Drug-Drug Interactions Between Cephalosporin Antibiotics and Probenecid

There are many reports on the drug-drug interactions between cephalosporin antibiotics and probenecid (157). As both cephalosporins and probenecid interact with OAT family transporters, some of these drug-drug interactions may be due to an OAT-mediated uptake process. Most cephalosporins are excreted in the urine, which may be partly mediated by OAT family transporters. The elimination rates of cephazedone, cefazolin, cefalexin, cefradine, cefaclor, cefmetazole, cefoxitin, cefuroxime, cefmenoxime, ceftizoxime, and cedftriaxone were significantly reduced by coadministration of probenecid, which may be partly caused by the inhibition of their renal excretions (157).

Marino & Dominguez-Gil have shown that the pharmacokinetics of cefadroxil is altered by coadministration of probenecid (158). In their report, the peak concentration and half-life of cefadroxil was increased 1.4- and 1.3-fold, respectively, following coadministration of probenecid. Its urinary excretion rate constant falls by 58%, supporting the possibility of drug-drug interaction at the renal excretion. Supplemental Table 1 suggests that OAT1- and OAT3-mediated transport should be decreased to at most 25%–47% and 25%–69% of the control, and, therefore, it may be partly explained by the OAT-mediated drug-drug interaction.

Probenecid has also been shown to alter the plasma concentrations of cefamandole and ceftriaxone (159). The maximum plasma concentration and half-life of cefamandole were increased 6- and 1.8-fold by coadministration of probenecid (159). Also, 71% of cefamandole is excreted in the urine, and this was reduced to 66% of the control (159). The elimination of ceftriaxone was slightly affected by coadministration of probenecid (160). Probenecid reduced the serum clearance of ceftriaxone to 73% of the control (160). It reduced the renal and nonrenal clearance to 80% and 68% of the control, respectively, suggesting that this drug-drug interaction is, to a minor extent, due to renal excretion (160).

Drug-Drug Interaction Between Methotrexate and NSAIDs

To date, there are reports that coadministration of MTX with penicillin, probenecid, and NSAIDs cause drug-drug interactions and several potential sites for these DDI have been reported: an increase in the protein unbound fraction of MTX, a decrease in the urine flow rate resulting from the inhibition of prostaglandin synthesis, and inhibition of the renal tubular secretion of MTX (161–164). Nozaki et al. analyzed the uptake mechanism of MTX in rat kidney slices and examined the effects of NSAIDs on its uptake (165). They showed that rat Oat3 and reduced folate carrier 1 (RFC-1) equally contribute to the renal uptake (30% each), with the remaining fraction being accounted for by passive diffusion and/or adsorption, whereas rOat1 makes only a limited contribution (165). Many NSAIDs inhibited both rOat3- and RFC-1-mediated uptake of MTX, but the K_i value for Oat3 was lower than that for RFC-1 (165). At their therapeutic concentrations, they inhibited only Oat3-mediated uptake of MTX. Therefore, the affect of NSAIDs on the renal uptake of

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MTX is expected to be nonextensive and partial. Many NSAIDs also inhibit human OAT3-mediated uptake of MTX with therapeutic relevant plasma concentrations of unbound drugs (26). However, also in humans, the contribution of OAT3 to the total renal uptake of MTX needs to be clarified for the identification of the mechanism of the clinically relevant DDI.

CONCLUSION

In addition to phase I and phase II enzymes, transporters also play an important role in drug elimination and distribution. Therefore, it is possible that transporter-mediated drug-drug interactions alter pharmacokinetics, and could result in severe side effects.

A large number of transporters have been characterized in rodents and humans, and the mechanism of the membrane transport of several compounds including endogenous compounds and therapeutic drugs has been clarified. However, the transport mechanism of most therapeutic drugs remains unknown. To predict a transporter-mediated drug-drug interaction, the transporters involved in the membrane transport of the drug need to be characterized. As multiple transporters have been characterized in the kidney and liver and their expression systems are available, it should be possible to predict a transporter-mediated drug-drug interaction by using these systems with the information of the contribution made by each transporter to the net transport in the kidney and liver.

We have estimated the possibility of a transporter-mediated drug-drug interaction from the R value calculated using the maximum unbound concentration of inhibitors. This method may avoid false negative predictions of drug-drug interactions. In conclusion, greater awareness of the possibility of transporter-mediated drug-drug interactions is necessary.

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ゲノム科学と創薬

遺伝子多型と薬物の効果の個人差

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ポイント

- 薬効の個人差を生み出す要因は，薬物動態的な要因と薬力学的な要因の２つに大別され，それぞれを決定するタンパク質の遺伝子多型が重要と考えられる．
- 体内動態にかかわる分子には，代謝酵素・トランスポーターがあり，それぞれ遺伝子多型によって生じる機能変化が薬物動態を大きく変え，ひいては効果・副作用を規定する事例が数多く報告されている．
- 薬効・副作用の感受性の個人差は，直接薬のターゲットタンパク質の遺伝子多型によるケース以外に，薬効標的と関連のないタンパク質の多型とリンクするケースがあり，今後広範囲な遺伝子多型と感受性の関連解析，原因の追究が望まれる．

はじめに

臨床において，同じ量の薬物を投与しても，適正な治療効果を示す人もあれば，まったく効果のない人や副作用がでる人もある．その手がかりとして，患者集団での薬物血中濃度のヒストグラムに多峰性が観察されたり，２種の薬物を個々の患者に投与したときに，それぞれの代謝物の総量に相関が認められたりすること〔たとえば，デブリソキンとスパルテイン[1]（CYP2D6の多型に起因する）〕，また一卵性双生児と二卵性双生児ペアに薬物を投与して動態をペア間で比較したところ一卵性双生児の方が有意にペア間の変動が小さいことなどから，薬物動態の個人差の原因として遺伝的要因が関与することが示唆されてきた．現在では，これらは薬物代謝酵素の遺伝子多型で説明がついている．

一般的に，薬効・副作用の個人間変動に影響する過程は，投与後の体内での薬物動態の個人差の部分（pharmacokinetics）と，薬物の標的部位との相互作用，シグナル伝達の感受性など薬力学的な個人差の部分（pharmacodynamics）に大別できる．前者は，薬物の吸収，分布，代謝，排泄を決定する代謝酵素・トランスポーターの遺伝子多型が主に関与し，投与設計，投与量の個別化で個人差を克服できる．一方，後者は，薬物の標的となるレセプター，転写因子や，その下流のシグナル伝達を行うアダプター分子群の遺伝子多型が該当し，個人差の克服には薬剤選択の個別化が必要となる．またより厳密には，薬効発現の程度は標的分子近傍の薬物濃度の関数として表されることから，細胞内にターゲットをもつ場合は，血中濃度のみならず，標的細胞内濃度を決定する要因も考慮に入れる必要がある（たとえば耐性癌細胞における排出トランスポーター）（図１）．

薬物の体内動態にかかわる分子の遺伝子多型

一般に薬物の排泄は，① 血中から臓器細胞へ

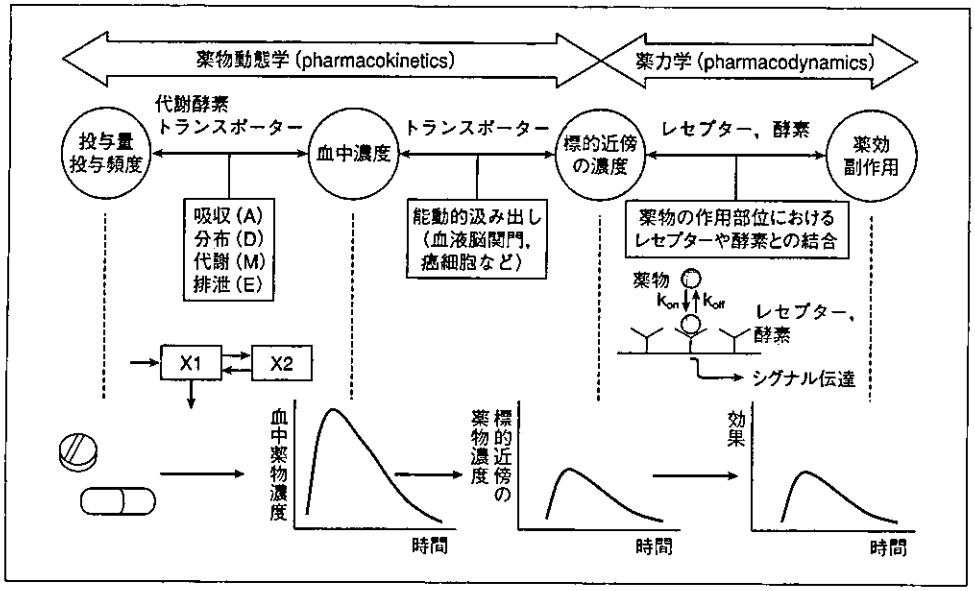

図1　薬物が効果・副作用を発現するまでの過程
(杉山雄一ほか 編：ファーマコキネティクス 演習による理解，p249，南山堂，2003[2] より改変)

の細胞膜を介した取り込み，②細胞内での主にシトクロム P-450（CYP）による酸化反応などの種々の物質変換，③グルクロン酸，グルタチオン，硫酸基など水溶性を上げる官能基を付加する抱合反応，④細胞内からの排出の各過程からなっており，それぞれの段階において多くの分子が機能している．①，④の段階においては，脂溶性が低く容易に膜透過できない物質について多くのトランスポーターが効率良い膜輸送を担っていることが明らかとなっており，これらの遺伝子多型も現在解析が進みつつある．

1．CYP の遺伝子多型と薬物動態の個人差

CYP は薬物の排泄にかかわる分子としていち早く遺伝子多型解析と体内動態との個人差の相関研究が行われた領域であり，臨床でも多くの事例が集積している．また多型のハプロタイプごとに体系化された命名法が確立している[3]．

CYP2D6 は，肝臓内における CYP 含量としては比較的少ないが，薬物代謝への寄与は，CYP3A4 に次ぎ薬物全体の約 20 % を占めるほど重要な酵素である[4]．現在までに，79 種類もの変異アレルの報告がある．多型の頻度には大きな人種差が認められている．欧米人には，機能欠損した遺伝子（主に*3，*4，*5）を両アレルにもつ poor metabolizer（PM）が 5～10 % 存在するのに対し，日本人では，PM の頻度は 1 % 未満（主に*5）である．さらに発現量と結合親和性の低下を示す*10 は，東洋人にのみ高頻度に出現し，PM と extensive metabolizer（EM，野生型）の中間の性質を示す intermediate metabolizer（IM，*5/*10，*10/*10）の分類が存在する．また，頻度は少ないものの，2D6 の遺伝子重複による発現の上昇がみられる ultra rapid metabolizer（UM）も存在する．2D6 の基質薬物には，β遮断薬（プロプラノロール，メトプロロール），抗うつ薬（ノルトリプチリン，フルオキセチン），抗不整脈薬（プロパフェノン）など治療域の狭い重要な薬物が多く，実際に PM，IM など遺伝子型に対応した血中クリアランスの変動がみられている[5,6]．また，向精神薬の副作用発現と 2D6

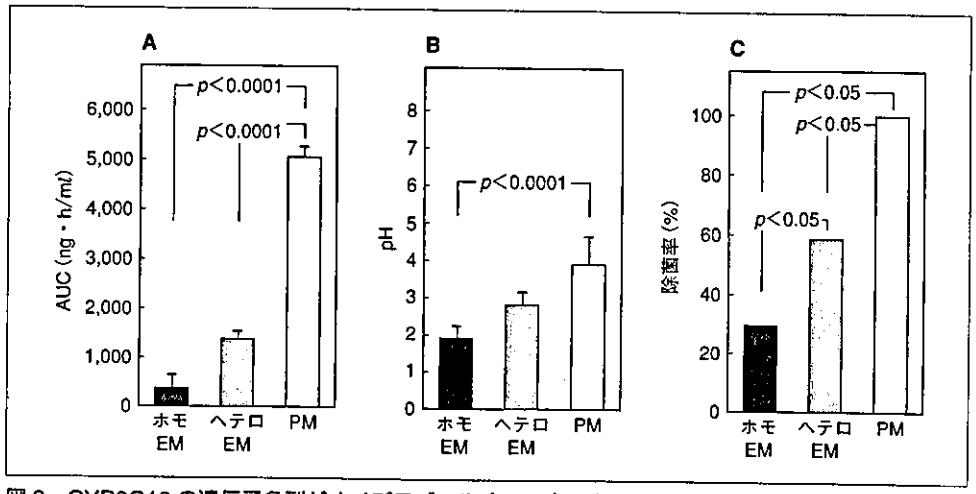

図2　CYP2C19の遺伝子多型がオメプラゾール（OMZ）に与える影響
A：OMZ 20 mg 経口投与時の AUC
B：OMZ 20 mg 経口投与後 24 時間の平均胃内 pH
C：OMZ 20 mg ＋アモキシシリン 2.0 g，2 週間投与後の *H.pylori* の除菌率
EM：extensive metabolizer，PM：poor metabolizer
（大河内秀昭：月刊薬事 43, 473-7, 2001[14] より）

の EM の割合との間にも相関が認められるとの報告がある[7]ことから，特に初期の投与量設定において多型診断は臨床上重要な役割を示すと考えられる．

CYP2C19 は，PM（主に*2, *3）の頻度は東洋人（20 %）が欧米人（10 %）より高い[8]．2C19 はプロトンポンプ阻害薬（PPI）を基質とすることから，2C19 の多型と，PPI の薬物動態・効果，また PPI は胃十二指腸潰瘍時の *H. pylori* 除菌療法に併用されるため除菌成績との関連が詳細に観察されている[9-13]．2C19 の PM では，オメプラゾール経口投与後の AUC（薬物血中濃度時間曲線下面積）が EM の 10 倍まで上昇する[11]．それに伴い，投与 24 時間後の平均胃内 pH の上昇も PM の方が有意に高い[11]．また，抗生物質アモキシシリンとの併用による除菌成績も PM の方が有意に高く，胃内 pH 上昇による抗生物質の安定化と *H. pylori* の増殖による感受性上昇によるものと考えられている[10]．実際に，オメプラゾールの投与量を増加すると除菌効率の上昇がみられること[12]，また 2C19 による代謝の寄与が比較的小さいラベプラゾールについては，上記のような変動は小さかったこと[9, 13]からも 2C19 の遺伝子型による PPI の投与設計が有効であると考えられる（図2）[14]．

CYP2C9 にも，*3 を保有する *1/*3 群が 2～3 %程度存在する．2C9 はきわめて治療域の狭い薬剤であるフェニトインやワルファリンの主要代謝酵素で，血中濃度の変動と中枢毒性や抗凝固能の変化といった重篤な副作用との相関が報告されており[15, 16]，多型の頻度は低いものの，遺伝子型による投与量の層別化が必要であろうと思われる．一方で，2C9 を主代謝経路にもつジクロフェナクは，*3 変異による代謝能の変化がみられないとの報告[17]があり，多型の影響が薬物により異なることが示唆されている．

2. CYP 以外の代謝酵素（抱合酵素）の遺伝子多型と薬物動態の個人差

1950 年代に，抗結核薬イソニアジドの *N*-

アセチル化代謝活性に個人差がみられることがすでに示唆されており，現在ではその原因がNAT（*N*-acetyltransferase）2にあることがわかっている．すなわち，通常は加水分解産物のヒドラジンがNAT2によりすみやかにアセチル化され解毒されるが，変異によりNAT2活性の低下があると，ヒドラジンが肝臓内に蓄積し，肝障害を発現すると考えられている[18]．また，抗不整脈薬プロカインアミドや潰瘍性大腸炎治療薬スルファサラジンについてはNAT2変異と全身性エリテマトーデス様の副作用発現の間に関連が認められている[19,20]．

CPT-11（カンプトテシン）の活性代謝物SN-38は，UGT（UDP-glucuronosyltransferase）1A1により肝臓で抱合を受け解毒・胆汁排泄される．UGTの変異は，もともと高ビリルビン血症であるCrigler-Najjar症候群およびGilbert症候群の解析から多数同定されてきた[21]．UGT1A1プロモーター領域のTAリピートが通常より1回多い変異では転写活性が半分に低下しており，UGTの活性低下からSN-38の消失遅延につながる可能性が考えられている[22]．また，TPMT（thiopurine *S*-methyltransferase）はプリン代謝拮抗薬メルカプトプリン（6-MP）の活性本体6-チオグアニンヌクレオチドの不活化を担っている酵素であり，特に*2，*3変異体は，発現量の低下による活性体の消失遅延，それに伴う骨髄抑制など重篤な副作用発現を引き起こす[23]．ドナーの赤血球ライセートと6-MPを混合して活性体の生成をみる方法もある[24]が，遺伝子多型判定が簡便になれば，TPMTの多型による投与量の層別化も今後可能になると思われる[25]．また，5-FU（5-fluorouracil）の代謝律速酵素であるDPD（dehydropyrimidine dehydrogenase）も活性の低下する多型があり，5-FUの神経毒性が低活性の人で有意に多くみられている[26]．

3. トランスポーターの遺伝子多型と薬物動態の個人差

近年，ヒトにおいても非常に多様なトランスポーターの発現が認められており，容易に膜透過できない化合物の濃縮的な取り込み・排出機構として働いていることが知られている．一般にトランスポーターは，きわめて広範な構造の化合物を認識することが知られており，それゆえ，トランスポーターの遺伝子多型による輸送活性，発現変動は，多くの基質化合物の輸送を変え，体内動態に影響を与えることが考えうる．

臨床において，遺伝子多型と薬物動態の個人差について関連解析が最も進んでいるのは，MDR1〔P-gp（P糖タンパク質），ABCB1〕であると思われる．MDR1は，肝臓の胆管側や小腸の管腔側，血液脳関門の血管側に発現が認められ，ATP水解活性を駆動力として細胞内からの排出を担うタンパク質である．現在では多くの遺伝子多型が同定されているが，なかでもアミノ酸変化を伴わない変異であるC3435Tは，興味深いことに，この変異の保有者の十二指腸におけるMDR1の発現量は有意に低く，経口投与したジゴキシンのAUCの上昇が認められている[27]．別の機能変化，発現変動を引き起こしうる変異との連鎖が想定され，実際，G2677（T, A），C1236Tとの間に連鎖不平衡が観察されている[28]．ただし*in vitro*発現系を用いた実験においては，2677，3435位の変異を考慮した多型体の機能解析では差異がみられないことが報告されている[29]．また連鎖不平衡ブロックの統計解析から，既知の変異の周辺40～80 kb以内に別の未知の変異がリンクする可能性が示唆されており[28]，今後の解析が待たれる．最近のハプロタイプ解析により，先に述べた3か所の変異保有者のみならず，7か所のイントロンのみの変異の保有者においてもSN-38の腎クリアランスの低下がみられており[30]，きわめて興味深い．しかし，多くの臨

床試験のなかで，先のジゴキシンの結果に矛盾する結果も報告されており[31]，今後情報の蓄積が望まれる．

また最近，肝臓に選択的な発現が認められる取り込みトランスポーターOATP2（SLC21A6, OATP1B1）についても多型に関して報告がされてきた．たとえば*in vitro*実験で，抗結核薬リファンピシンの輸送能力が多型により変動し，そのためPXR（pregnane X receptor）を介した転写誘導能にも影響を与える可能性を示唆する報告もある[32]．日本人においては，特に*15（Asn130Asp，Val174Ala）が約10％存在しており，HMG-CoA（hydroxymethylglutaryl-CoA）還元酵素阻害薬プラバスタチンの腎外クリアランス（すなわち肝クリアランス）が*15保有者で有意に低下することが臨床事例として初めて報告された（図3）[33]．一方，筆者らは*in vitro*実験系を用いて*15変異が単位タンパク質あたりの輸送量を低下させることを示し，腎外クリアランス低下の程度と半定量的に一致することを見出している（投稿中）．OATP2は基質認識性が非常に広範であることから，多様な薬物の肝クリアランスの第1ステップを担う取り込みトランスポーターの変異が体内動態に影響を与えうる可能性は十分に考えられ，今後のさらなる臨床試験が待たれる．

薬物の効果の感受性にかかわる分子の遺伝子多型

近年，多くの薬物に関して薬効の標的分子やその後のシグナル伝達，転写制御のカスケードが明らかになるにつれ，それらの遺伝子多型と効果との関連を示す報告が相次いでいる．特にレセプター分子や酵素との結合を介して効果を発揮する薬物にとって，直接相互作用する分子の遺伝子多型は薬効に影響を与える可能性が強く，解析が進んでいる．最近の主な事例を簡単に表1にまとめた[34-36]．

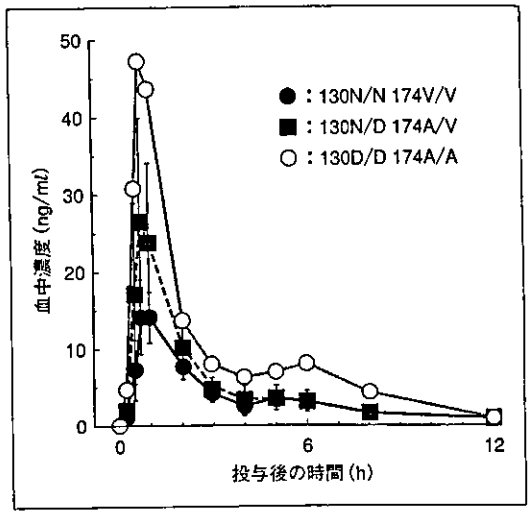

図3 OATP2遺伝子多型とプラバスタチン血中濃度推移との関連

（Nishizato Y, et al : *Clin Pharmacol Ther* 73, 554-65, 2003[33]より）

この領域で最も報告の多いβ_2アドレナリンレセプターの遺伝的多型は，特に頻度が高いArg16Gly，Gln27Gluの2か所について多くの検討がなされている[37]．*in vitro*によるイソプロテレノールを用いた検討では，Arg16Gly，Gln27Gluそれぞれ単独の変異体においては，レセプターと薬物の親和性やアデニル酸シクラーゼの活性には影響ないが，前者では脱感作の亢進がみられたのに対し，後者では逆にまったくみられなかった[38]．ヒトにおける変異解析の結果から，Arg16-Gln27，Gly16-Glu27の間で強い連鎖不平衡がみられ[39]，一方，Gly16-Glu27両方の変異を入れたタンパク質は，脱感作の亢進（Gly16の性質）がみられた[38]ことから，*in vitro*実験においてもハプロタイプを考慮した実験デザインの重要性がうかがえる．薬効との関連では，喘息患者にβ_2刺激薬アルブテロールを経口投与した後，血中濃度は多型により変化しないが，喘息の改善効果の指標となるFEV_1（forced expiratory volume in the first second）の値の変化は，Arg16/Arg16が，Gly16/Arg16，Gly16/Gly16群に比べて有意に大きな改善がみられている（図4）[40]．ほかに

表1　薬物の効果・副作用に遺伝子多型が関連しうる報告のある遺伝子群

遺伝子名	薬剤のカテゴリー・代表的薬物名	影響がみられる事象
1. 直接薬効ターゲットに関係する遺伝子		
アンジオテンシン変換酵素 (ACE)	ACE阻害薬 (エナラプリル)	血圧の低下，腎保護作用など
β_2アドレナリンレセプター	β_2刺激薬 (アルブテロール)	気管支拡張作用，心血管作用など
アラキドン酸5-リポキシゲナーゼ	ロイコトリエン阻害薬	FEV_1の改善
ドーパミンレセプター (D_2, D_3, D_4)	抗精神病薬 (プロムペリドール)	抗精神病作用
エストロゲンレセプターα	エストロゲン	骨密度上昇，HDL上昇など
グリコプロテインⅢaサブユニット	アスピリン，グリコプロテインⅡb/Ⅲa阻害薬	抗血小板作用
セロトニントランスポーター (5-HT)	抗うつ薬 (クロザピン)	抗うつ作用
アンジオテンシンレセプター (AT1R)	AT1R拮抗薬 (カンデサルタン)	腎血液循環改善作用
スルホニルウレアレセプター (SUR1)	SU系糖尿病薬 (トルブタミド)	インスリン分泌促進作用
ビタミンDレセプター	活性型ビタミンD_3	くる病改善効果
2. 副作用に関係する遺伝子		
ブラジキニンB2レセプター	ACE阻害薬 (エナラプリル)	(ACE阻害薬により誘発される) から咳
ドーパミンレセプター (D_2, D_3, D_4)	抗精神病薬 (プロムペリドール)	運動障害，静座不能
HLA	アバカビル (抗HIV薬)	過敏症反応
K^+チャネルおよび関連タンパク質 (HERG, KvLQT1, Mink, MiRP1)	エリスロマイシン，テルフェナジン，シサプリド，クラリスロマイシンなど	薬剤誘導性のtorsade de pointesのリスク上昇
プロトロンビンおよび第Ⅴ因子	経口避妊薬	血栓症のリスク上昇
過敏症反応	カルバマゼピン (抗痙攣薬)	過敏症反応
3. 間接的に効果に関係する遺伝子 (作用が明らかでないものも含む)		
アンジオテンシン変換酵素 (ACE)	HMG-CoA還元酵素阻害薬	血中LDL減少，動脈硬化進展など
adducin	利尿薬	心筋梗塞の危険因子
アポリポタンパク質E	タクリン (Alzheimer病治療薬)	病状の改善
	HMG-CoA還元酵素阻害薬	生存率上昇
コレステロールエステル転送タンパク質 (CETP)	HMG-CoA還元酵素阻害薬	動脈硬化進展の遅延
メチルグアニンメチル転移酵素 (MGMT)	カルムスチン (抗腫瘍薬)	gliomaに対する効果
Parkin	デボドパ	Parkinson病の治療効果改善
stromelysin-1	HMG-CoA還元酵素阻害薬	心血管イベントの減少など
リポタンパク質リパーゼ	HMG-CoA還元酵素阻害薬	動脈硬化進展の遅延
インターロイキン6	HMG-CoA還元酵素阻害薬	血中LDL減少
Toll-likeレセプター4	HMG-CoA還元酵素阻害薬	心血管イベントの減少など
レプチンレセプター	HMG-CoA還元酵素阻害薬	血中コレステロール低下

(Evans WE & McLeod HL : *N Engl J Med* 348, 538-49, 2003[34] を中心にまとめた)

もさまざまな臨床研究がなされているが，同じ指標で評価しているにもかかわらず，結果が互いに相反する事例が多くみられる．その原因の一つとして，多くの研究が単独のSNPを対象としており，対象集団のなかで連鎖する他の変異の影響を考慮していないために，的確な層別化が図れていない可能性が示唆される．実際，77人の13か所の変異についてハプロタイプ解析を行ったところ，わずか12種類に分類され，アルブテロール投与後のFEV_1の変化を層別化して評価したところ，単独のSNPとはいずれも相関が認められなかったのに対し，ハプロタイプ別の解析では有意な相関がみられたとする報告がある[39]．

また，先天的QT延長症候群の発症に関与する候補遺伝子の多型に関しては数多くの報告があり，タイプごとに適した治療法が提案されてきたが，一方，多種の薬剤について，薬剤誘導性のQT延長が臨床上重篤な副作用として問題視されている．現に，これが原因で上市されな