1 AGENT IDENTIFICATION AND PRECLINICAL TESTING

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INTRODUCTION

This chapter provides an overview of the preclinical screening assays and animal efficacy testing models currently utilized by the drug discovery and development program of the Division of Cancer Prevention (DCP), National Cancer Institute (NCI) to identify chemical agents or natural biological products which may be efficacious in preventing human cancers. The first step of the drug discovery process involves subjecting candidate agents to a sequential series of mechanism-based assays which target specific biochemical and molecular pathways known to be involved in carcinogenesis. These in vitro mechanistic assays provide quantitative data used to establish chemopreventive efficacy and also to assist in classifying and prioritizing agents for further evaluation in whole animal models. Major classes of chemopreventive agents identified thus far include, for example, signal transduction modulators, such as Ras farnesyl transferase inhibitors; retinoids; protein tyrosine kinase inhibitors (PTK), such as epidermal growth factor receptor (EGFR) inhibitors; peroxisome proliferator-activated receptor (PPAR) modulators; hormone modulatory agents, such as anti-androgens, anti-estrogens, and aromatase inhibitors; anti-inflammatories including NSAIDs, cyclooxygenase (COX-2), and lipoxygenase (LOX) inhibitors; antimutagens, such as phase II enzyme inducers; antioxidants, such as selenium and vitamin E; and angiogenesis inhibitors, including collagenase and matrix metalloproteinase blocking agents. In the second step promising chemopreventive compounds or combinations of agents possessing pleiotropic activities are subsequently developed both in carcinogen-induced and genetically manipulated animal tumor models, including models of colon, lung, bladder, mammary, prostate, and skin cancer, as well as in batteries of toxicity and pharmacokinetic tests. Development of chemopreventive drugs using this targeted approach affords a strategic framework for evaluating agents according to defined criteria and not only provides evidence of agent efficacy but also serves to generate dose-response, toxicity, and pharmacokinetic data needed to consider
Phase 1 clinical safety and pharmacokinetic evaluations.

Identification and validation of new molecular and biochemical targets in carcinogenesis is also a high priority of the preventive agent development program. The revolution in information processing and remarkable progress in molecular and cellular biology has provided potential new molecular targets for prevention and new approaches for characterizing efficacy and toxicity. These include development of transgenic and gene knock-out animal models that mimic specific characteristics of human carcinogenesis or carry well-defined genetic lesions and the use of DNA microarray technology to characterize the mechanisms of action of candidate chemopreventive agents in modulating gene expression in tissues undergoing neoplastic transformation. This technology will undoubtedly increase our understanding of underlying genetic and molecular pathways involved in cancer development and progression and will also facilitate discovery of new chemopreventive agents with novel mechanisms of action.

Drug Development Overview

The goal of the cancer preventive agent development program in the DCP, NCI is to identify safe and effective agents for the primary and secondary prevention of human cancers (mainly epithelial cancers at this time) and to bring the agents to clinical testing. By definition, cancer chemoprevention refers to the intervention or use of either naturally occurring biological substances or synthetic chemical agents to prevent initiation and to delay or reverse progression of early neoplastic lesions to clinical disease or metastatic cancer (1). The scientific rationale for chemoprevention is based largely on the concept that carcinogenesis is a multistep process that can be blocked, delayed, or reversed in its early neoplastic stages by intervention with pharmacological agents. Epidemiological evidence suggesting that dietary components may reduce cancer incidences is an important source for identifying potential preventive agents (2). Examples of these have included dietary constituents, vitamins, and minerals such as isoflavones (3), polyphenols (4), lycopene (5), vitamin E (6), calcium (7), and selenium (8,9). A second resource for identifying and characterizing potential preventive agents is experimental carcinogenesis studies which have shown that specific chemical substances can prevent progression of premalignant lesions to invasive cancers (10, 11). Thus far, several thousand chemical agents have been identified; some are distinct chemical entities, while others are mixtures. However, only carefully controlled clinical chemoprevention trials can determine whether these may be safe and effective cancer preventive agents in humans.

Another objective of the drug development program is to validate and optimize existing assays, and to discover new strategies for models in cancer prevention. This includes standardizing assay protocols using well-established chemopreventive agents, and determining test parameters such as carcinogen dose, treatment time, and dietary considerations. Research to develop new cell