e-PKGene

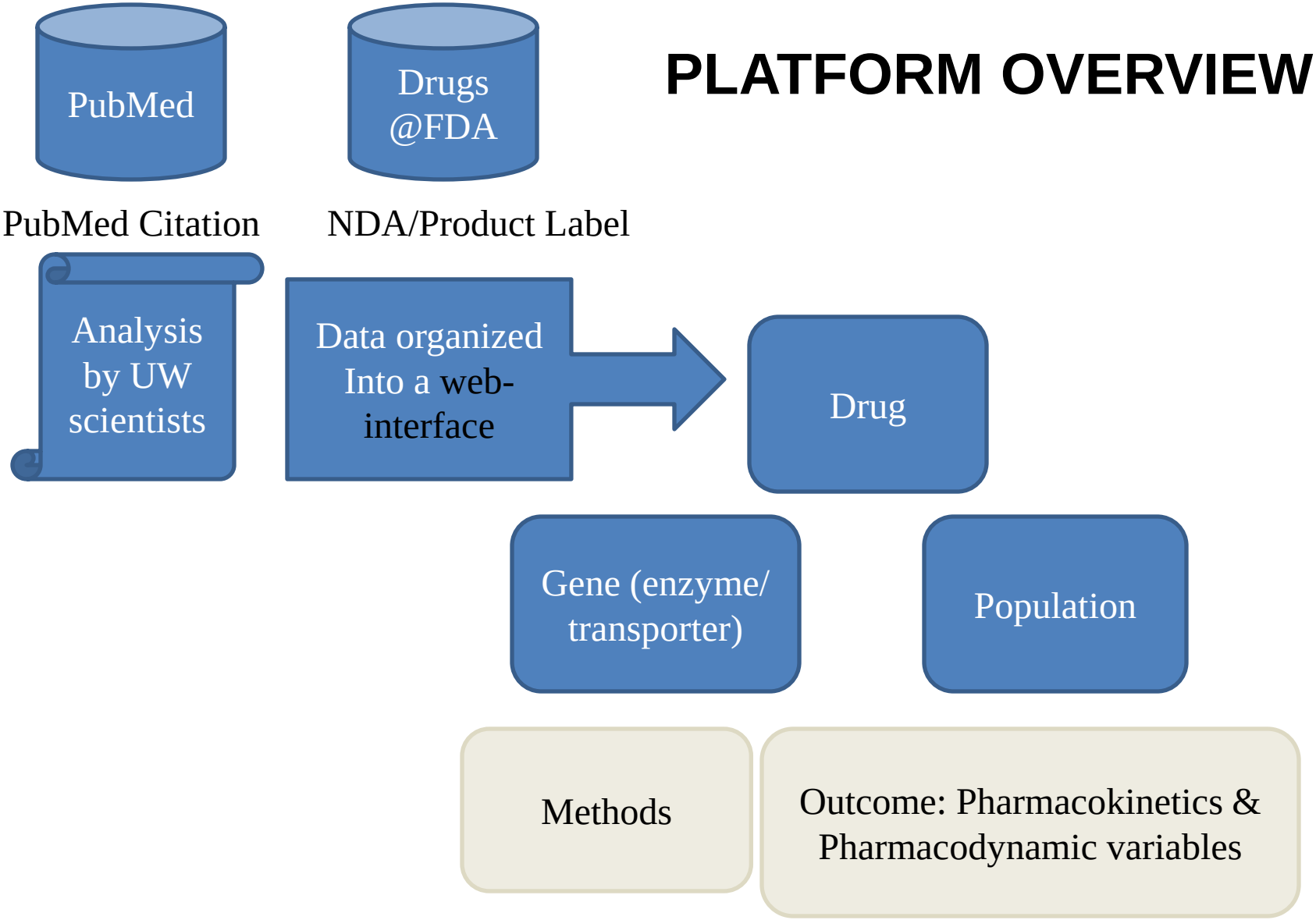
Pharmacogenetics Database
Background and Examples of Use

DIDB Program

Department of Pharmaceutics
School of Pharmacy
University of Washington

Background

PLATFORM OVERVIEW



e-PKGene Definitions

- **Articles** included in the e-PKGene database focus on drug exposure (AUC, Cl, C) in relation to pharmacogenetic variation. Studies can also include PD, clinical efficacy, and side effects.

- **Study**: set of assessments (PK, PD and safety) following the administration of a target compound to a well defined population.

- The **population** is usually divided into a “reference” group and an “impaired” group. The reference group usually consists of “extensive metabolizers” or carriers of two copies of the wild type allele of the gene of interest.

e-PKGene- Definitions

- e-PKGene defines each **variant allele** by one SNP which is unique to the allele. This SNP is identified as "diagnostic SNP", but it is important to note that *the diagnostic SNP may not be responsible for the observed functional changes*.

Genetic characteristics for the most common CYP2D6 alleles (diagnostic Single Nucleotide Polymorphism) [1] [2] [4] [5] [6] [7] [8] [9] [10] [11] [12] [15] [16]

Allele Name	Diagnostic SNP		SNP position	Effect of Variation	Amino Acid Change NP_000097.2	rs number	Activity Score Model-B	Enzyme activity in vivo
	Gene M33388	NM_000106.4						
CYP2D6*1	none			"wild type" [reference]	NA		1	normal
CYP2D6*2	**2850C>T** **4180G>C**	c.886T>C c.1457C>G	exon 6 exon 9	substitution (missense)	R296C T486S	rs16947 rs1135840	1	normal
CYP2D6*3	**2549delA** (also seen as 2637delA)	c.775delA	exon 5	frameshift (nonsense)	R259	**rs4986774**	0	none
CYP2D6*4	**1846G>A** (also seen as 1934G>A)	c.506-1G>A (IVS3 -1G>A)	intron 3	splicing defect (nonsense)	182ter	**rs3892097**	0	none
CYP2D6*5	**whole gene deletion**						0	none
CYP2D6*6	**1707 delT** (also seen as 1795delT)	c.454del T	exon 3	frameshift (nonsense)	W152X	**rs5030655**	0	none

e-PKGene Definitions

- Assignment of **Impact**: The classification is assigned by the database editorial team to provide users with a contextual framework that rapidly highlight variants of interest. Based on a predefined cut-off given by the statistical analysis performed by the authors (statistical significance).

e-PKGene rules (Impact of Variant)

1: statistical analysis must be performed by the authors of the study

2: control group is genotype *wt/*wt and/or Extensive Metabolizers

➢ **Active parent compound**

- Change in AUC
- Change in CL
- Change in Concentration

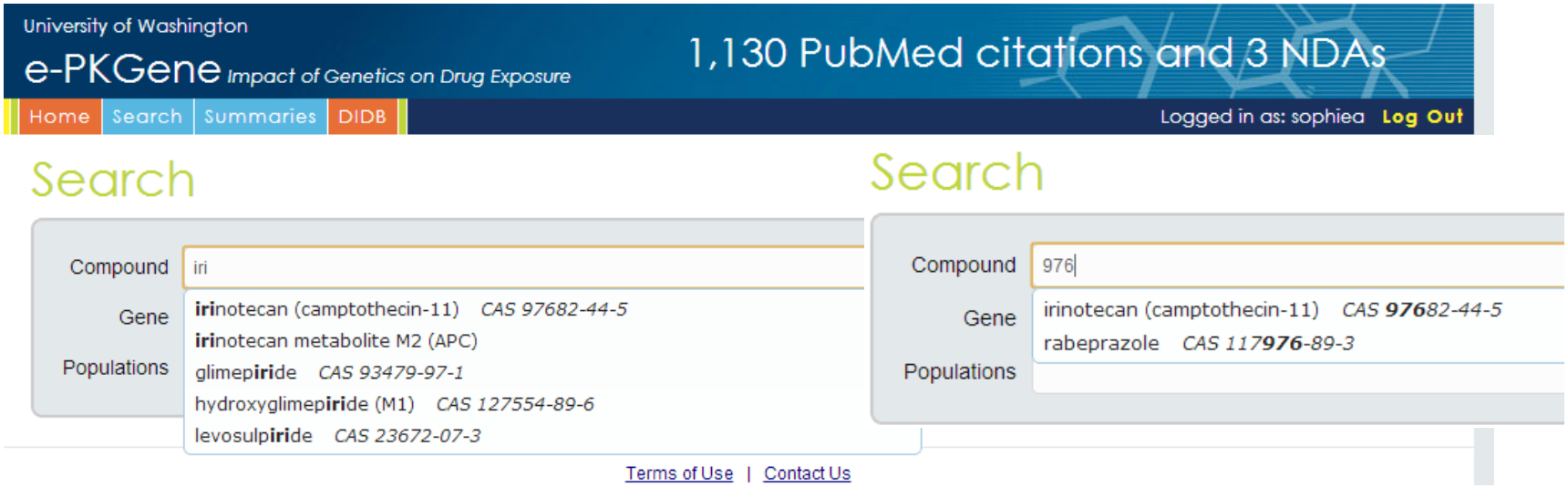
➢ **Metabolites**

- Change in CL (formation)
- Change in AUC ratio
- Change in Concentration ratio

Examples of Queries

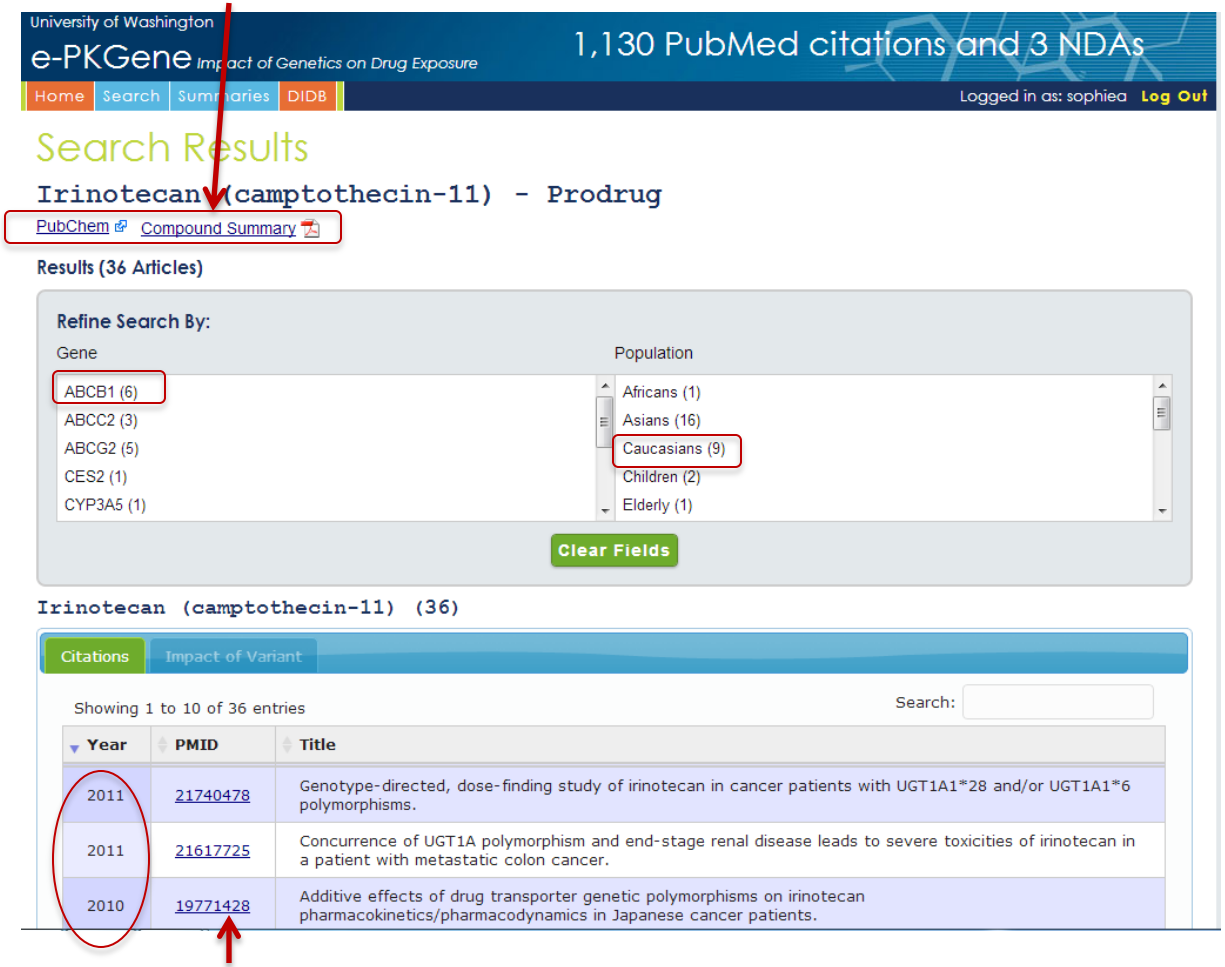
Example 1: **Compound** Search
Irinotecan

- Enter first letters of compound generic name OR first digits of compound CAS number
- Select the compound of interest



Search Results

Links to PubChem and compound summary



Link to citation display

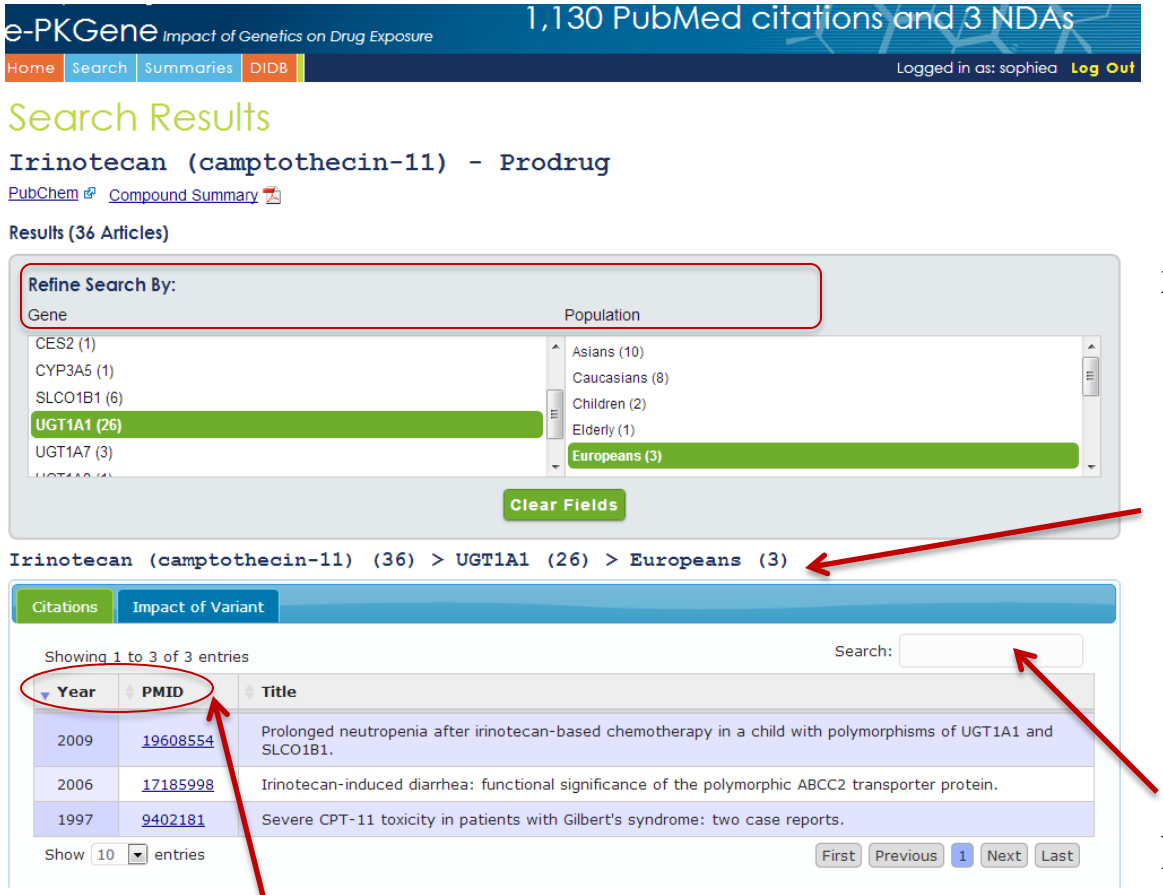
- **Gene box**: number of citations for each polymorphic gene studied

- **Population box**: number of citations performed in a given population (ethnic, health status)

- **Citations**: list ordered chronologically

-**Links**: PubChem compound, compound summary, citation display

Refine the search: by **Gene** and/or **Population**



Select gene and/or population of interest. This will:

1) Narrow the number of citations listed according to criteria of selection

1) Allows the **access to "Impact of Variant" box**

Specific additional search by key word, date or PMID

Reorder by ascending or descending years and PMIDs

Impact of Variant

Binary classification of genotypes/compound pairs based on PK differential observed across the various citations retrieved

Irinotecan (camptothecin-11) (36) > UGT1A1 (26) > Caucasians (8)

Citations | Impact of Variant

Specific genetic variants have been investigated in only 5 of the 8 citations retrieved.

UGT1A1*1/*28			
Yes		**No**	
SN-38 (7-Ethyl-10-hydroxycamptothecin)	17185998	irinotecan (camptothecin-11)	17185998
		SN-38 (7-Ethyl-10-hydroxycamptothecin)	15286088 15523087
		SN-38 glucuronide (7-ethyl-10-hydroxycamptothecin glucuronide)	15523087 17185998

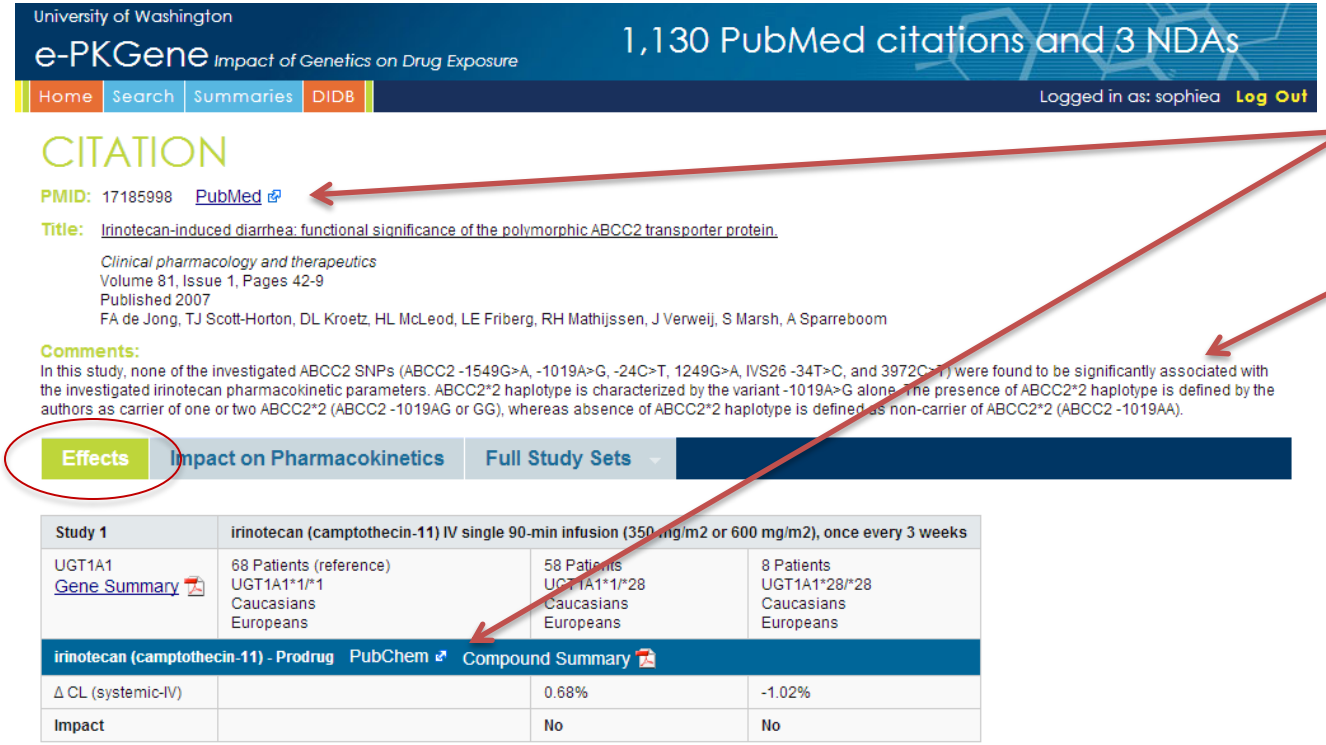
UGT1A1*28/*28			
Yes		**No**	
SN-38 (7-Ethyl-10-hydroxycamptothecin)	15523087 16809730	irinotecan (camptothecin-11)	17185998

Access to citation display

- Based on change of exposure between reference group (wild-type) and variant group
- Predefined cut-off: statistical analysis
- Based on Parent or metabolite(s) exposure

*Impact is assigned by the database team based on the following criteria: Impact=YES if the change in AUC, CL or Cmax is statistically significant (as determined by the study author). If changes are not statistically significant, the database team has assigned Impact=NO. Impact is assigned based on changes for the parent compounds, and metabolites.

Citation Display: **Effects** page screen



Features:

- Links to PubMed, PubChem, gene and compound summary
- Comments (curator)

Effects: summary of the impact of variant(s) on parent compound PK in the different populations (based on ΔAUC, ΔCL, or Δconc)

Citation Display: **Impact on Pharmacokinetics**

CITATION

PMID: 17185998 PubMed

Title: Irinotecan-induced diarrhea: functional significance of the polymorphic ABCC2 transporter protein.

Clinical pharmacology and therapeutics
Volume 81, Issue 1, Pages 42-9
Published 2007
FA de Jong, TJ Scott-Horton, DL Kroetz, HL McLeod, LE Friberg, RH Mathijssen, J Verweij, S Marsh, A Sparreboom

Comments:
In this study, none of the investigated ABCC2 SNPs (ABCC2 -1549G>A, -1019A>G, -24C>T, 1249G>A, IVS26 -34T>C, and 3972C>T) were found to be significantly associated with the investigated irinotecan pharmacokinetic parameters. ABCC2*2 haplotype is characterized by the variant -1019A>G alone. The presence of ABCC2*2 haplotype is defined by the authors as carrier of one or two ABCC2*2 (ABCC2 -1019AG or GG), whereas absence of ABCC2*2 haplotype is defined as non-carrier of ABCC2*2 (ABCC2 -1019AA).

Effects | Impact on Pharmacokinetics | Full Study Sets

Study 1	irinotecan (camptothecin-11) IV single 90-min infusion (350 mg/m2 or 600 mg/m2), once every 3 weeks		
UGT1A1 Gene Summary	68 Patients (reference) UGT1A1*1/*1 Caucasians Europeans	58 Patients UGT1A1*1/*28 Caucasians Europeans	8 Patients UGT1A1*28/*28 Caucasians Europeans
irinotecan (camptothecin-11) - Prodrug PubChem Compound Summary			
CL (systemic-IV) (L/h) (Medians (Range))	29.5 (13.1 - 50.41)	29.7 (11.7 - 50.4)	29.2 (17.3 - 41.1)
Δ CL (systemic-IV) %		0.68	-1.02
SN-38 (7-Ethyl-10-hydroxycamptothecin) - active metabolite PubChem			
AUC ratio (metabolite/parent) (Medians (Range))	3 (1.08 - 7.35)	3.64 (1.88 - 24.5)	4.49 (2.4 - 8.1)
Δ AUC ratio (metabolite/parent) %		21.33	49.67*
Biliary Index (Medians (Range))	7.95 (1.92 - 27.1)	9.04 (0.97 - 24.5)	18.8 (7.1 - 34.3)
Δ Biliary Index %		13.71	136.48*
CL (systemic-IV) (L/h) (Medians (Range))	387 (199 - 1683)	316 (107 - 833)	266 (68.8 - 495)
Δ CL (systemic-IV) %		-18.35*	-31.27*
SN-38 glucuronide (7-ethyl-10-hydroxycamptothecin glucuronide)			
AUC ratio (metabolite/parent) (Medians (Range))	4.41 (1.26 - 12.2)	4.02 (1.84 - 36.3)	2.24 (1.38 - 3.97)
Δ AUC ratio (metabolite/parent) %		-8.84	-49.21*
CL (systemic-IV) (L/h) (Medians (Range))	57.6 (23.7 - 214)	53.8 (2.24 - 163)	69.6 (34.3 - 125)
Δ CL (systemic-IV) %		-6.6	20.83

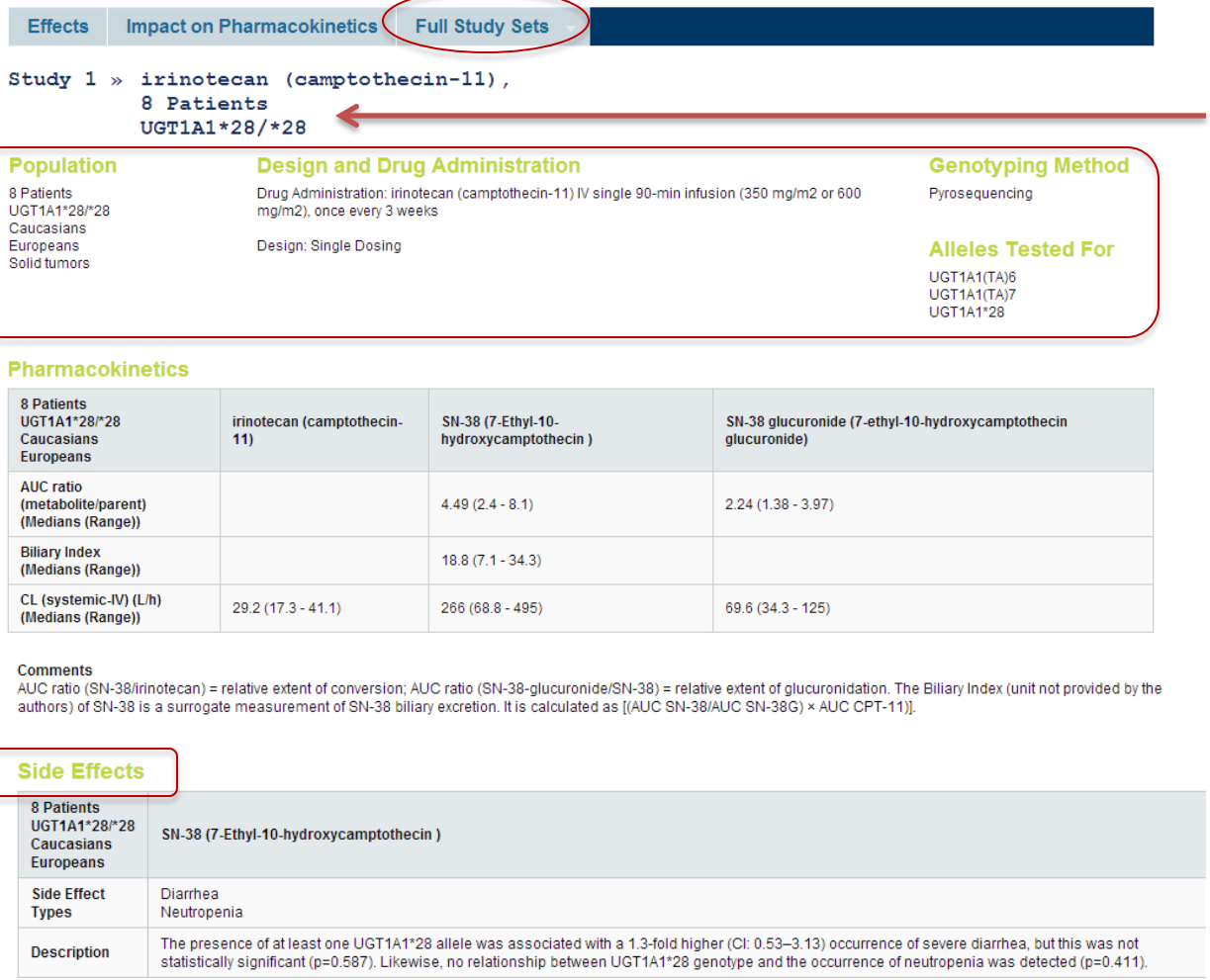
* Change is statistically significant (P<0.05) or Confidence interval outside equivalence boundaries.

- Overview of all PK parameters measured in each population groups reported in the citation

- for parent compound and metabolites

- statistical significance *vs* reference group.

Citation Display: **Full Study Sets**



Effects | Impact on Pharmacokinetics | Full Study Sets

Study 1 » irinotecan (camptothecin-11),
8 Patients
UGT1A1*28/*28

Population

8 Patients
UGT1A1*28/*28
Caucasians
Europeans
Solid tumors

Design and Drug Administration

Drug Administration: irinotecan (camptothecin-11) IV single 90-min infusion (350 mg/m2 or 600 mg/m2), once every 3 weeks

Design: Single Dosing

Genotyping Method

Pyrosequencing

Alleles Tested For

UGT1A1(TA)6
UGT1A1(TA)7
UGT1A1*28

Pharmacokinetics

8 Patients UGT1A1*28/*28 Caucasians Europeans	irinotecan (camptothecin-11)	SN-38 (7-Ethyl-10-hydroxycamptothecin)	SN-38 glucuronide (7-ethyl-10-hydroxycamptothecin glucuronide)
AUC ratio (metabolite/parent) (Medians (Range))		4.49 (2.4 - 8.1)	2.24 (1.38 - 3.97)
Biliary Index (Medians (Range))		18.8 (7.1 - 34.3)	
CL (systemic-IV) (L/h) (Medians (Range))	29.2 (17.3 - 41.1)	266 (68.8 - 495)	69.6 (34.3 - 125)

Comments
AUC ratio (SN-38/irinotecan) = relative extent of conversion; AUC ratio (SN-38-glucuronide/SN-38) = relative extent of glucuronidation. The Biliary Index (unit not provided by the authors) of SN-38 is a surrogate measurement of SN-38 biliary excretion. It is calculated as [(AUC SN-38/AUC SN-38G) × AUC CPT-11)].

Side Effects

8 Patients UGT1A1*28/*28 Caucasians Europeans	SN-38 (7-Ethyl-10-hydroxycamptothecin)
Side Effect Types	Diarrhea Neutropenia
Description	The presence of at least one UGT1A1*28 allele was associated with a 1.3-fold higher (CI: 0.53–3.13) occurrence of severe diarrhea, but this was not statistically significant (p=0.587). Likewise, no relationship between UGT1A1*28 genotype and the occurrence of neutropenia was detected (p=0.411).

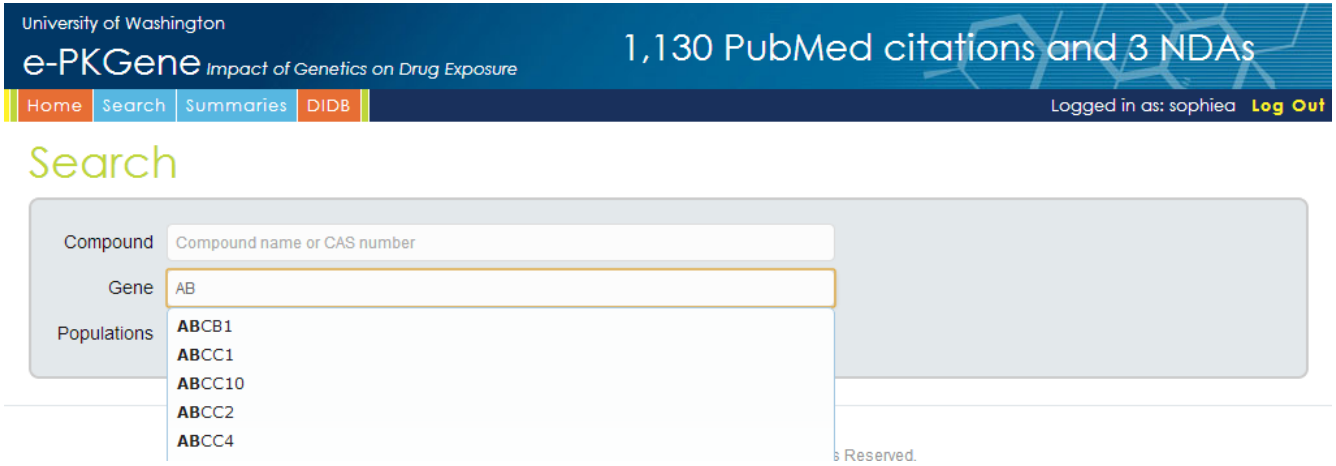
- Displays data captured for a study in **one population of interest**

- May include:
•Population description
•Study design/dosing regimen
•Phenotyping/Genotyping Methods
•Alleles tested for

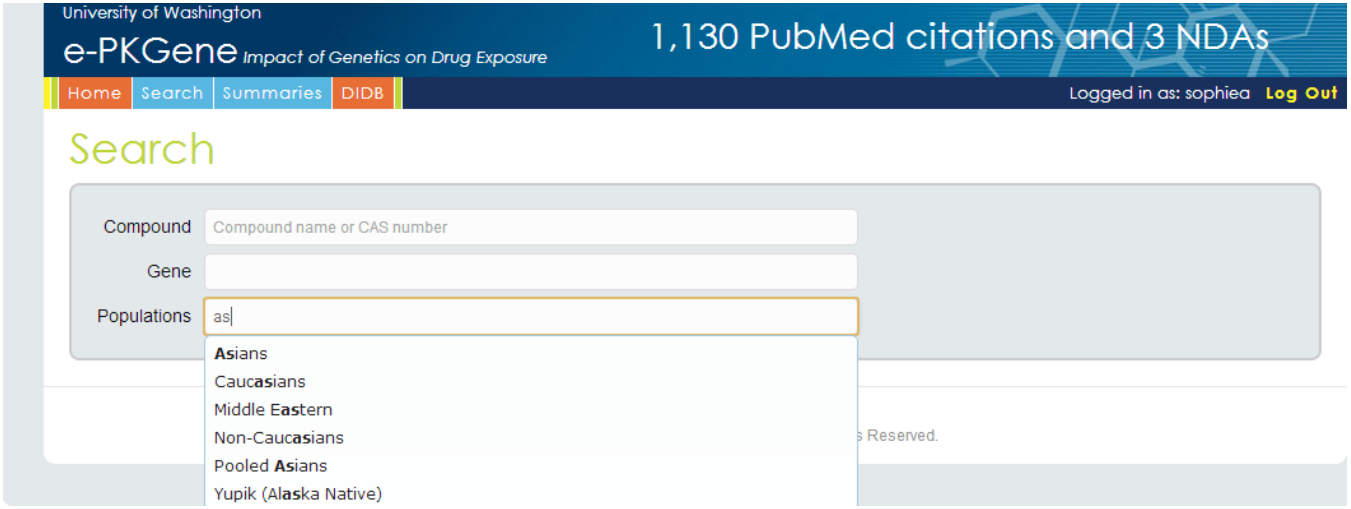
-**Pharmacodynamic** and/or **side effects** when these assessments are performed

Other Search Criteria

Search by **Gene**

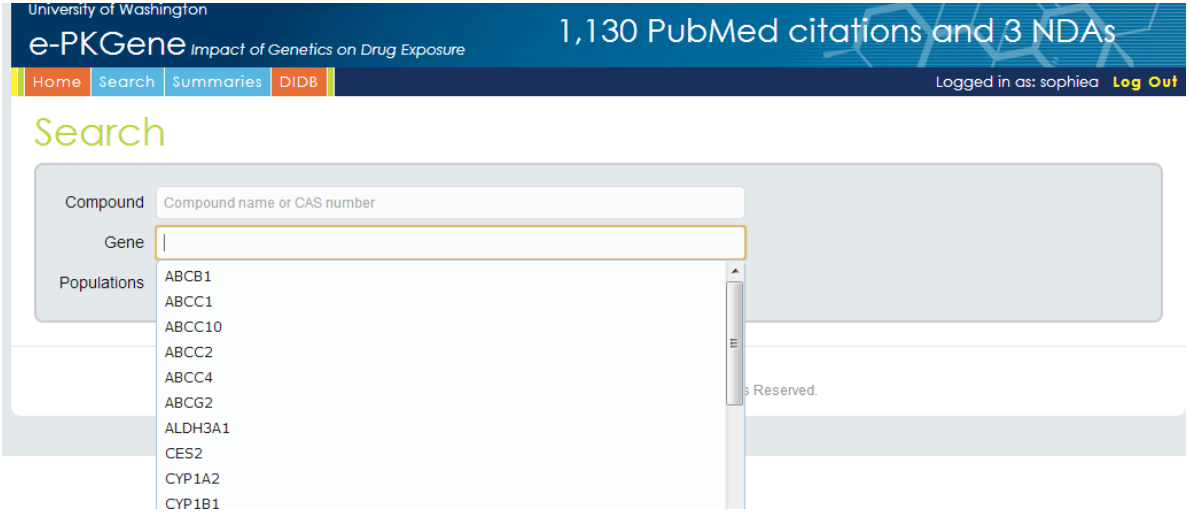


Search by **Population**

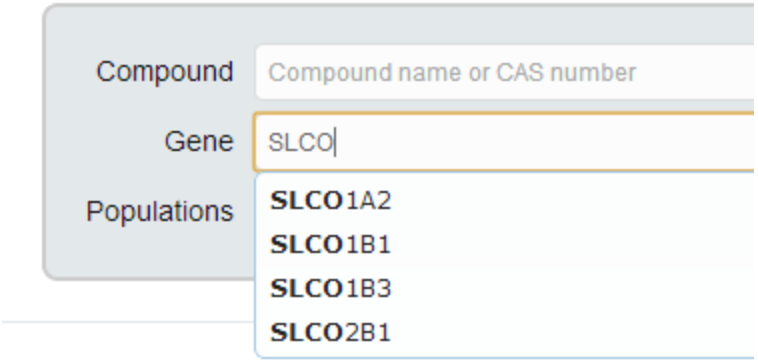


Example 2: **Search by gene -** SLCO1B1

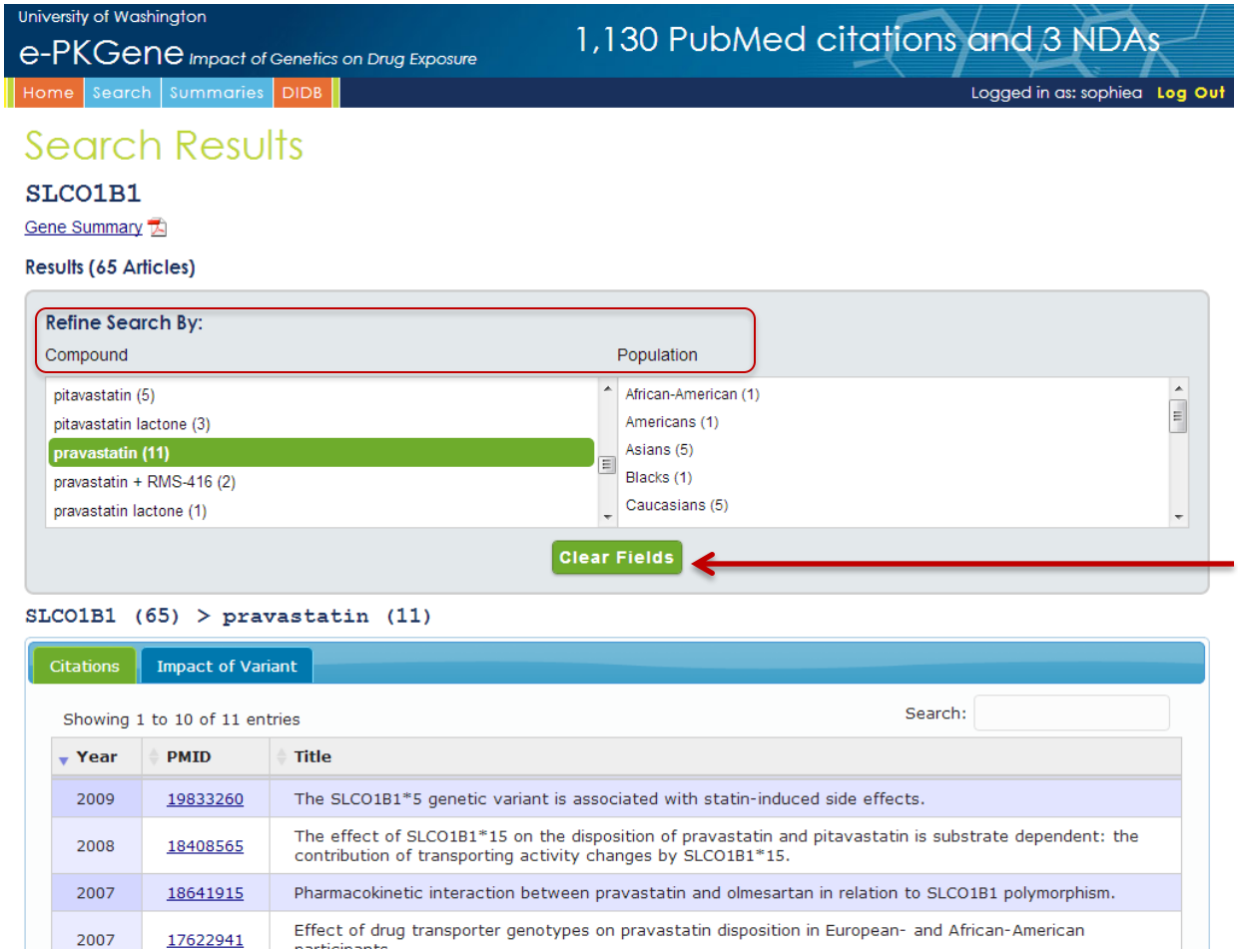
- Curser in the gene box: drop box with the whole gene list by default



- Type first letters of gene and select gene of interest



SLCO1B1 Search



- **Refine search**: select a **compound** and/or **population**

- "Clear Field" to perform a new search

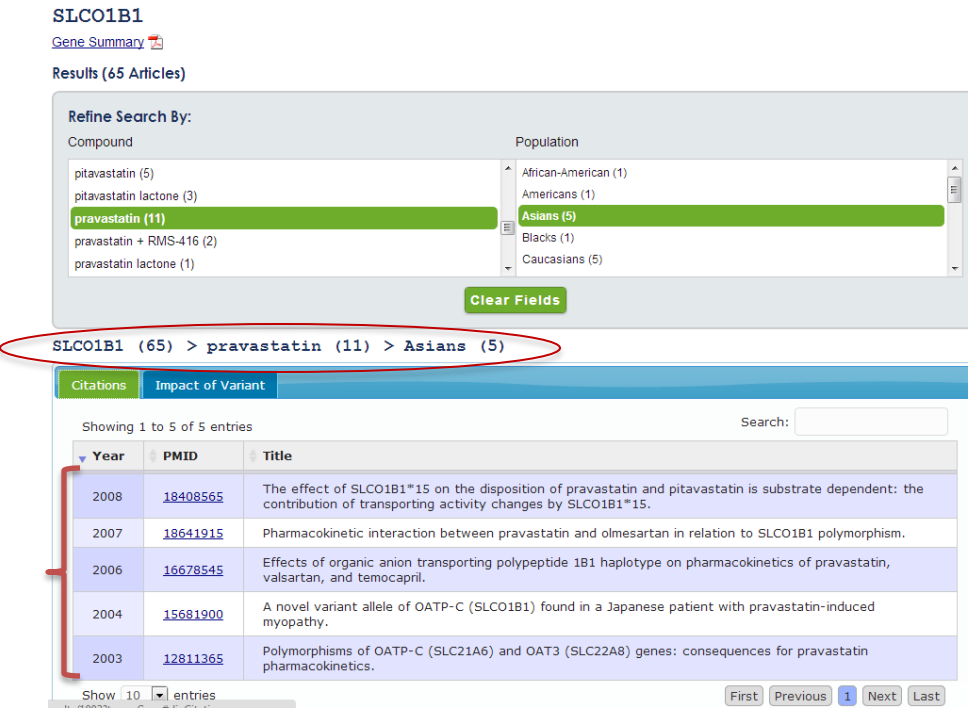
- **Citation list** displays only citations studying the compound in the population selected

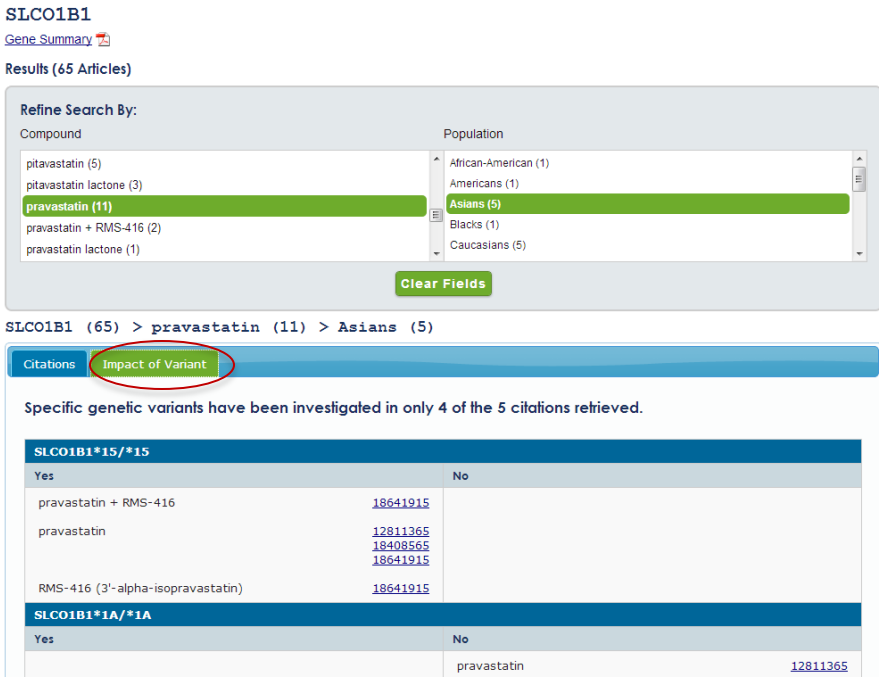
SLCO1B1 Search

Pravastin and **Asians** selected

List of citations refined

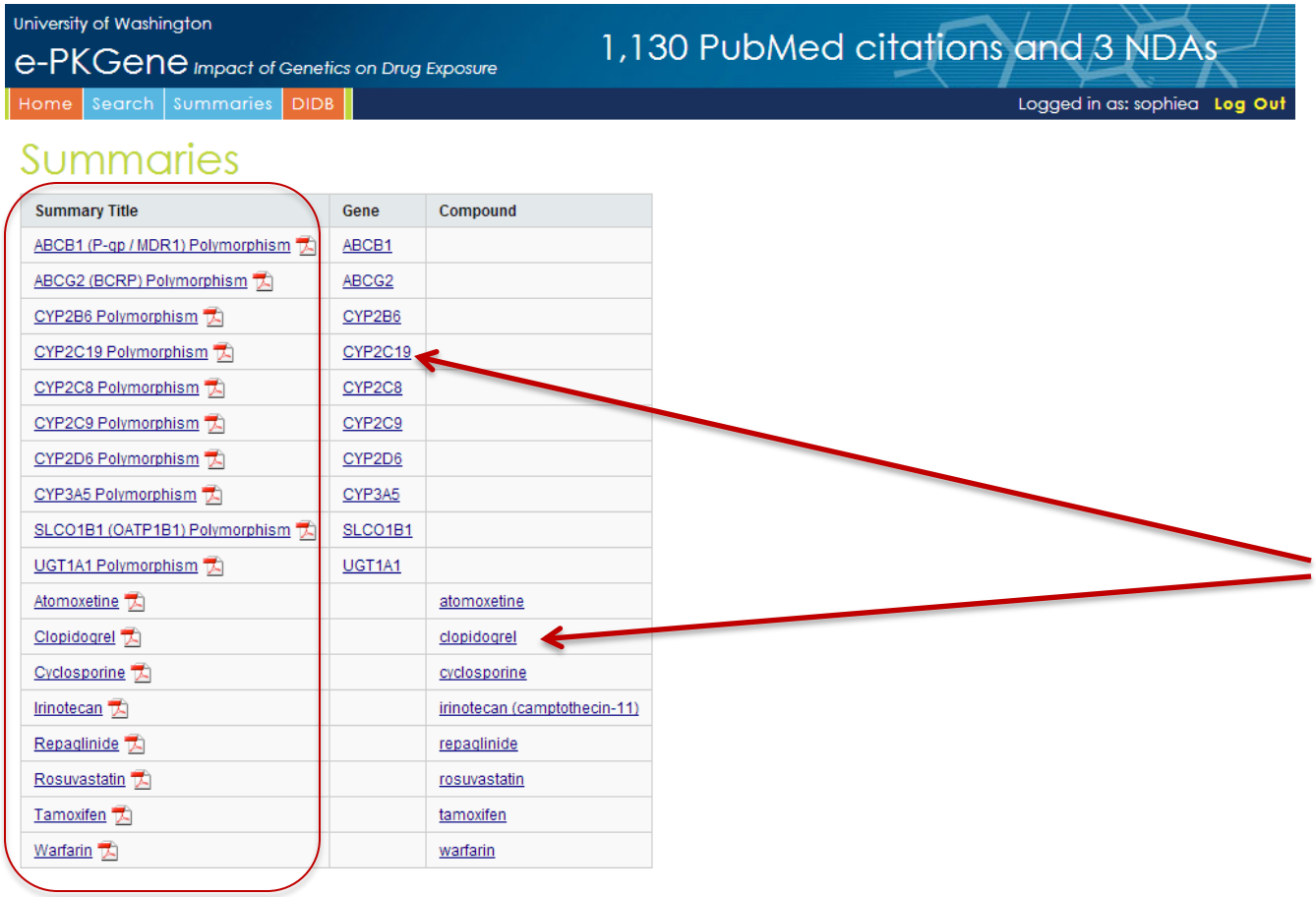
Impact box opens





Summaries

available in PDF format for **Drugs** and **Genes**



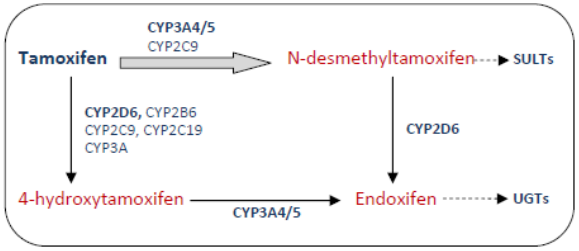
- PDF summaries for Genes and Drugs
- Direct access to gene results or compound results

Example of Drug Summary: **Tamoxifen**

ePKGene Summary May 2010

Tamoxifen

Background: Metabolism and Pharmacokinetics



Tamoxifen is a prodrug that requires metabolic activation to active metabolites 4-OH tamoxifen and 4-OH-N-desmethyltamoxifen (endoxifen) (1). Both CYP2D6 and CYP3A4 play a role in this activation. Loss or decrease of CYP2D6 function by genetic polymorphism may be associated with poorer clinical outcome compared with patients that have normal or ultrarapid metabolizer status (2).

1 Borges S, Desta Z, et.al. *Clinical Pharmacology & Therapeutics* (2006) **80**, 61–74
2 Kiyotani K, Mushiroda T, et. al. *J Clin Oncol*. (2010) **28**, 1287-93.

Pharmacokinetics/Pharmacodynamics Summary for Tamoxifen

Max % Change by CYP2D6 Genotype/Phenotype (6 studies evaluated)

			Homozygous EM or *1/*1	*1/*10	Range	*10/*10	Range	*4/*4	Range	Heterozygous EM	Range	Homozygous Variant (PM)	Range	Ultra-rapid (UM)	Range
PK	Tamoxifen	Css	Control	31	*	3.7	*	-22.4	*	-27.5	-1.1 to -27.4	-8.9	*	7.8	*
	N desmethyl tamoxifen	Css	Control	**	**	**	**	1.6	*	11.1	5.2 to 11.1	**17.5**	*	**-14.7**	*
	4-OH tamoxifen	Css	Control	-10.7	-1.9 to -10.7	**-46.4**	-22.6 to -46.4	-25.3	*	-12.6	-1.7 to -12.6	**-12.1**	*	1.7	*
	Endoxifen	Css	Control	-9	*	**-60.3**	*	**-74.4**	*	**-75.3**	-5.2 to -75.6	**-56.2**	-27.5 to -56.2	-11.5	*
PD	Not Available														

* Single study ** Not studied **BOLD**=statistically significant

Homozygous EM= assigned based on genotype to include *X/*Y (X=1, 2, 1xN, 41, Y=1, 2, 35, 1xN, 2xN, 41)

Heterozygous EM= assigned based on genotype to include one allele with compromised activity (10, 4, 5, 6)

Homozygous PM= assigned based on genotype to include two alleles with compromised activity (3,4, 5, 9, 10, 17, 21, 41)

Ultrarapid UM=assigned based on genotype to include *1/*2x2

Background: Summary of drug disposition characteristics

Maximum changes in exposure found in the literature for a given drug and a given genotype

Example of Gene Summary: **CYP2D6**

CYP2D6 Gene Polymorphism Summary

Official Full Name: cytochrome P450, family 2, subfamily D, polypeptide 6
Alias Names: RP4-669P10.2, CPD6, CYP2D, CYP2D@, CYP2DL1, MGC120389, MGC120390, P450-DB1, P450C2D, P450DB1
CYP2D6 Location: chromosome: 22; Location: 22q13.1
Gene Reference: M33388
Gene Identification Number: 1565
gene information from NCBI dbSNP database(Entrez Gene) [1]

Definitions

Single nucleotide polymorphism (SNP)[3]
A single nucleotide polymorphism (SNP) is a variation occurring in the DNA when a single nucleotide — A, T, C, or G —differs between a paired chromosomes in an individual. SNPs are the most common genetic variations in humans.

CYP2D6 allele classification[5][6]

- The CYP2D6*1 allele is the **wild-type** allele (it has no SNP) and encodes an enzyme with normal activity. It defines the **Extensive Metabolizers** status and is considered as the "Reference" allele.

Allele Name	Diagnostic SNP		SNP position	Effect of Variation	Amino Acid Change NP_000097.2	rs number	Activity Score Model-B	Enzyme activity in vivo
	Gene M33388	NM_000106.4						
CYP2D6*1		none		"wild type" [reference]	NA		1	normal
CYP2D6*2	**2850C>T** **4180G>C**	c.886T>C c.1457C>G	exon 6 exon 9	substitution (missense)	R296C T486S	rs16947 rs1135840	1	normal

Relation Genotype/Phenotype

It is important to note that phenotypes displayed in the citations are the ones the authors assigned to their

Activity Score	>2	2	1 < AS < 2	0 < AS ≤ 1	0
Phenotype	Ultrarapid Metabolizers (UMs)	Extensive Metabolizers (EMs)	Slow Extensive Metabolizers (SEMs)	Intermediate Metabolizers (IMs)	Poor Metabolizers (PMs)

- General description of Gene
- Definitions
- Classification of alleles
- Genetic description for some alleles
- Relation Genotype/Phenotype (when available)
- Frequencies by ethnicity
- References

For Comments and Questions

Contact DIDB Program at didbase@uw.edu