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Drug Safety Evaluation in the United States of America

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Abstract. General steps in the discovery and development of novel drugs in the United States are presented. The first step is the discovery of novel drugs. Brief histories and mechanisms of a few novel drugs in the American market are outlined. In this presentation, preclinical animal toxicologic studies(drug safety evaluation) are emphasized in regard to drug development. When preclinical animal studies have defined the toxicity and the doses at which it occurs, an Investigational New Drug Application (IND) is submitted to the Food and Drug Administration (FDA). An IND notifies the FDA the intention to begin testing a novel drug in human subjects.

INTRODUCTION

It is difficult to predict how long it will take and how much it will cost to discover and develop a novel drug. In some cases, hundreds or even thousands of compounds are tested before the search is abandoned or before a satisfactory one is found. Total research and development expenditures by research-based pharmaceutical firms in the U.S.A. are projected to reach $ 15.0 billion (approximately 21.2% of gross sales) in 1997, up from $ 13.4 billion (approximately 19.9% of gross sales) in 1996 and $ 11.9 billion (19.4% of gross sales) in 1995[6]. Surveys[6] in 1996 found that parmaceutical manufacturers in the U.S.A. spent an average of $ 304 million and 14.7 years to develop a new drug from chemical synthesis to New Drug Application (NDA) approval by FDA. U.S. FDA spent 21.6 months of average review time for each NDA and approved 131 NDAs in 1996[6]. Drugs must be effective with no or minimal side effects. A novel drug reaches the market through two big steps: discovery and development including drug safety evaluation.

(A) DRUG DISCOVERY

Biological problems are like mazes: there are always unerring pathways to the right answer: but they may be difficult to find. A novel drug can be discovered through tireless research using hit-or-miss technology to delve into the biological mazes, or by rationally targeting a site, such as an enzyme, receptor, membrane channel, or abnormal gene that can be manipulated to alleviate or cure the disease. Because of highly advancing sciences including combinatorial chemistry, molecular biology, biochemistry, and immunology, drugs in the near future will be rationally designed and developed rather than by hit-or-miss techniques

The finding of lowering blood pressure with the venom of a poisonous Brazilian snake inspired the development of the first angiotensin I converting enzyme inhibitor, captopril, for the treatment of hypertension in the early 1970's. Since then, many angiotensin I converting enzyme inhibitors have been developed. These inhibitors have proven to be very effective drugs for the treatment of hypertension and have prolonged and saved the lives of millions of people. In 1982, it

was reported that derivatives of imidazole-5-acetic acid attenuated vasoconstriction induced by angiotensin II. Since then, a clever series of stepwise molecular modifications of the compound had led to the development of an orally active, potent, and selective nonpeptide angiotensin II receptor antagonist which was marketed in 1995. In the last two years, several other nonpeptide angiotensin II receptor antagonists have been developed as very effective antihypertensive drugs.

HMG-CoA reductase catalyzes the conversion of HMG-CoA to mevalonate, which is an early and rate-limiting step in the biosynthesis of cholesterol in the cytosol of hepatocytes. The first HMC-CoA reductase inhibitor was isolated from cultures of Penicillium species in 1976. Since then, several chemically modified and synthetic HMC-CoA reductase inhibitors have been developed for the treatment of high cholesterol. Squalene is also an intermediate product as well as HMG-CoA in the pathway of cholesterol synthesis. As far as inhibitors of HMG-CoA reductase are effective cholesterol-lowering agents in animals and in man, inhibitors of squalene synthase should also serve as cholesterol-lowering agents. Therefore, squalene synthase inhibitors have become the next target for pharmacological intervention to achieve cholesterol lowering therapy. However, to date, no company has achieved success in developing a squalene-synthase inhibitor as a drug.

Genetic diseases had been traditionally treated by intervening in the biological events resulting from aberrant genes, but not by altering the aberrant genes. For the last several years, many fierce attempts on gene therapies by correcting the aberrant genes have been made. Most attempts added useful genes into selected cells to compensate for a missing or inefficient version or to instill some entirely new property via vectors (viruses, liposomes, etc.) delivering therapeutic genes by *in* or *ex vivo* techniques. In one earlier approach, called "antisense therapy", short stretches of synthetic DNA act on mRNA transcripts of aberrant genes to prevent the transcripts from being translated into abnormal proteins. A similar tactic utilizes small RNA molecules called "ribozymes" to degrade mRNAs that are copied from aberrant genes. A rather different effort provides a gene for a protein, called an "intracellular antibody", that can block the activity of the mutant protein itself. Some other therapeutic strategies rely on the design of hybrids of DNA and RNA that might direct the repair of mutant gene. Modern genetic researchs are moving from gene sequencing and mapping into functional or protein-based genomics (organizing massive amounts of information about what these genes do and what goes wrong with them in the disease state). Many researchers are looking for promising opportunities to correlate genetics with various genetic and non-genetic diseases and response to drugs. Continuous tireless research on genomics will hopefully lead to the treatment and prevention of genetic and even non-genetic diseases in the future.

(B) DRUG DEVELOPMENT

The FDA oversees the complex and demanding approval process to ensure that only safe and effective drugs can reach the American market. The developmental process of a new drug is, in many ways, different for each new drug. However, the general process consists of two principal steps: preclinical animal studies and clinical trials in human subjects.

(1) Preclinical Animal Studies

Before new drugs are administered to human subjects, preclinical animal studies are undertaken to determine the pharmacologic effects and safety in laboratory animals, in compliance with the Good Laboratory Practice(GLP) Regulations of FDA. The studies involve years of work and include many different types of studies. However, the testing is generally divided into two areas of

pharmacology and toxicology, and pharmacology work is generally conducted first.

(a) Pharmacologic Studies

Pharmacologic studies are performed to determine in vivo efficacy, mechanism of the action, duration of the action, dose-response relationship, and pharmacodynamics. In addition, pharmcokinetics are thoroughly studied to determine the absorption, distribution, metabolism, and excretion of the drug.

(b) Toxicologic Studies (Drug Safety Evaluation)

The primary goal of toxicologic studies (drug safety evaluation) in laboratory amimals is to measure the safety of the drug. Toxicologic studies include genotoxicity, acute, subacute, chronic, carcinogenicity, reproductive, and irritation studies. Depending on the nature of the drug and the proposed duration of use in humans, appropriate studies are conducted.

Genotoxicity studies are required by the FDA. These involve a battery of *in vivo* and *in vitro* tests designed to determine if the compound can cause any gene, chromosome, or DNA damage. The International Conference on Harmonization(ICH) has recommended the following standard test battery for genotoxicity: a) a test for gene mutation in bacteria; b) an *in vitro* test with cytogenetic evaluation of chromosomal damage with mammalian cells or an *in vitro* mouse lymphoma *tk* assay; and c) *in vivo* test for chromosomal damage using rodent hematopoietic cells[7].

Acute toxicity is the toxicity produced by a pharmaceutical when it is administered in one or more doses during a period not exceeding 24 hours. The information obtained from these studies is useful in choosing doses for repeat-dose studies, providing preliminary identification of target organs of toxicity, and occasionally, revealing delayed toxicity. These studies may also aid in the selection of starting doses for Phase 1 human studies, and provide information relevant to acute overdosing in humans. Generally, acute toxicity studies are designed to determine the short-term toxicity by administering a single dose to at least five rodents per sex. In acute studies, after the administration of a single dose, overt clinical signs are observed for at least two weeks. Gross pathologic examinations are conducted on all animals that die or are sacrificed in poor condition and on all control and high-dose animals after the two-week observation. If 50% or more of high-dose group are sacrificed for gross pathologic examinations. When drug-related gross lesions are found in high-dose animals, all target organs of toxicities from all treated animals are microscopically examined.

Subacute toxicity studies are designed to determine the toxicity in animals administered the drug for at least 2 weeks through 90 days.

Chronic toxicity and carcinogenicity studies are required for drugs that are used for a long-term period, exhibit any genotoxicity, or have some suspected chemical structures as carcinogens. The duration of the most common chronic toxicity studies are six months and one year. The FDA generally requires carcinogenicity studies of at least two years in the rat and at least 18 months in mice. Carcinogenicity studies are usually conducted at the same time chronic toxicity tests are underway. The animals used in chronic studies are sacrificed at appropriate intervals and important tissues are carefully examined histopathologically.

In subacute and chronic toxicity studies, the test compound is usually administered daily to at least one rodent (rats or mice) and one non-rodent (dogs or monkeys) species at three or more dose levels by the route that will be used in human subjects. In these studies, the high dose is determined as the maximum tolerated dose.

The low dose should not produce any toxicity. The intermediate dose is generally measured as the geometric mean of the high and low dose levels. Every study has control animals administered vehicle only. Controls are treated in exactly the same fashion as treated animals. The total number of animals of each sex of each dose in each study must be statistically sufficient, and generally larger numbers of animals are used for longer-term studies to allow for mortality. Usually, the total number of animals is about 80 to 100 rodents and 16 to 30 non-rodents in a chronic study, and 500 to 800 rodents in a carcinogenicity study. Each study generally includes blood chemistry, electrolytes, urinalysis, toxicokinetics, organ weights, gross pathology, microscopic pathology, and sometimes electron-microscopic pathology. The toxicity of each compound is determined by the statistical and/or biological significance of the data. All of the repeat-dose toxicity studies generally have a toxicokinetics component to evaluate systemic drug exposure and to correlate this information with the toxicologic findings.

Reproductive and teratologic studies are primarily designed for durg to be used in women, but are usually conducted for most drugs. Rats, mice, or rabbits are used for these studies. Reproduction testing includes three different types of studies, designated segment I, II and III studies. Rats are the most commonly accepted species for segment I studies. In segment I studies, drugs are administered to virgin males from the beginning of spermatogenesis (from 42 days old in male rats for 28 days before mating), to females for two estrous cycle (14 days in female rats) before mating, and through the gestation period. Examinations involve the investigation of spermatogenesis, oogenesis, genital function, mating behavior, estrous cycle, conception rate, gestation period, parturition, and lactation of the first generation. Based on the practice of concurrently evaluating male and female rodents in the same Segment I study, Segment I studies must be completed prior to Phase III clinical trial[8]. In segment II studies, the drug is administered to pregnant animals (usually rat and rabbits) during the period of organogenesis (late two-third period of gestation). This study is designed to measure the embryo-toxicity and teratogenic effects of drug, and must be completed prior to any outpatient trial that includes women of child-bearing potential[8]. The segment III study is designed to determine potential perinatal and postnatal effects of drugs. The drug is administered to pregnant animals from the final one-third of gestation through the weaning period. Examinations are concentrated on late fetal development, labor, delivery, lactation, neonatal viability, and growth of the newborn. Segment III studies must be generally completed prior to final registration. If pediatric trials are included in Phase III clinical trials, Segment III studies must be completed prior to the start of the pediatric trials[8].

When preclinical animal studies, that are similar in duration to the desired length of clinical treatment on human subjects with the new drug, are satisfactorily completed, generally the new drug can be tested on human subjects[8]. Then, the pharmaceutical company can propose to FDA to test the new drug in human subjects. This proposal includes all data from the completed preclinical animal tests, detailed information on the drug, and detailed protocols for clinical studies. This proposal is called the IND. If the FDA has any concerns about the proposal, the agency contacts the company to delay clinical studies. If the company is not notified by the FDA within 30 days after submitting the IND, the company can initiate clinical studies on humans.

(2) Clinical Trials

Clinical trials proceed strictly and sequentially in at least three different phases according to the

protocols submitted to the FDA. Phase I clinical trials focus primarily on the safety of the new drug in human subject. In these studies, the drug is administered to 20-80 normal humans, and possibly some mild but stable patients. Phase II clinical trials are designed to detect pharmacological efficacy in addition to safety in 100 to 200 human patients (chronic animal studies generally begin at this point). Phase III clinical trials are designed to gather broader information, including the dosages, in addition to safety and pharmacologic efficacy in larger population of patients ranging from several hundred to several thousand. In these three types of studies, a relatively complete investigation is conducted on the pharmacologic efficacy, safety, and dosages for humans including the mechanism of pharmacologic action, pharmacokinetics, pharmacodynamic, and side effects. The results of these three clinical phases provide the overall adequate basis for physician labeling.

With the satisfactory completion of all preclinical animal studies and clinical studies, the pharmaceutical company submit the NDA. The NDA is the vehicle through which the pharmaceutical company formally requests the FDA to approve a new drug for marketing in the United States. One NDA is composed of thousands of pages of preclinical and clinical data, drug chemistry details, and manufacturing descriptions to show that the drug is safe and effective in its proposed use or indication. The time for reviewing an NDA according to the Federal Food, Drug and Cosmetic Act is 180 days, but historically many drugs have required much longer time, reaching 3 to 5 years. With the introduction of user's fees, the review time has been cut substantially, to one year for conventional drugs and six months for life-saving therapies.

The pharmaceutical company must comply with Good Manufacturing Practices of the FDA in manufacturing the new drug, and after regulatory review and approval, the novel drug reaches the market. After a new drug is in the market, the post-marketing surveillance program may include post-approval trials are not required by the FDA, but most pharmaceutical companies voluntarily initiate these studies to gain more data which can further prove safety and efficacy of their drugs, and can contribute to some unexpected competitive advantage of their products.

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