Cancer Chemother Pharmacol (2001) 48 (Suppl 1): S102–S106
DOI 10.1007/s002800100315

Nagahiro Saijo · Tomohide Tamura
Noboru Yamamoto · Kazuto Nishio

New strategies for cancer therapy in the 21st century

Abstract The development of new anticancer drugs, radiation therapy devices, and surgical techniques has improved the survival and quality of life of cancer patients. Despite these advances, many adverse events prevent patients from receiving treatment in comfort, and a majority of patients die from recurrent disease. The limitations of treatment in terms of effectiveness and tolerability have driven researchers to develop new strategies to reduce treatment-related toxicity and improve the survival rates of cancer patients.

Keywords Pharmacogenetics · Pharmacogenomics

Promising target-based therapy

The anticancer drugs of the future will be relatively specific and nontoxic at clinically effective doses. Such agents are called target-based drugs because they will be rationally screened based on their molecular targets [7, 15, 18, 26, 33, 36, 40, 44, 46]. They can be classified into cytotoxic and noncytotoxic drugs. Cytotoxic drugs are currently being screened by the same strategy. Some noncytotoxic agents have been demonstrated to possess direct antitumor activity, therefore, making it difficult to differentiate between cytotoxic and noncytotoxic drugs. Methodologically, the newer treatments can be classified into small-molecule therapy, antibody therapy, gene therapy, and immunotherapy, some of which are tumor-specific and some nonspecific. Target-based drugs include signal transduction inhibitors such as receptor tyrosine kinase (RTK) inhibitors and farnesyl transferase inhibitors, CDK inhibitors, and angiogenesis inhibitors such as matrix metalloproteinase inhibitors and vascular endothelial growth factor (VEGF) inhibitors (Table 1) [1, 3, 32].

This work was presented at the 16th Bristol-Myers Squibb Nagoya International Cancer Treatment Symposium, "Hematologic malignancies: pioneers in cancer therapy across the century from mustard to molecular targets and beyond," 27–28 October 2000, Nagoya, Japan.

N. Saijo (✉) · T. Tamura · N. Yamamoto
Medical Oncology, National Cancer Center Hospital,
Tokyo, Japan
E-mail: nsaijo@gan2.ncc.go.jp
Tel.: +81-3-3542-2511
Fax: +81-3-3542-6220

K. Nishio
Pharmacology Division,
National Cancer Center Research Institute, Tokyo, Japan

Antibodies

Monoclonal antibodies (mAbs) for cancer therapy have undergone the transition from basic research to tools for clinical cancer treatment. In November 1997, the mouse/human chimeric anti-CD20 mAb, rituximab (Rituxan) became the first antibody to obtain approval from the US Food and Drug Administration for the treatment of B cell lymphoma [13, 14, 27]. In October 1998, the antibody product mouse/human chimeric anti-HER2 receptor mAb trastuzumab (Herceptin) was approved for the treatment of HER2-positive breast tumors. A randomized, controlled trial comparing chemotherapy alone vs chemotherapy plus trastuzumab demonstrated the superiority of the combined modality in terms of survival, time to progression, and response rate [4, 5, 9, 12, 31, 39].

The immunoglobulin G2a mAb 17-1A edrecolomab has been approved in Germany for the treatment of colorectal cancer in the adjuvant setting. Other promising antibodies under clinical evaluation include C225 anti-epidermal growth factor receptor (EGFR) mAb [6], anti-VEGF mAb [8, 38], and anti-GM2 or GD2 mAb [16, 20].

Tyrosine kinase inhibitors

RTKs, including many growth factor receptors, are transmembrane glycoproteins with a single membrane-

Table 1 Target-based therapy

Cytotoxic drug therapy	Topoisomerase inhibitors, tubulin binders, minor groove binders, antimetabolites, etc
Target-based therapy in the narrow sense (Small molecules)	
Tumor-specific noncytotoxic drugs	CDK inhibitors, FTI, TK inhibitor
Tumor-nonspecific noncytotoxic drugs	Angiogenesis inhibitors, TK inhibitor, matrix metalloproteinase inhibitors
(Macromolecule therapy)	Gene therapy (Ad p53), cytopathic virus (ONYX type)
	Immunotherapy (peptide Ag)
	Cytokine
	Antibody (trastuzumab)

spanning domain and a conserved cytoplasmic tyrosine kinase domain. There are 18 known families in vertebrates, comprising 56 receptors. The subfamilies of RTKs include the receptors for EGF, insulin, platelet-derived growth factor (PDGF), VEGF, fibroblast growth factor (FGF), hepatocyte growth factor (HGF), and the neurotropins, as well as a number of orphan receptors for which no ligands have been identified. RTKs and their downstream signaling molecules have emerged as important targets for the development of inhibitors because of their role in positive growth control [24, 25]. The degree of expression of many tyrosine kinases found in different tumor types has been associated with progression of cancer, increasing grade of malignancy, and a poorer prognosis.

SU-101 targets the PDGF signaling pathway, and in clinical trials in patients with glioblastoma multiforme it has been demonstrated to be metabolized rapidly to active compounds [17, 28]. The molecular pathways on which SU-101 acts remain unknown because of the difference in concentrations that inhibit molecular targets and tumor growth.

Sugen Co. (South San Francisco, Calif.) is developing two other compounds, SU-5416 [43] and SU-6668 [41]. SU-5416 is an angiogenesis inhibitor that targets the VEGF receptor, Flk-1, and is undergoing phase I, II, and III evaluations in various tumors. A phase I study of SU-6668 has recently been initiated. It is a more broadly targeted RTK inhibitor and has been reported to cause tumor shrinkage in animal experiments [23, 34].

ZD-1839 targets the EGF RTK. The EGF receptor is considered an important target because it is found in many tumors, including breast, ovarian, renal, and lung, and its overexpression frequently indicates a highly aggressive cancer. During global phase I trials of ZD-1839, at least seven partial responses have been recorded in non-small cell lung cancer among the 173 patients enrolled [11]. ZD-1839 is important because it is active against cisplatin- and/or taxane-refractory tumors. Phase III trials of standard chemotherapy with and without ZD-1839 in non-small cell lung cancer have been initiated in the USA and Europe. Two independent phase II studies of ZD-1839 alone have also been instituted for second- or third-line treatment.

The tyrosine kinase inhibitor STI-571, developed by Novartis (Basle, Switzerland) for the treatment of chronic myelogenous leukemia, is a bcr-abl tyrosine kinase inhibitor. Complete responses have been reported in a phase I study [42]. The concentration of STI-571 required to block tumor growth is much higher than that needed to shut down the target signaling pathways, suggesting that it also works through another unknown pathway.

Pharmacogenetics

Factors that influence the variability of drug toxicity and response can be divided into two categories: pharmacokinetics and pharmacodynamics. Pharmacokinetics, the relationship between time and plasma concentration, is influenced by: (1) dose, route, and schedule of administration; (2) metabolism (anabolism or catabolism); and (3) protein binding, which strongly influences drug distribution. Pharmacokinetically, metabolism is influenced by genetic factors regulating metabolic enzyme activity. Many anticancer drugs have a narrow therapeutic index, and anticancer drug doses are usually calculated on the basis of body surface area. However, there is considerable individual pharmacokinetic variation even when the dose is based on body surface area.

Drug metabolism is a major factor in pharmacokinetic variability. Cytochrome P450 (CYP3A4) is the enzyme responsible for the metabolism of a wide variety of compounds [30]. A number of isozymes of P450 are known to exist, and they have been classified into families and subfamilies. CYP3A4 is present in human liver microsomes and plays an important role in the metabolism of many anticancer drugs. It exhibits at least a five- to ten-fold interindividual variability in the disposition of the drugs, and thus it is important to determine its activity before administration to patients.

Three major noninvasive in vivo methods for estimation of interpatient variability of CYP3A4 activity have been reported [21, 22, 30]: the erythromycin breath test, the urinary dapsone recovery test, and measurement of the ratio of endogenous urinary 6-β-hydroxycortisol (6β-OHF) to free cortisol (FC) (6β-OHF/FC). However, none of these methods provides satisfactory estimates. Measurement of endogenous 6β-OHF/FC is considered the simplest and most practical method, and

while it has been reported to be able to assess intrapatient variability due to enzyme induction and inhibition, for example, estimation of interpatient variability is considered impossible. The reason for the difficulty has not been completely elucidated. One possibility is that the small amount of endogenous substrate does not reflect the actual activity of CYP3A4. We have hypothesized that administration of a large dose of exogenous cortisol would allow more precise estimation of interpatient variability of CYP3A4 activity [45].

A group of 30 patients with advanced non-small-cell lung cancer were enrolled in the study, and urinary 6β-OHF and FC were measured after administration of hydrocortisone 300 mg i.v. More than 2 days later, docetaxel 60 mg/m^2 i.v. was administered, followed by pharmacokinetic sampling. The correlation between docetaxel pharmacokinetics and the interpatient variability of CYP3A4 activity estimated by our method was assessed. After cortisol administration, 24-h urinary 6β-OHF (t6β-OHF) increased by about 60-fold compared with pretreatment levels to an average of $12{,}273 \pm 4{,}076$ mg/day (mean $\pm$ SD). Docetaxel clearance (CL) and area under the concentration-time curve averaged 24.5 ± 6.4 l/h/m^2 and 2.66 ± 0.91 mg/l, respectively, and an excellent correlation between docetaxel CL and t6β-OHF was observed ($r = 0.867$). Multivariate analysis revealed that t6β-OHF ($P < 0.001$), α-1-acid glycoprotein ($P < 0.004$), AST ($P = 0.007$), and age ($P = 0.022$) were significantly correlated with docetaxel CL. The interpatient variability of CYP3A4 activity and docetaxel CL were accurately predicted by measuring t6β-OHF after cortisol administration.

Recently, single-nucleotide polymorphisms (SNPs), a molecular technique for the detection of genetic polymorphisms, have become available. The SNPs of the genes encoding drug-metabolizing enzymes are being studied intensively. It is expected that the interpatient variability of drug-metabolizing enzyme activity at the gene level will be determined [10, 19, 29].

Pharmacogenomics

Genome-wide studies of gene expression have become possible due to the availability of all the sequences of protein-coding genes. Genome-wide expression studies are being carried out in various laboratories using DNA microarrays that are being applied for molecular evaluation not only of tumor cell lines but also of tumor tissues. DNA microarrays have shown that gene expression patterns in tumor cells are associated with biological and clinical characteristics [35, 37], and clusters of genes with similar function are regularly found that can be explained by their biological roles. Their patterns of expression are associated with proliferation, cytokine induction, and cell type. In studies using tumor tissue, the patterns of gene expression in tumor cells are also associated with those in nonmalignant cells, such as stromal and infiltrative lymphocytes. Some of these associations have been confirmed by immunohistochemistry using antibodies to the gene clusters. These procedures enable analysis of a much higher level of molecular events.

Specimens obtained by repeated biopsy and from primary and metastatic sites are reported to show a similar expression pattern, suggesting the validity of this technique. We analyzed the expression profiles of genes in lung cancer and neighboring normal lung tissue obtained from three lung cancer patients and found that their expression in cancer tissues was similar to that in normal tissue in the same patient, but there were large differences in lung cancer tissue from different patients. Increased expression of angiogenesis-related genes was observed in lung cancer tissue compared with normal lung tissue from the same patient [2]. Clustering of expression profiles revealed that angiogenesis-related genes can be classified into three groups. Studies of many tumor tissues may succeed in defining subclasses of tumors in different biological and clinical categories, and application of this technology may lead to an understanding of the function of gene

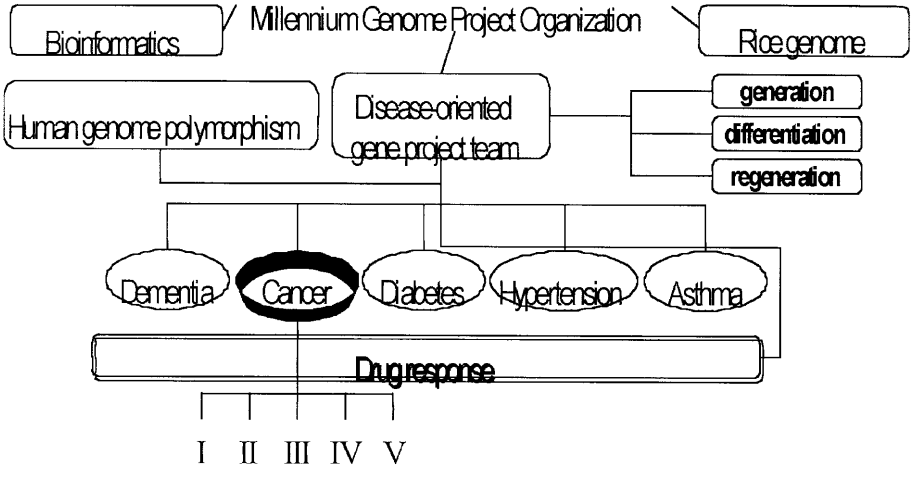

Fig. 1 The disease-oriented Japanese genome project

Table 2 The Japanese Millennium Project

Basic technology for genome research	Immortalization of small samples
	Gene amplifier
	SNP-based genome research
	High-throughput SNP typing
	Construction of clinical and genome databases
Guidelines for ethical aspects	Informed consent
	Genetic counseling
	Protection of privacy
	Establishing ethics committees
	Auditing
	Harmonization with the EU and USA
Identification of gene polymorphisms related to disease and clinical applications	Expression profiles
	Somatic mutations
Identification of gene polymorphisms related to PK/PD and clinical applications	Drugs: taxanes, irinotecan, fluorouracil, etc
	Genes: metabolism and transporters
	SNPs: CYP, DPD, etc
	Expression profiling
	Sample sources: peripheral lymphocytes immortalized by Epstein-Barr virus, animal models, rat liver cells, human primary hepatocytes
Individualization based on genome analysis	Lung, pancreas, gastric, colon, and esophageal carcinomas and genetic tumors
	Known cSNP analysis
	Whole genome association study
	Genomic analysis of animal carcinogenesis models

expression, provide a means of diagnosis, and identify targets for therapy.

Millennium Project in Japan

A disease-oriented genome project called the Millennium Project was inaugurated in Japan in 2000 (Fig. 1). One of the issues addressed under this project is cancer, which is divided into the five projects shown in Table 2. The author's group will work on the fourth project, "Identification of genomic polymorphisms related to pharmacokinetics/pharmacodynamics and their clinical applications". Genes encoding drug-metabolizing enzymes for the taxanes, irinotecan and fluorouracil and drug transporters will be analyzed. Changes in genes, such as in expression and mutations, will be evaluated for their correlation with pharmacokinetics and pharmacodynamics. The samples for genetic analyses will be peripheral lymphocytes, rat liver cells, human primary hepatocytes, etc. The techniques used for pharmacogenetic and pharmacogenomic analyses will be SNPs and cDNA expression arrays. We will use these techniques to try to identify the genes governing drug sensitivity and resistance in the hope of finding new molecular targets for chemotherapy and to develop order-made, tailor-made, customized, individualized, and personalized therapy.

References

1. Adjei AA (2000) Signal transduction pathway targets for anticancer drug discovery. Curr Pharm Res 6:361
2. Akutagawa S, Nakamura T, Usuda J, Fukumoto H, Koh Y, Tamura T, Saijo N, Nishio K (2000) Differential gene expression in human UCN-01-resistant small cell lung cancer cells. Proc 91st Am Assoc Cancer Res 41:31 (A203)
3. Arap W, Pasqualini R, Ruoslahti E (1998) Chemotherapy targeted to tumor vasculature. Curr Opin Oncol 10:560
4. Baselga J, Tripathy D, Mendelsohn J, Baughman S, Benz CC, Dantis L, Sklarin NT, Seidman AD, Hudis CA, Moore J, Rosen PP, Twaddell T, Henderson IC, Norton L (1996) Phase II study of weekly intravenous recombinant humanized anti-p185HER2 monoclonal antibody in patients with HER2/neu-overexpressing metastatic breast cancer. J Clin Oncol 14:737
5. Baselga J, Tripathy D, Mendelsohn J, Baughman S, Benz CC, Dantis L, Sklarin NT, Seidman AD, Hudis CA, Moore J, Rosen PP, Twaddell T, Henderson IC, Norton L (1999) Phase II study of weekly intravenous trastuzumab (Herceptin) in patients with HER2/neu-overexpressing metastatic breast cancer. Semin Oncol 26:78
6. Baselga J, Pfister D, Cooper MR, Cohen R, Burtness B, Bos M, D'Andrea G, Seidman A, Norton L, Gunnett K, Falcey J, Anderson V, Waksal H, Mendelsohn J (2000) Phase I studies of anti-epidermal growth factor receptor chimeric antibody C225 alone and in combination with cisplatin. J Clin Oncol 18:904
7. Boral AL, Dessain S, Chabner BA (1998) Clinical evaluation of biologically targeted drugs: obstacles and opportunities. Cancer Chemother Pharmacol 42:S3
8. Brekken RA, Overholser JP, Stastny VA, Waltenberger J, Minna JD, Thorpe PE (2000) Selective inhibition of vascular endothelial growth factor (VEGF) receptor 2 (KDR/Flk-1) activity by a monoclonal anti-VEGF antibody blocks tumor growth in mice. Cancer Res 60:5117
9. Burris HA III (2000) Docetaxel (Taxotere) in HER-2-positive patients and in combination with trastuzumab (Herceptin). Semin Oncol 27:19
10. Cardon LR, Idury RM, Harris TJ, Witte JS, Elston RC (2000) Testing drug response in the presence of genetic information: sampling issues for clinical trials. Pharmacogenetics 10:503
11. Ciardiello F, Caputo R, Bianco R, Damiano V, Pomatico G, De Placido S, Bianco AR, Tortora G (2000) Antitumor effect and potentiation of cytotoxic drugs activity in human cancer cells by ZD-1839 (Iressa), an epidermal growth factor receptor-selective tyrosine kinase inhibitor. Clin Cancer Res 6:2053
12. Cobleigh MA, Vogel CL, Tripathy D, Robert NJ, Scholl S, Fehrenbacher L, Wolter JM, Paton V, Shak S, Lieberman G, Slamon DJ (1999) Multinational study of the efficacy and safety of humanized anti-HER2 monoclonal antibody in women who have HER2-overexpressing metastatic breast

cancer that has progressed after chemotherapy for metastatic disease. J Clin Oncol 17:2639
13. Davis TA, White CA, Grillo-Lopez AJ, Velasquez WS, Link B, Maloney DG, Dillman RO, Williams ME, Mohrbacher A, Weaver R, Dowden S, Levy R (1999) Single-agent monoclonal antibody efficacy in bulky non-Hodgkin's lymphoma: results of a phase II trial of rituximab. J Clin Oncol 17:1851
14. Davis TA, Grillo-Lopez AJ, White CA, McLaughlin P, Czuczman MS, Link BK, Maloney DG, Weaver RL, Rosenberg J, Levy R (2000) Rituximab anti-CD20 monoclonal antibody therapy in non-Hodgkin's lymphoma: safety and efficacy of re-treatment. J Clin Oncol 18:3135
15. Ferrante K, Winograd B, Canetta R (1999) Promising new developments in cancer chemotherapy. Cancer Chemother Pharmacol 43:S61
16. Foon KA, Lutzky J, Baral RN, Yannelli JR, Hutchins L, Teitelbaum A, Kashala OL, Das R, Garrison J, Reisfeld RA, Bhattacharya-Chatterjee M (2000) Clinical and immune responses in advanced melanoma patients immunized with an anti-idiotype antibody mimicking disialoganglioside GD2. J Clin Oncol 18:376
17. Garber K (2000) Tyrosine kinase inhibitor research presses on despite halted clinical trials. J Natl Cancer Inst 92:967
18. Gibbs JB (2000) Mechanism-based target identification and drug discovery in cancer research. Science 287:1969
19. Gray IC, Campbell DA, Spurr NK (2000) Single nucleotide polymorphisms as tools in human genetics. Hum Mol Genet 9:2403
20. Hanai N, Nakamura K, Shitara K (2000) Recombinant antibodies against ganglioside expressed on tumor cells. Cancer Chemother Pharmacol 46:S13
21. Hunt CM, Watkins PB, Saenger P, Stave GM, Barlascini N, Watlington CO, Wright JT Jr, Guzelian PS (1992) Heterogeneity of CYP3A isoforms metabolizing erythromycin and cortisol. Clin Pharmacol Ther 51:18
22. Kinirons MT, O'Shea D, Downing TE, Fitzwilliam AT, Joellenbeck L, Groopman JD, Wilkinson GR, Wood AJ (1993) Absence of correlations among three putative in vivo probes of human cytochrome P4503A activity in young healthy men. Clin Pharmacol Ther 54:621
23. Laird AD, Vajkoczy P, Shawver LK, Thurnher A, Liang C, Mohammadi M, Schlessinger J, Ullrich A, Hubbard SR, Blake RA, Fong TA, Strawn LM, Sun L, Tang C, Hawtin R, Tang F, Shenoy N, Hirth KP, McMahon G, Cherrington V (2000) SU6668 is a potent antiangiogenic and antitumor agent that induces regression of established tumors. Cancer Res 60:4152
24. Levitzki A (1999) Protein tyrosine kinase inhibitors as novel therapeutic agents. Pharmacol Ther 82:231
25. Levitzki A, Gazit A (1995) Tyrosine kinase inhibition: an approach to drug development. Science 267:1782
26. Lush RM, Rudek MA, Figg WD (1999) Review of three new agents that target angiogenesis, matrix metalloproteinases, and cyclin-dependent kinases. Cancer Control 6:459
27. Maloney DG, Grillo-Lopez AJ, White CA, Bodkin D, Schilder RJ, Neidhart JA, Janakiraman N, Foon KA, Liles TM, Dallaire BK, Wey K, Royston I, Davis T, Levy R (1997) IDEC-C2B8 (Rituximab) anti-CD20 monoclonal antibody therapy in patients with relapsed low-grade non-Hodgkin's lymphoma. Blood 90:2188
28. Marx GM, McCowatt S, Boyle F, Pavlakis N, Levi JA, Bell DR, Freilich R, Cook R, Biggs M, Little N, Wheeler HR (2000) Phase II study of thalidomide as an anti-angiogenic agent in the treatment of recurrent glioblastoma multiforme (GBM). Proc Am Soc Clin Oncol 19:A613
29. McCarthy JJ, Hilfiker R (2000) The use of single-nucleotide polymorphism maps in pharmacogenomics. Nat Biotechnol 18:505
30. Parkinson A (1996) An overview of current cytochrome P450 technology for assessing the safety and efficacy of new materials. Toxicol Pathol 24:48
31. Pegram MD, Lipton A, Hayes DF, Weber BL, Baselga JM, Tripathy D, Baly D, Baughman SA, Twaddell T, Glaspy JA, Slamon DJ (1998) Phase II study of receptor-enhanced chemosensitivity using recombinant humanized anti-p185HER2/neu monoclonal antibody plus cisplatin in patients with HER2/neu-overexpressing metastatic breast cancer refractory to chemotherapy treatment. J Clin Oncol 16:2659
32. Powis G (1994) Signalling pathways as targets for anticancer drug development. Pharmacol Ther 62:57
33. Powis G (1995) Anticancer drugs acting against signaling pathways. Curr Opin Oncol 7:554
34. Rosen L, Hannah A, Rosen P, Kabbinavar F, Mulay M, Gicanov N, DePaoli A, Cropp G, Mabry M (2000) Phase I dose-escalating trial of oral SU006668, a novel multiple receptor tyrosine kinase inhibitor in patients with selected advanced malignancies. Proc Am Soc Clin Oncol 19:A708
35. Ross DT, Scherf U, Eisen MB, Perou CM, Rees C, Spellman P, Iyer V, Jeffrey SS, Van de Rijn M, Waltham M, Pergamenschikov A, Lee JC, Lashkari D, Shalon D, Myers TG, Weinstein JN, Botstein D, Brown PO (2000) Systematic variation in gene expression patterns in human cancer cell lines. Nat Genet 24:227
36. Saijo N, Tamura T, Nishio K (2000) Problems in the development of target-based drugs. Cancer Chemother Pharmacol 46:S43
37. Scherf U, Ross DT, Waltham M, Smith LH, Lee JK, Tanabe L, Kohn KW, Reinhold WC, Myers TG, Andrews DT, Scudiero DA, Eisen MB, Sausville EA, Pommier Y, Botstein D, Brown PO, Weinstein JN (2000) A gene expression database for the molecular pharmacology of cancer. Nat Genet 24:236
38. Schlaeppi JM, Wood JM (1999) Targeting vascular endothelial growth factor (VEGF) for anti-tumor therapy, by anti-VEGF neutralizing monoclonal antibodies or by VEGF receptor tyrosine-kinase inhibitors. Cancer Metastasis Rev 18:473
39. Shak S (1999) Overview of the trastuzumab (Herceptin) anti-HER2 monoclonal antibody clinical program in HER2-overexpressing metastatic breast cancer. Herceptin Multinational Investigator Study Group. Semin Oncol 26:71
40. Stadler WM, Ratain MJ (2000) Development of target-based antineoplastic agents. Invest New Drugs 18:7
41. Stopeck A (2000) Results of a phase I dose-escalating study of the antiangiogenic agent, SU5416, in patients with advanced malignancies. Proc Am Soc Clin Oncol 19:A820
42. Talpaz M, Sawyers CL, Kantarjain H, Resta D, Reese SF, Ford J, Druker BJ (2000) Activity of an ABL specific tyrosine kinase inhibitor in patients with BCR-ABL positive acute leukemias, including chronic myelogenous leukemia in blast crisis. Proc Am Soc Clin Oncol 19:A6
43. Via LE, Gore-Langton RE, Pluda JM (2000) Clinical trials referral resource. Current clinical trials administering the antiangiogenesis agent SU5416. Oncology 14:1312
44. Weinstein JN, Buolamwini JK (2000) Molecular targets in cancer drug discovery: cell-based profiling. Curr Pharmacol Res 6:473
45. Yamamoto N, Tamura T, Kamiya Y, Sekine I, Kunitoh H, Saijo N (2000) Correlation between docetaxel clearance and estimated cytochrome P450 activity by urinary metabolite of exogenous cortisol. J Clin Oncol 18:2301
46. Zunino F, Capranico G, Pratesi G, Spinelli S (1992) Current approaches to new drug development in cancer chemotherapy. Farmaco 47:1115