その位置が決定されている場合，その近くの多型マーカーを用いた連鎖解析を行うことができる．1例としては同胞対解析が挙げられる．マーカーと責任遺伝子の染色体上の距離により，一定の確率で連鎖に誤差が生じる．これは減数分裂時に組み換えが生じるからである．1回の減数分裂あたり，平均30ヵ所の組み換えが起こるとされており，ヒトゲノムを3×10^9塩基対とすれば10^8塩基に1ヵ所で組み換えが起こる．即ち1000kb(10^6bp)離れた2点の間では1%の可能性で組み換えが起きる．

遺伝子変異と易罹病性の一例として，感染症に対する易罹病性を挙げてみる．古くから知られている例であるが，HIV感染に際してのchemokine receptor-5(CCR-5)の関与である．このreceptorはCD4のco-receptorとして働くが，変異型CCR-5(32塩基対の欠失)が白人一般人口の中にも比較的高頻度にみられる．変異型は受容体機能を消失しており，HIV感染が成立しにくくなると予想される．実際，変異型CCR-5の対立遺伝子頻度は白人一般人口では9.2%である一方，変異型ホモ接合体の頻度はHIV感染者の中ではゼロであり，変異型ヘテロ接合体の頻度はHIV感染者で一般人口より有意に低かったという[2]．

動脈硬化の原因のひとつに軽度の高ホモシステイン血症が知られている．近年発表された2つのメタアナリシスにより，Methylene-tetrahydrofolate reductase(MTHFR)遺伝子C677T多型のホモ変異は，軽症高ホモシステイン血症を介して動脈硬化性疾患の独立した危険因子であることが明らかにされた．変異のアリル頻度は38%ともいわれ，一般人での軽症高ホモシステイン血症が問題となる．高血圧症や高脂血症と並んで冠状動脈イベントなどと関連する可能性がある．しかも軽症高ホモシステイン血症は葉酸などの摂取による治療が可能であり，臨床的意義は高いと考えられる．C677T変異によってこの酵素にAla→Val変異が起こり，活性が消失すると考えられている．共同研究者の宮木らはこの変異のgenotype毎に葉酸補充によるホモシステイン低下効果を検討した(**図1**)[3]．これはMTHFR遺伝子C677T多型で層別化した二重盲検プラセボ使用ランダム化比較試験で，介入としては葉酸1mgを含むカプセルおよび同形のプラセボカプセル1日1錠を経口投与90日間とした．その結果，どの多型においても介入1ヵ月後から実薬群で有意なホモシステイン低下が観察されたが(**図2**)[3]，その減少幅は野生型およびヘテロ変異体と比較してホモ変異体において最大で，統計的にも有意差があった(**図3**)[3]．本変異のホモ接合体(日本人一般集団での割合は10%以上)における低下量は変異のない人に比べ約2.4倍であることが1ヵ月後および3ヵ月後の2度にわたり定量的に示された．すなわち葉酸補充による恩恵を最も享受できる集団であることが示された[3]．

一方，遺伝子変異は薬物治療における効果や副作用の発現に関係することがあり，遺伝子検査が重要である．塩酸イリノテカン(CPT-11)はトポイソメラーゼIを阻害することにより消化器癌，肺小細胞癌に効果を示すが，UGT-1A1遺伝子多型の影響を受け，UGT-1A1*28を有する症例では薬の副作用が顕著になりやすい(**図4**)．このため「投与量を減量すること」がFDA勧告により薬剤添付文書に記載されることになった．また最近，抗凝固薬ワルファリン至適投与量を決定する因子が解析され，それを加味した適正使用量の推計式も考案されつつある．近年の研究で，個人個人のワルファリン投与量を規定する因子が詳しく研究され，年齢，性，体表面積，弁置換の有無に加えて遺伝子型(Cytochrome P450[CYP]2C9とVitamin K Epoxide Reductase Complex 1[VKORC1])がその決定要素として重要であると考えられており，これらを用いた至適量予測式も提案されている．これらの変数によりワルファリン投与量の50%以上が規定されるともいわれる(**図5**)．米国FDAでは2005年11月の会議(Clinical Pharmacology Subcommittee of the Advisory Committee for Pharmaceutical Science)において，“Current evidence related to the pharmacogenetics of warfarin as a potential basis for label updates”と題し，ワルファ

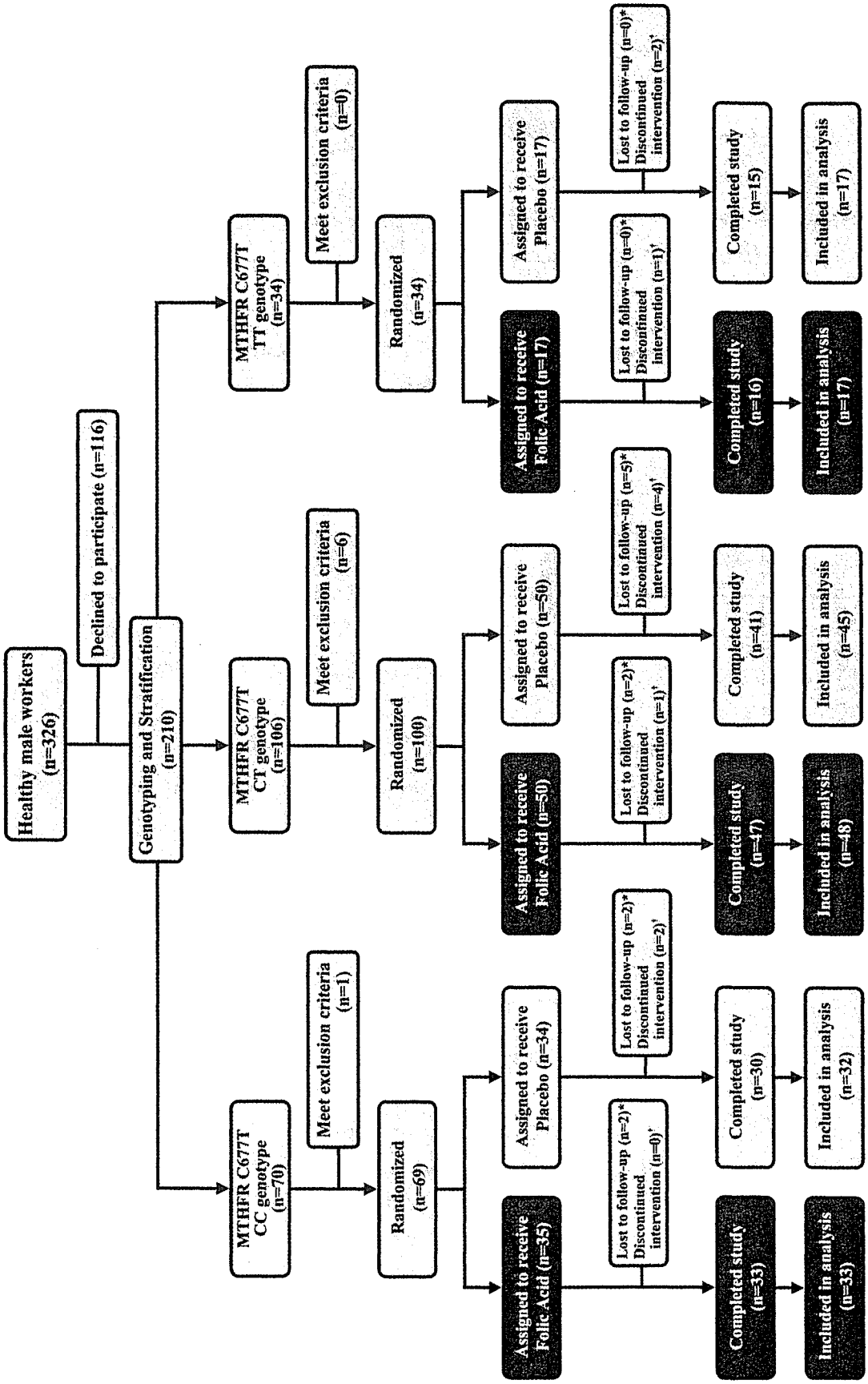

図1　Overall follow up rate after 3 months : 94.6%（CC : 92.8%, CT : 93.0%, TT : 100%）

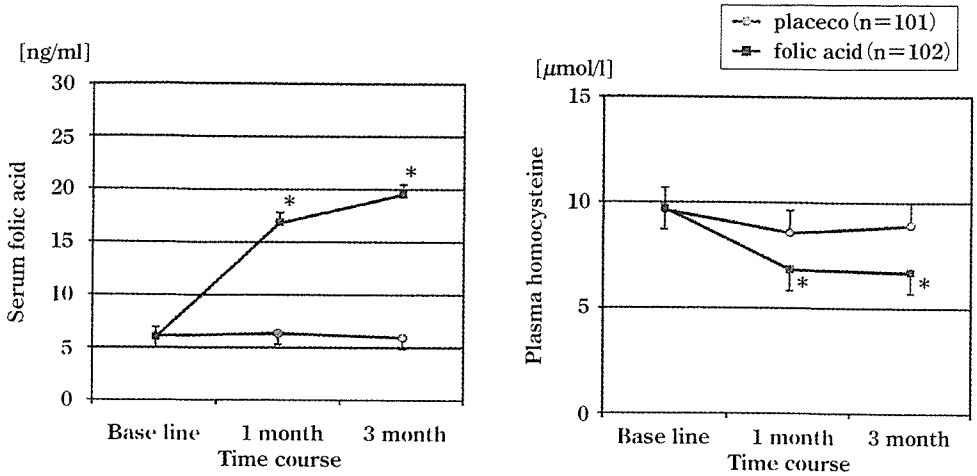

図2 Serum folic acid and plasma total homocysteine (tHcy) levels in folic acid and control groups

*p＜0.001 for the comparison with the placebo group. Mean and S. E. M. are shown.

J Hum Genet 2005; 50 (5) より転載

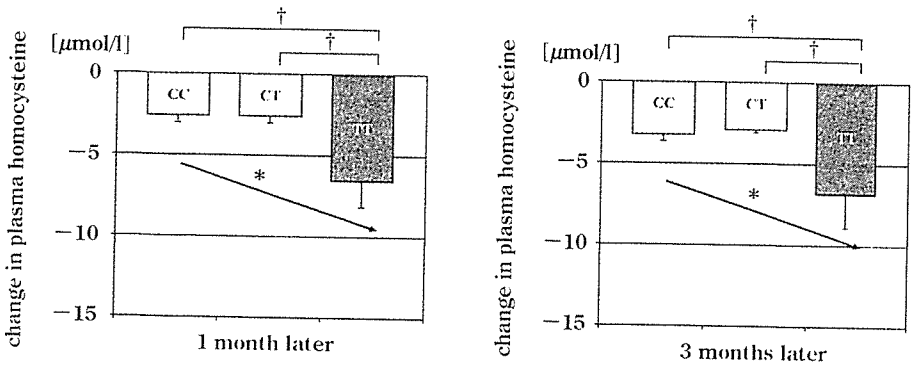

図3 Differences between MTHFR C677T genotypes in the decrease of plasma total homocysteine (tHcy) in the folic acid group

Only folic acid group is shown (Mean and SEM). * stand for p＜0.01 for trend test.

These are significant even after being adjusted with age, BMI, smoking status, and alcohol consumption. † stand for p＜0.01 for Tukey's post-hoc test of analysis of variance.

J Hum Genet 2005; 50 (5) より転載

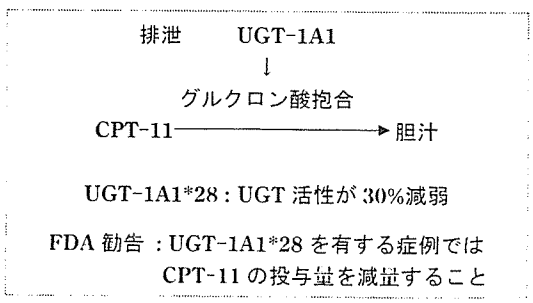

図4 塩酸イリノテカン (CPT-11)

(トポイソメラーゼⅠ阻害) 消化器癌，肺小細胞癌に効果

リン使用に際して個々の患者に遺伝子型のタイピングを行うよう添付文書に記載すべきか，が議論されている[4]．2006年2月に開催された日本血栓止血学会 Scientific Standardization committee (SSC)

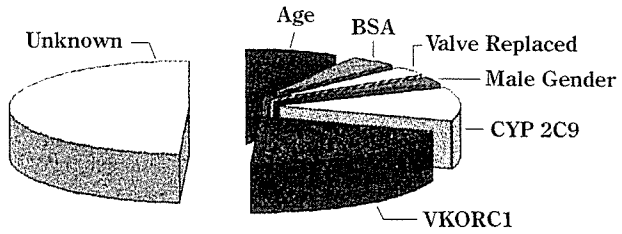

図5　Predicting the Stable Dose of Warfarin
Michael D. Caldwell. http://www.fda.gov/OHRMS/DOCKETS/AC/05/slides/2005-4194S1_05_Caldwell_files/frame.htm

会議でもこの問題が取り上げられワルファリン投与量と関係する幾つかの因子(VKORC1, CYP 2C9, GGCX)の日本人での SNP 探索とその結果ならびにワルファリン投与量との関連が報告され，また人種差について，VKORC1，CYP 2C9 の遺伝子型頻度が異なることが示されている．ワルファリン投与量が少量で済む VKORC1 1173T の頻度が日本人で非常に高いことも報告された．また INR 目標値に関する意識の違い，同じ INR でも日本人では本当に出血しやすいのか，などの臨床的エビデンスの充実が望まれる一方で，出血リスクを減らす意味での遺伝子型判定は重要，などの論議がなされた．

いわゆるテーラーメード医療のために，今後遺伝子検査が診療に必要となることは間違いないが，臨床現場でいつ行うべきか，遺伝学的検査の情報を如何に使用するか(発症予測，薬効予測，副作用予測など)，加えて社会的有用性(倫理性，医療費削減効果)も吟味される必要がある．

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To the editor:

Protein S–K196E mutation as a genetic risk factor for deep vein thrombosis in Japanese patients

Deep vein thrombosis (DVT) is a multifactorial disease caused by interactions between acquired risk factors and coagulation abnormalities.[1] In whites, the factor V–Leiden and the prothrombin-20210G>A are widely recognized as genetic risk factors for DVT. However, these 2 mutations are not present in Japanese populations, and little is known about the genetic risk factors for DVT in these populations. In this study, we evaluated the genetic contributions of 5 polymorphisms in Japanese DVT patients. The plasminogen-A620T mutation, formerly referred to as plasminogen-Tochigi, and the protein S–K196E mutation, formerly referred to as protein S–Tokushima, exhibited decreased activities of plasminogen and protein S despite normal antigen levels.[2-4] The ADAMTS13-P475S mutation exhibited low von Willebrand factor–cleaving activity in vitro.[5] The factor XII–4C>T substitution in the 5′-untranslated region, formerly referred to as 46C>T, showed decreased plasma levels of both antigen and activity.[6] The plasminogen activator inhibitor-1 (PAI-1) 4G/5G polymorphism is related to in vitro differences in transcription activity.[7] We genotyped subjects for these 5 polymorphisms and compared their genotypic frequencies between 161 DVT patients and 3655 population-based controls. The protocol for this study was approved by the ethical review committee, and only those subjects who provided written informed consent for genetic analyses were included in this study. All participants of this study were Japanese. The controls were from a general population randomly selected from the residents of Suita City located in the second largest urban area in Japan (the Suita Study).[8] One hundred sixty-one DVT patients, 78 men and 83 women, were registered by the Study Group of Research on Measures for Intractable Diseases, working under the auspices of the Ministry of Health, Labor, and Welfare of Japan. Six centers (Tochigi, Tokyo, Nagoya, Kyoto, and 2 in Osaka) participated in this study. The patients' mean age was 49.5 years (range, 12-87 years) and their mean body mass index was 23.6 ± 3.3. Thirteen percent of patients had a family history of thrombosis, and 16% of the patients had recurrent thrombosis.

Of all the polymorphisms tested, only the frequency of protein S–K196E was statistically different between the 2 groups ($\chi^2 = 38.3$, $P < .001$) (Table 1). No other frequency differences were statistically significant. Two DVT patients were homozygous for the protein S–196E allele; however, no homozygotes were identified in the control group. One patient with the 196EE genotype first developed DVT following surgery at age 47, while the other patient developed DVT during pregnancy at age 32.

The mutant protein S with the E allele has already been intensively studied as protein S–Tokushima.[11] The protein S mutant showed the reduced activated protein C cofactor activity compared with wild-type protein S, suggesting a direct link between the protein S–K196E mutation and the development of DVT. By the genotyping of the general population, the protein S–196E allele frequency was estimated as 0.009. Thus, a substantial portion of the Japanese population harbors this mutant allele and is at higher risk for DVT.

Table 1. Numbers and genotypic frequencies of protein S–K196E mutation in the DVT and control groups

Genotypes	General population, no. (%)	DVT group, no. (%)
Additive model*		
KK	3585 (98.2)	146 (90.7)
KE	66 (1.8)	13 (8.1)
EE	0 (0.0)	2 (1.2)
Total	3651 (100.0)	161 (100.0)
Dominant model†		
KK	3585 (98.2)	146 (90.7)
KE + EE	66 (1.8)	15 (9.3)
Total	3651 (100.0)	161 (100.0)

DNA genotyping was performed by the TaqMan allele discrimination method.[9] We have adopted the numbering standards of the Nomenclature Working Group, wherein the A of the ATG of the initiator Met codon is denoted as nucleotide + 1, and the initial Met residue is denoted as amino acid + 1, resulting in the renaming of several mutant alleles.[10] Comparisons between the DVT cases and the controls were analyzed using a χ^2 test with the genotypes as independent variables (indicated by P and OR) or using multiple logistic analyses with the genotypes as independent variables and age and sex as covariates (indicated by P' and OR′).

*For comparison of general population to DVT group, P was not determined.

†For comparison of general population to DVT group, $P < .001$; OR = 5.58 (3.11-10.01); $P' < .001$; OR′ = 4.72 (2.39-9.31).

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