DESIGNING SMARTER CANCER PREVENTION TRIALS

In April 2010, an international team of researchers from academia and drug company GlaxoSmithKline reported that dutasteride, a drug already approved for the treatment of benign prostatic hyperplasia, reduced the chances that men considered at high risk for prostate cancer would develop the disease. The four-year trial included more than 8,100 men and met the gold standard for clinical trials: it was randomized, double-blind, and placebo-controlled; it studied parallel groups at multiple medical centres; and it assessed outcomes with biopsies at two years and four years. In the end, men who took dutasteride were 23% less likely to have a positive biopsy for cancer than those on the placebo. GSK submitted this data in its application to the US Food and Drug Administration to market the drug for prostate cancer prevention

JAHI CHIKWENDIU/THE WASHINGTON POST VIA GETTY IMAGES

— this January, the FDA said No.

Although it is not unusual for the FDA to reject a drug application supported by apparently positive data, this case illustrates the particular challenges surrounding clinical trials for cancer prevention. When the aim is to decrease the incidence of cancer in large populations, studies on preventive agents require large patient cohorts — sometimes approaching 20,000 participants — and take years or even decades to perform. This combination makes them especially unwieldy compared to tests with therapeutic compounds, which can much more quickly be seen to work, or not, by testing them exclusively in people who have the disease. In cancer prevention drug trials, the usual gold standard barely rates a bronze.

Since preventives are intended for apparently healthy patients, trials require a high confidence that the anticipated anticancer benefit will outweigh any harmful side effects. In the dutasteride trial, statistical analysis showed that the decrease in cancer was driven mainly by a reduction in less serious tumours that might not even require treatment. In addition, men who took the drug were slightly more likely than those on a placebo to develop more aggressive tumours. The FDA's expert advisory panel concluded that the prevention benefits failed to outweigh this risk.

Researchers say two things are needed to decrease the length and size of prevention studies. One is

The PI3K pathway might also be used for chemoprevention. Early trials have shown that the administration of a compound that decreases PI3K activity causes regression of abnormal lesions in the airways of high-risk smokers[3].

DNA DAMAGE

As part of daily living, DNA frequently sustains damage. If not repaired, this can lead to mutations that replicate, resulting in abnormal and then cancerous growths. Certain mechanisms usually prevent this from occurring. The enzyme 8-oxoguanine DNA glycosylase (OGG1) repairs DNA by excising damaged bases (see DNA repair duties, page S21). Biochemists Zvi Livneh and Tamar Paz-Elizur, at the Weizmann Institute in Rehovot, Israel, discovered that levels of OGG1 can also be used to predict an individual's risk of developing lung cancer.

By measuring OGG1 concentration in blood samples, Livneh and Paz-Elizur found that 40% of people with lung cancer had low levels of the enzyme compared to 4% of healthy individuals. Smokers with low OGG1 activity were 5 to 10 times more likely to develop lung cancer than smokers with normal OGG1; when compared to non-smokers with normal OGG1 activity, the risk skyrocketed to 120 times more likely. The same blood test could be broadened to other cancers. For example, smokers with lower OGG1 activity are 70 times more likely to develop head and neck cancer than non-smokers with normal enzyme activity[4].

➲ NATURE.COM
read Missing the mark in this week's *Nature*
go.nature.com/4Ejeum

OGG1 is only one of an unknown number of DNA repair enzymes; low levels of any of them could be associated with cancer development. Livneh and Paz-Elizur have expanded their research to include two additional DNA repair enzymes — AAG and APE1 — to cover people with "different risk factors to develop a certain cancer", says Livneh. A study is underway to access their performance and results are expected in mid-2011.

It is unlikely that any single test, however many markers included, will be sufficient to gauge the risk of cancer development. "We have an additional ongoing study which explores a two-stage protocol for lung cancer prevention," says Livneh. The first stage involves Livneh and Paz-Elizur's DNA repair biomarkers plus five biomarkers developed by other groups. These biomarkers measure: alteration in gene expression; levels of DAP kinase, an enzyme involved in programed cell death; antibodies to mutant p53, a sign that a cell's tumour suppressor system is damaged; markers of inflammation; and variations in cancer-related genes. "Together these biomarkers are expected to yield a better risk assessment than one type alone," says Livneh. Individuals identified as high risk will be tested using spiral computed tomography (CT). "For such a high-risk group, spiral CT early detection of lung cancer might be cost-effective and life saving," adds Livneh.

In the initial stages of cancer, the body is often able to recognize abnormal cell changes and raise a response, producing auto-antibodies. However, this response is limited, and in the later stages of cancer, the immune system becomes compromised and can no longer identify and attack cancer cells. Auto-antibodies are therefore prime candidates for biomarkers of early stage cancer.

By examining auto-antibody formation in presymptomatic individuals who later went on to develop lung cancer, Samir Hanash, at the Fred Hutchinson Cancer Center in Seattle, Washington, has identified three important antigens — annexin-1, 14-3-3 Theta and LAMR-1 — regarded by the immune system as foreign[5]. So far, specificity of these biomarkers is high but sensitivity lingers around 60%. The challenge for Hanash is to find additional candidate antigens that improve on the performance of this 3-antigen panel.

These figures might be improved by looking for even earlier signs of cancer. Through the Women's Health Initiative and Physician's Health Study, Hanash has access to blood samples that were collected up to eight years before a patient was diagnosed with lung cancer. In addition, he is searching for biomarkers of lung cancer in former smokers and in people who never smoked. "It turns out that most of the blood markers we have identified among smokers are also applicable to non-smokers," says Hanash.

In spite of major investment in biomarker development over the past 15 years, the field of cancer prevention biomarkers looks woefully thin. One of the main reasons, according

to identify high-risk populations to be the preferred subjects for the trials. The second is surrogate endpoints that can provide evidence of whether a preventive drug is working — and do this in just a few years, rather than decades. The key to both is finding better biomarkers — the genes, proteins, and cellular metabolites that can be measured and associated with the development of cancer.

Patterns of these biomarkers that can be uniquely linked with one type of cancer can make it easier to estimate an individual's cancer risk. Selecting highest-risk patients for studies increases the statistical power of trials with a smaller number of participants. As a second benefit, high-risk cohorts can also shorten trials. If epidemiological studies show, for example, that a known percentage of patients carrying a certain gene will develop cancer within five years, researchers can restrict a prevention trial to those patients and run it for just that duration. Moreover, patients and regulators are likely to be more tolerant of side effects if the targeted users have a high chance of developing cancer without intervention.

The designers of the dutasteride trial did select participants judged to be at higher risk of developing prostate cancer. However, they did so by looking for elevated levels of prostate-specific antigen (PSA) — a protein whose utility as a biomarker for prostate cancer is a matter of debate. If a fully validated biomarker for prostate cancer had existed, GSK might have been able to design a dutasteride trial that required fewer participants and could have yielded a more definitive outcome. In particular, looking at the drug's effect (or lack thereof) on the biomarker may have clarified whether the increase in detected higher-grade cancers was due to the drug or simply an artefact of the tumours becoming more easily detected owing to dutasteride's shrinking of the prostate.

Some biomarkers may even function as the surrogate endpoints needed to shorten prevention trials. If, say, a specific group of proteins reliably increases in the blood of patients during the earliest, precancerous stages of disease, doctors could monitor those proteins rather than relying on biopsies to detect malignancy. Molecular biomarkers of potential toxicity, such as the activity of drug-metabolizing enzymes, could also help researchers monitor subjects' safety and response to drug candidates in clinical trials.

Scott Lippman, an oncologist and cancer prevention researcher at the University of Texas MD Anderson Cancer Center, has proposed fully integrating biomarkers chemoprevention development. After evaluating biomarkers in animal models, researchers would do epidemiologic studies linking the biomarkers to human cancers. They would next model the likelihood that patients with specific biomarkers will develop cancer. Then, in a 'phase 0' step between preclinical and phase I clinical trials, researchers could test sub-therapeutic doses to assess a drug's behaviour in healthy patients without risking harm. Lippman argues that this approach could yield better decisions on whether to undertake a lengthy, and costly phase III trial — and speed the development of preventive agents. Indeed, the fact that GlaxoSmithKline skipped some of these steps might have played a role in the FDA's decision on dutasteride. The drug inhibits the enzyme that converts testosterone to the more potent 5α-dihydrotestosterone. But neither molecule is yet a validated biomarker for prostate cancer.

Erika Jonietz *is a science writer in Austin, Texas.*

1. Andriole, G. L. *et al. N. Engl. J. Med.* **362**, 1192–1202 (2010).
2. Thompson, I. M. *et al. N. Engl. J. Med.* **349**, 215–224 (2003).

to Eleftherios Diamandis, a clinical biochemist at the University of Toronto, is because of poor study design with weak endpoints and little statistical rigor[6]. Furthermore, most research efforts have focused on biomarkers that monitor treatment. In fact, most biomarkers in clinical use are not suitable for population screening or for early diagnosis, observes Diamandis.

Diamandis claims that previous research into cancer biomarkers were looking in the wrong places. Too often efforts have focused on genetic markers, which in terms of cancer "represent 'digital information' — yes or no. This is not true for metabolomic or proteomic biomarkers, which are associated with quantitative changes", he says. But such biological markers are delicate. "They can be influenced by sample collection and storage methods, benign diseases, and even diet and drugs," he explains. A difficulty of identifying quantitative biomarkers that are both highly sensitive and highly specific is that data analytical biases are introduced. "It is not surprising that seemingly spectacular data on new biomarkers are subsequently found to be not reproducible, and therefore unsuitable for use in clinical practice," Diamandis concludes.

George Poste, head of the Complex Adaptive Systems Initiative at Arizona State University in Tempe, agrees that biomarker research is yet to deliver on its promise for these and other reasons. Part of the problem, he says, is that until recently, most investigator-initiated research has been too small and non-uniform to yield meaningful results. A lack of standardization in sample collection and processing, the use of cell lines instead of patient biopsies for research, and an insufficient number of patient samples are reasons for the dearth of meaningful biomarker development. Moreover, the field needs much more funding to encourage collaborative research and a 'big science' approach, says Poste. Government and industry funding must step up to the plate, he adds.

Poste cites the US National Cancer Institute's Cancer Human Biobank and the UK's Biobank as successful examples of big science and what it can do when the community invests in this research. He notes that historically the lion's share of cancer funding has gone to treatment, not prevention. "But the real issue is how can we catch cancer very early on, before it spreads," says Poste. This is the realm of biomarkers. "If we can find cancer in its earliest stages, it might be possible in the future to prevent it." ■

Vicki Brower *is a freelance writer in New York.*

1. Zhang, X. *et al. Physiological Genomics* **41**, 1–8 (2010).
2. Gustafson, A. *et al. Science Translational Medicine* **2** , 26ra25 (2010).
3. Lam, S. *et al. Cancer Epid. Mark. Prev.* **15**(8): 1526–1533 (2006).
4. Paz-Elizur, T. *et al. Cancer Research* **66**(24),11683–11689 (2006).
5. Qui, J. *et al. J. Clinical Oncology* **26**, 5060–5066 (2008).
6. Diamandis, E. J. *Natl Cancer Inst.* **102**, 1449 (2010).

DNA REPAIR DUTIES

8-Oxoguanine DNA glycosylase (OGG1) removes bases that have been damaged by tobacco smoke, ionizing radiation or oxidative stress.