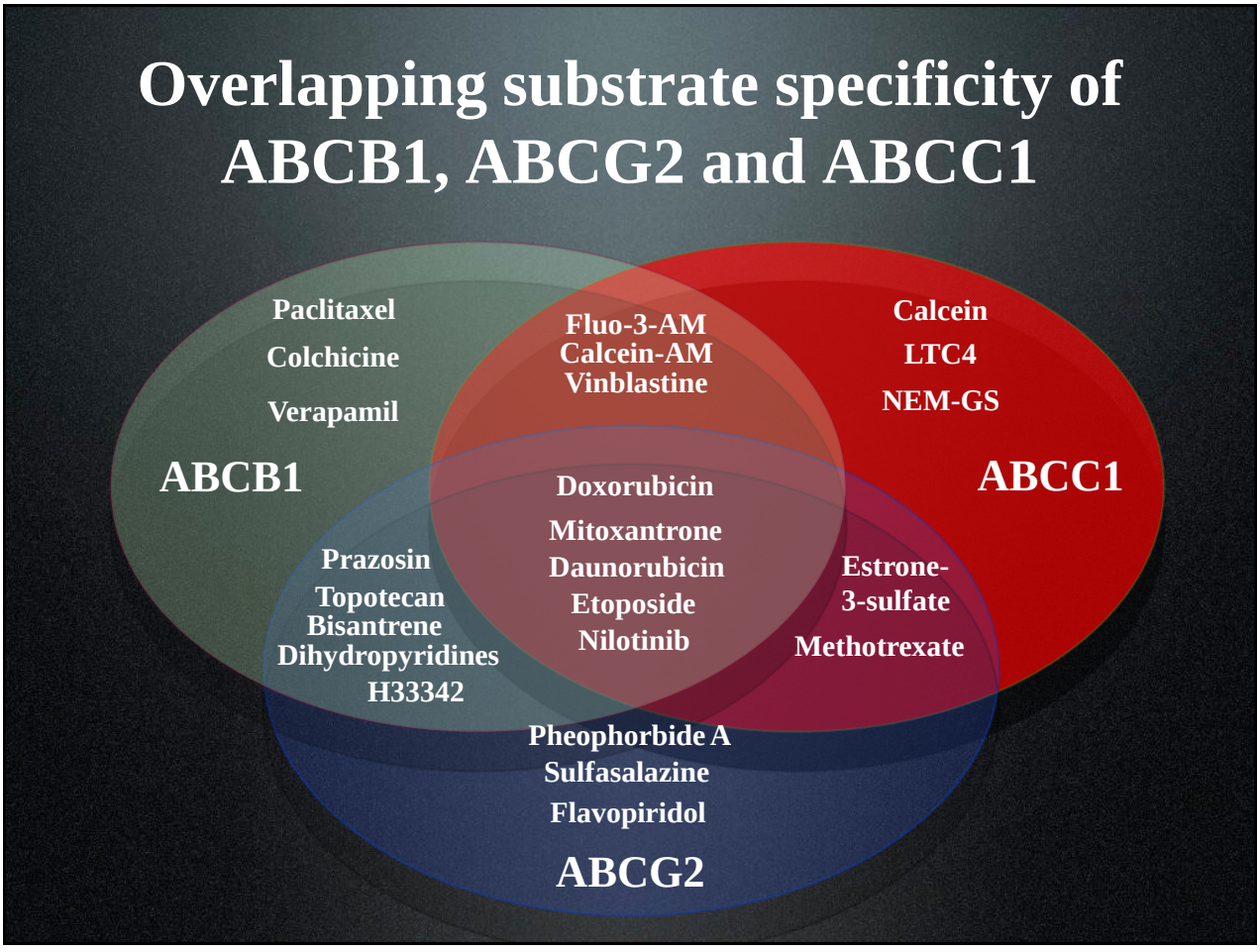
Human diseases associated with an ABC Transporter

Disease	Transporter
Cancer	ABCB1, ABCC1, ABCG2
Cystic fibrosis	ABCC7 (CFTR)
Stargardt disease & AMD	ABCA4 (ABCR)
Tangier Disease (HDL deficiency)	ABCA1 (ABC1)
Progressive familial intrahepatic cholestasis	ABCB11(SPGP), ABCB4 (MDR2)
Dubin-Johnson syndrome	ABCC2 (MRP2)
Pseudoxanthoma elasticum	ABCC6 (MRP6)
Persistent hypoglycemia of infancy, neonatal diabetes	ABCC8 (SUR1), ABCC9 (SUR2)
Sideroblastic anemia and ataxia	ABCB7 (ABC7)
Adrenoleukodystrophy	ABCD1 (ALD)
Sitosterolemia	ABCG5, ABCG8
Immune deficiency	ABCB2 (Tap1), ABCB3 (Tap2)

ABC transporters that confer MDR: Domain organization



Overlapping substrate specificity of ABCB1, ABCG2 and ABCC1



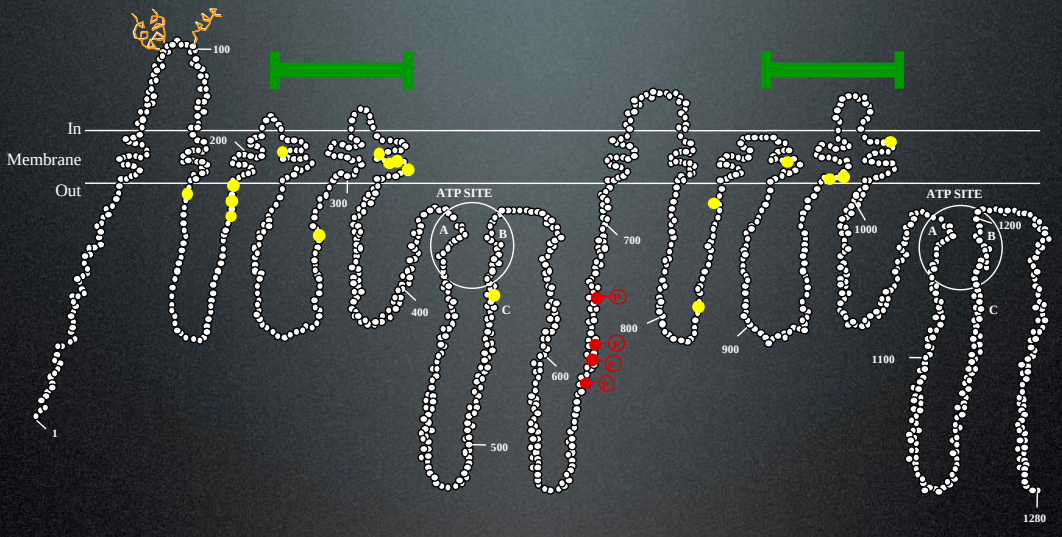
Multiple ABC Transporters Confer Resistance to Anti-Cancer Drugs

		ABC transporters overexpressed in cell lines selected for resistance						ABC transporters shown to confer drug resistance in transfection studies					
		ABCA2	ABCB1	ABCC1	ABCC2	ABCC4	ABCG2	ABCB5	ABCB11	ABCC3	ABCC5	ABCC6	ABCC11
Vinca alkaloids	Vinblastine		Selected	Confers resistance	Confers resistance								
	Vincristine		Confers resistance	Confers resistance	Confers resistance								
Anthracyclines	Daunorubicin		Selected	Confers resistance			Confers resistance	Confers resistance				Confers resistance	
	Doxorubicin		Selected	Selected	Confers resistance		Confers resistance	Confers resistance				Confers resistance	
	Epirubicin		Confers resistance	Confers resistance	Confers resistance		Confers resistance						
Epipodophyllotoxins	Etoposide		Confers resistance	Confers resistance	Confers resistance			Confers resistance		Confers resistance		Confers resistance	
	Teniposide		Confers resistance							Confers resistance		Confers resistance	
Taxanes	Docetaxel		Confers resistance										
	Paclitaxel		Confers resistance		Confers resistance				Confers resistance				
Kinase inhibitors	Gleevec		Confers resistance	Confers resistance			Confers resistance						
	Flavopiridol						Confers resistance						
Campthotecins	Irinotecan (CPT-11)		Confers resistance	Confers resistance	Confers resistance	Confers resistance	Confers resistance						
	SN-38		Confers resistance	Confers resistance	Confers resistance	Confers resistance	Confers resistance						
	Topotecan				Confers resistance	Confers resistance	Selected						
Thiopurines	6-mercaptopurine					Selected					Confers resistance		
	6-thioguanine					Selected					Confers resistance		
	5-FU										Confers resistance		Confers resistance
Other	Bisantrene		Confers resistance				Confers resistance						
	Cisplatin				Selected						Confers resistance	Confers resistance	
	Arsenite			Confers resistance	Selected								
	Colchicine		Selected	Selected									
	Estramustine	Selected											
	Methotrexate		Confers resistance	Confers resistance	Confers resistance	Confers resistance	Confers resistance			Confers resistance	Confers resistance		Confers resistance
	Mitoxantrone	Confers resistance			Confers resistance		Selected	Confers resistance					
	Saquinivir		Confers resistance										
	PMEA					Selected					Confers resistance		Confers resistance
	Actinomycin-D		Confers resistance										
	AZT					Confers resistance	Confers resistance				Confers resistance		

Confers resistance

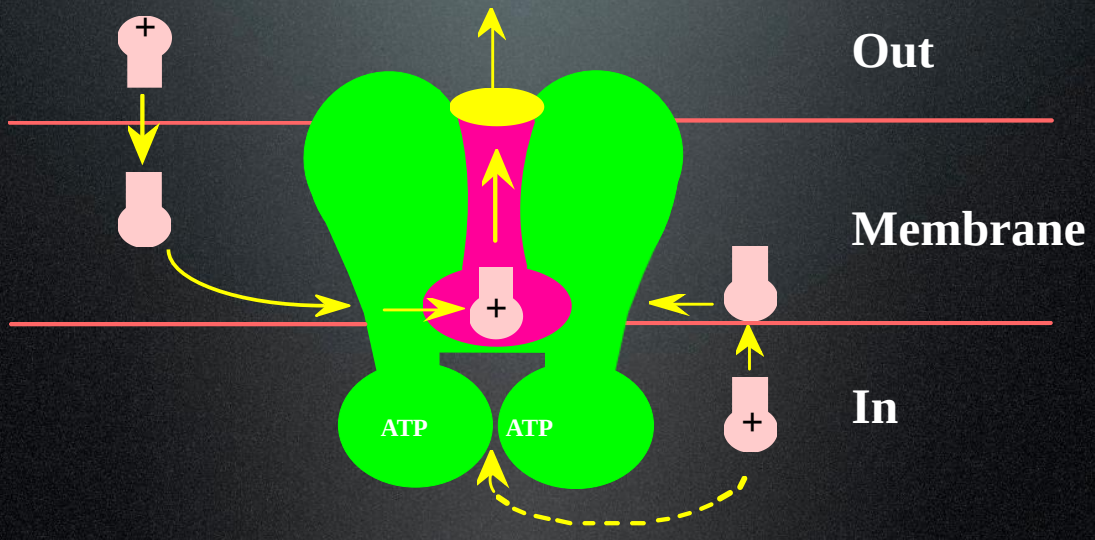
Selected

Hypothetical Model of Human P-glycoprotein

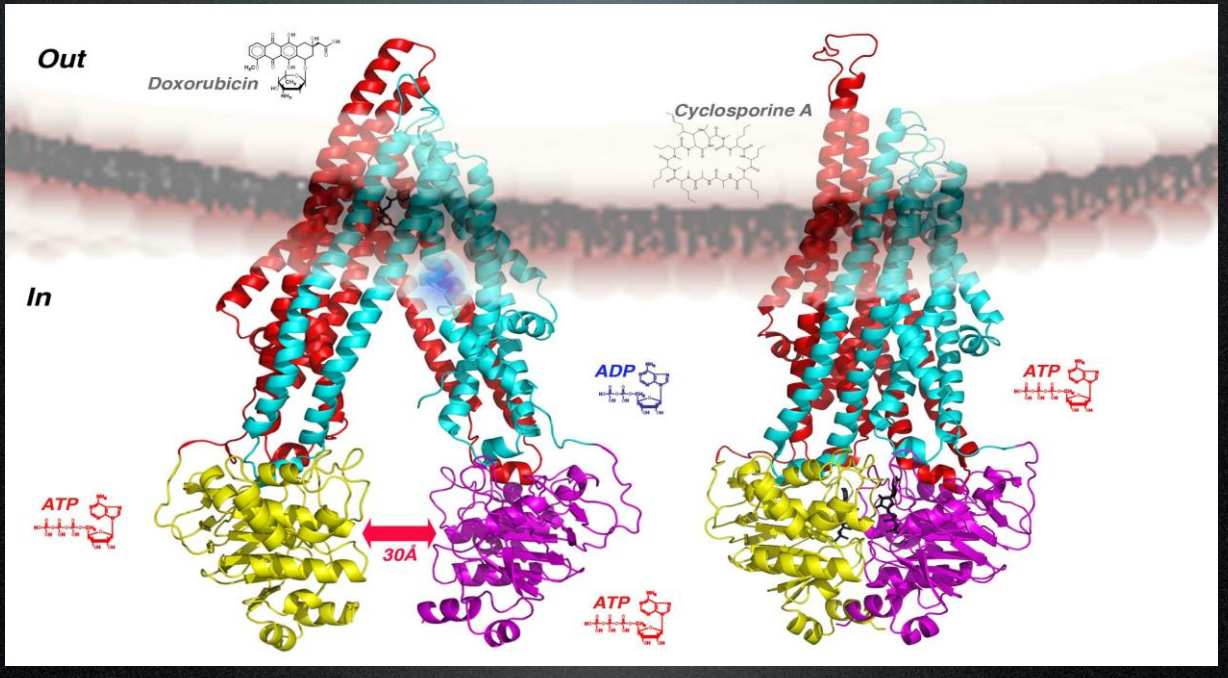


POINT MUTATIONS (●),
PHOTOAFFINITY LABELED
REGIONS (⊢⊣), AND
PHOSPHORYLATION SITES (Ⓟ)

P-glycoprotein removes hydrophobic substrates directly from the plasma membrane



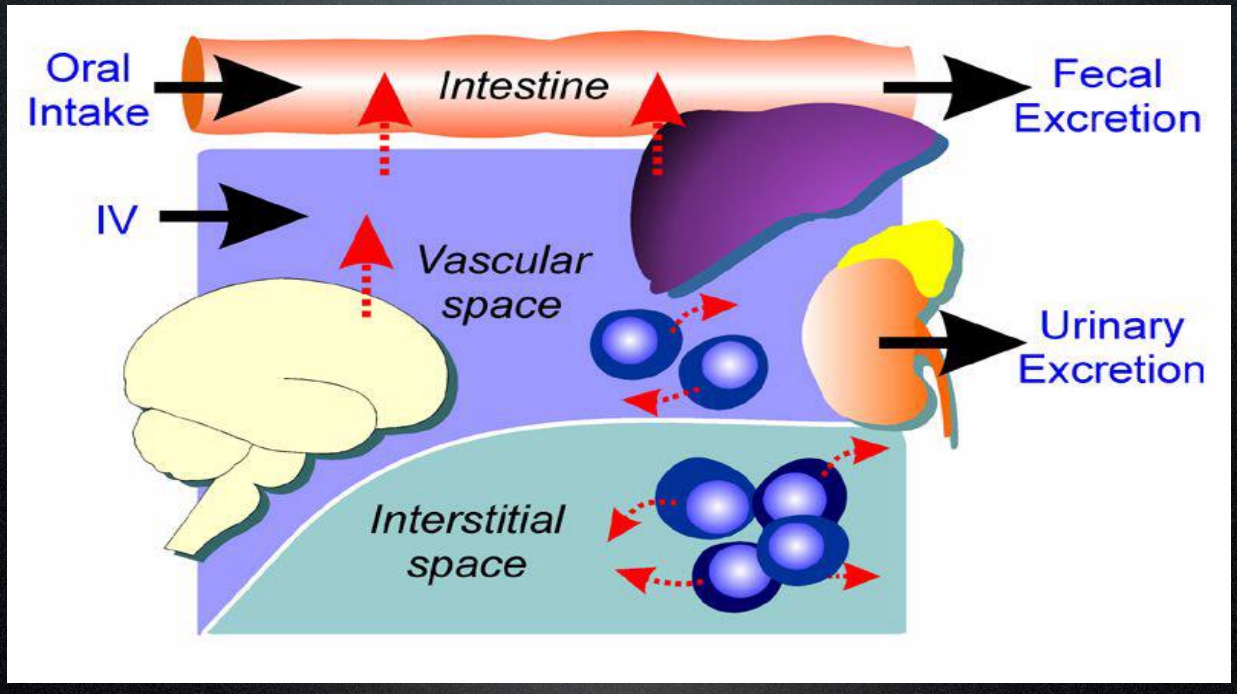
Atomic models of the structures of P-gp



Mouse P-gp at 3.8Å (Aller and Chang)

Human P-gp model based on Sav1866 (Xia)

Physiologic Role of P-glycoprotein



Role of P-glycoprotein in cancer

- Approximately 50% of human cancers express P-glycoprotein at levels sufficient to confer MDR
- Cancers which acquire expression of P-gp following treatment of the patient include leukemias, myeloma, lymphomas, breast, ovarian cancer; preliminary results with P-gp inhibitors suggest improved response to chemotherapy in some of these patients
- Cancers which express P-gp at time of diagnosis include colon, kidney, pancreas, liver; these do not respond to P-gp inhibitors alone and have other mechanisms of resistance
- Animal models with human cancer xenografts and BRCA1-driven mouse mammary cancers show role for P-gp in MDR (Pajic et al., Cancer Res. 69, 6396-6404, 2009)