ILLUSTRATION BY MATTHEW HOLLISTER

CHEMOPREVENTION

First line of defence

Combinations of drugs are showing some promise as therapeutic agents that stop cancer before it starts.

BY LAUREN GRAVITZ

The mice in Xiangwei Wu's lab at the MD Anderson Cancer Center in Houston seem to be cheating death. Despite being genetically pre-programed for colon cancer, they're staving off disease with a novel course of 'cancer cleansing'. Every three weeks, Wu injects the mice with a drug combo that targets only the mutant cells, prompting the cells to self-destruct. There are no side effects, nor is there any visible damage. These mice are receiving the most cutting-edge therapy conceivable: short-term treatment for long-term prevention.

Taking drugs to prevent cancer — rather than treat it — is not a new idea. In fact, over the past few decades a new specialty known as 'chemoprevention' has grown up around the concept. It's a tantalizing notion: swallow a pill that can thwart disease before it manifests. The reality, of course, may never be that simple. An untold number of genetic changes can trigger cells to become cancerous — predicting what those changes will be and who will get them is still incredibly difficult (see Portents of malignancy, page S19). Studying the changes as they occur is even harder, at least in humans. Researchers cannot investigate a developing disease until they know it's there, which means that stopping it is a biomolecular game of Whac-a-Mole. Complicating the problem is the thorny issue of people who feel fine starting a long-term drug regimen — one that can cause troublesome side effects or even put them at risk for another disease.

The process of carcinogenesis can be exceedingly slow. "Cancer begins 20 years before a woman feels the lump in her breast," says Michael Sporn, a pharmacologist at Dartmouth and the person who introduced the concept of cancer chemoprevention 35 years ago. That long span of time offers several opportunities for intervention. There's the mutation of a cell into something not-quite-healthy but also not-quite-cancerous. There's the transformation of a population of these precancerous cells into something that is immortal and grows unchecked. And then in most cancers, there's the metastatic movement of some of these cells away from the tumour, which establish a foothold elsewhere in the body.

A chemoprevention agent that blocks the very first step would be best. But most researchers would consider a drug successful if it could stop the disease progressing from any stage to the next.

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QUENCHING INFLAMMATION

Breaking the first link in the carcinogenesis chain is the ideal place to start: kill off any mutant cells that the immune system misses and do so before they have a chance to become cancerous. Yet this is a difficult proposition. Not only is it nearly impossible to observe these early changes in humans, but a drug for such early prevention would have to be absolutely benign in order to justify a near lifelong prescription.

Many in chemoprevention are beginning to think that perhaps the best way to catch cancer is to target inflammation. Chronic inflammation appears to encourage tumours by prompting the growth of new blood vessels and a remodelling of the extracellular matrix — creating a prime setting for normal cell growth to turn malignant.

Inflammation may be at the root of a host of serious ailments, from heart disease to diabetes. Stemming the tide of inflammation might prevent not only cancer but a number of other diseases. This theory led chemoprevention researchers to turn to two drugs — celecoxib and aspirin — that target the cyclooxygenase enzymes (COX-1 and COX-2), which play key roles in inflammation and pain. Both of the drugs are non-steroidal anti-inflammatories (NSAIDs), which epidemiological studies suggest may reduce the risk of colon and other cancers.

Aspirin inhibits COX-1, while celecoxib (Celebrex) inhibits COX-2. COX-1 is produced in tissues throughout the body, and is known to mediate the production of prostaglandins — chemical messengers that control a number of physiological functions, such as lowering blood pressure, regulating body temperature and controlling inflammation. COX-2, on the other hand, is strictly regulated and tends to spike during inflammation and other stress — an abundance of COX-2 has been linked to the growth and proliferation of cancerous and pre-cancerous cells. Inhibiting the COX pathways can alter cancerous and precancerous cells by decreasing blood vessel formation and cell growth. COX inhibition enhances a mutant cell's ability to commit

suicide in a process known as apoptosis and enables the immune system to recognize and target the cells for destruction.

Celecoxib — along with other COX-2 inhibitors such as rofecoxib (Vioxx), since removed from the market — were developed as alternatives to aspirin, which can cause gastrointestinal bleeding after prolonged use. And, for a while, they seemed promising not just for pain but also for cancer prevention. The US Food and Drug Administration (FDA) approved daily doses of celecoxib for reducing colon cancer risk in people with a rare genetic disease called familial adenomatous polyposis (FAP), which causes dense outcroppings of intestinal polyps. Early reports indicated that daily doses of COX-2 inhibitors could decrease the risk of breast, skin and colon cancer in high-risk individuals. While COX-2 inhibitors are still a promising drug for chemoprevention, no drugs that work through this pathway are now on the market.

But it become apparent that prolonged use of COX-2 inhibitors increased the risk of stroke and heart attack, so the FDA banned most from the market. Celecoxib remained available until early February 2011, when the manufacturer voluntarily removed labeling for this use due to its inability to do follow-up studies. A number of cancer prevention researchers continue to pursue COX-2 inhibitors, trying to develop compounds with the same inflammation- and cancer-fighting effects as celecoxib but without the risk of cardiovascular problems.

Aspirin comes with its own set of risks. "People die from it," says Leslie Ford, associate director for clinical research in the cancer prevention division of the National Cancer Institute (NCI) in Washington. "There are about 16,000 deaths a year from gastro-intestinal bleeds in people taking aspirin. But these risks aren't preventing clinical trials, and the evidence is mounting that aspirin can decrease a person's risk of colon cancer, as well as lung, prostate and even brain cancer.

Other systemic, inflammation-targeted drugs may also help ward off a variety of cancers. Some studies suggest that statins — developed for cholesterol management — might disrupt the growth and proliferation of cancer cells. Reports suggest that medications taken to increase insulin sensitivity in type 2 diabetes could lower risk for several types of cancers, including head, lung and neck. Such drugs include metformin, pioglitazone and rosiglitazone (although the latter has recently been linked to heart problems).

Precisely how or why these drugs might aid cancer prevention remains a mystery. "Every time I think I have a specific agent for a specific pathway, when it's tested, I find all sorts of other activities that may play into it as well." says Ernest Hawk, head of the Division of Cancer Prevention & Population Sciences at MD Anderson Cancer Center. "This really gets at the question: do we really ever know the mechanism of anything?"

CURBING PREMALIGNANT CELLS

Currently, most chemoprevention aims to prevent premalignant cells from completing the process of carcinogenesis. These agents have clear cellular targets and are intended to treat people at high risk — those with a family history of disease or a known genetic mutation, or who are already known to have precancerous cells.

Because different cancers evolve in various ways, a one-size-fits-all approach is not on the horizon. "One of the biggest challenges of this field is that it's really not one field," says Eva Szabo, a researcher in the cancer prevention division of the NCI. "Before we understood the complexity that is cancer, the thought was that we could have a generalized strategy that could prevent the transformation of cells, or could arrest progression." Now, she says, it's apparent that lung or breast or colon cancers can take multiple forms. And because the pathogenesis is different, the prevention has to be different, too.

Most of the medications approved by the FDA for the purpose of treating precancerous cells or reducing cancer risk (see Table opposite) fall into this category. Women who fall into high-risk categories for breast cancer, for example, can take raloxifene (Evista) or tamoxifen (Nolvadex), both of which have been shown to cut a woman's risk of estrogen-receptor positive breast cancers by as much as 50%. And five different creams and ointments have been approved to prevent skin lesions from developing into skin cancers such as squamous cell carcinoma.

Xiangwei Wu's colon cancer-prone mice belong in this category, too. Wu's two-drug combination takes advantage of mutations specific to precancerous cells in the colon, prompting only those mutant cells to self-destruct. "Because most preventive drugs don't get rid of the bad cells, people have to be on them continuously for a long time," says Wu. Such is the case with raloxifene and tamoxifen.

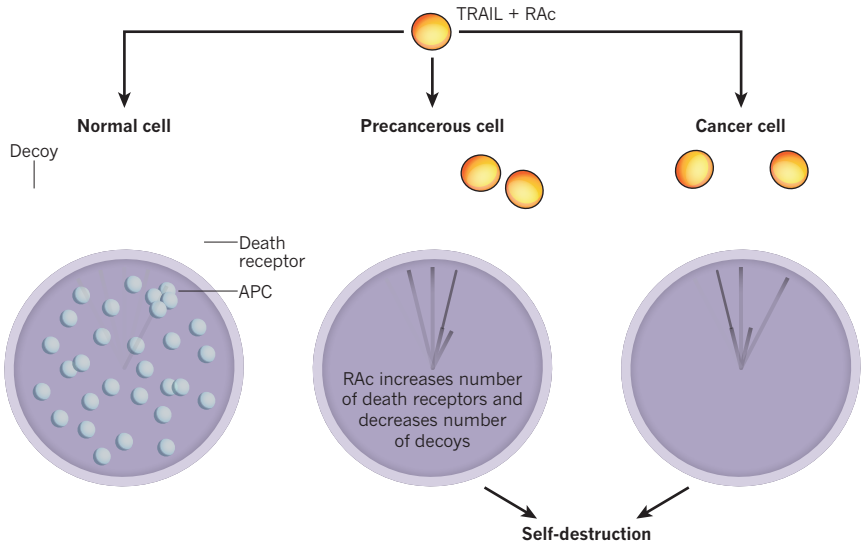
Wu's mice seem to be experiencing the best of all possible outcomes: they receive intermittent therapy that kills emerging cancer cells, but benefit from breaks between doses to recover from any side effects. In addition, the two agents — tumour necrosis factor-related apoptosis-inducing ligand (TRAIL) and all-trans-retinyl acetate (RAc) — work synergistically. Most researchers see this as important in preventing a rebound of drug-resistant populations. TRAIL has already proven promising for treating cancer because it appears to leave healthy cells unharmed while inducing cancer cells to self-destruct. In combination with RAc, Wu has found, TRAIL can induce precancerous cells to commit suicide, too (see Cancer-stopping combo).

Another two-drug combo aimed at colon cancer is one of the most promising chemoprevention prospects in the pipeline. Low daily doses of sulindac, an NSAID, along with difluoromethylornithine (DFMO) appear to do far more good together than either does alone — and with minimal toxicity, according to researchers at the University of California, Irvine. "We wanted to find the lowest dose of each at which we could find a relevant effect in the colon," says Frank Meyskens, director of the university's Chao Family Comprehensive Cancer Center.

The combination is one of push and pull. Cancerous cells have a hard time regulating the metabolism of polyamines and so have abnormally high levels of these compounds, which healthy cells use for growth and development. DFMO prevents polyamine synthesis, while

CANCER-STOPPING COMBO

TRAIL binds to receptors to trigger cells to self-destruct. Normal cells are protected both by decoy receptors and by the protein APC, which blocks the death-signal pathway. RAc helps kill precancerous cells by lowering the fraction of decoys.



FDA-APPROVED CHEMOPREVENTION DRUGS

Drug	Brand name(s)	Cancer type	Year first approved	Target / mechanism	Dosing (how long, how often)	Original manufacturer
Tamoxifen	Nolvadex Istubal Valodex	Breast	1998	Selective estrogen receptor modulator (SERM)	Daily, for 5 years	AstraZeneca
Raloxifene	Evista	Breast	2007	SERM	Daily, duration unknown	Eli Lilly
HPV vaccine	Gardasil Cervarix	Cervix Vulva Vagina/Anus	2006	Elicit immune response to prevent infection by the most common cancer-causing types of HPV	3 doses over the course of 6 months	Merck & Co GlaxoSmithKline
Porfimer sodium + photodynamic therapy (PDT) & omeprazole	Photofrin	Esophageal	2003	Lodges in precancerous cells and upon exposure to certain light produces an active form of oxygen that kills nearby cancer cells	Single injection, followed by light therapy three days later. Can be repeated after 90 days	Axcan
Fluorouracil	Efudex Fluoroplex Carac	Skin	1970	Interferes with DNA synthesis and leads to cell death	Apply to affected areas twice daily until lesions are gone, as long as 10-12 weeks.	Valeant
Diclofenac sodium 3%	Solaraze	Skin	2000	Exact mechanism is unknown	Apply to lesion twice daily, for 60-90 days	PharmaDerm
5-aminolevulinic acid + PDT*	Levulan	Skin	1999	Solution kills precancerous cell when exposed to light [1]	Apply a topical solution to lesion, then single photodynamic therapy treatment 14 to 18 hours later.	DUSA
Imiquimod	Aldara (5 %) Zyclara (3.75%)	Skin	2004	Enhances immune response and promotes apoptosis	Zyclara: Applied topically for two 2 weeks, then a 2 week break Aldara: Applied twice a week for 16 weeks	Graceway Pharmaceuticals

sulindac triggers cells to purge it. Together, in low doses, the two compounds work to lower polyamines in precancerous tissue; the result is fewer precancerous growths, which are risk factors for colon cancer. In clinical trials, Meyskens' team showed that after three years of therapy, the combined treatment reduced the occurrence of polyps by 70% and a greater than 90% reduction in advanced adenomas—the ones most likely to go on to develop cancer. The researchers found a company, Cancer Prevention Pharmaceuticals, to take the treatment through phase III trials into the market.

PREVENTING METASTASIS

With most solid-tumour cancers, the biggest danger is not the tumour itself but its ability to metastasize. While the primary tumour continues to grow, rogue cells break off, work their way into the blood stream, and move on to colonize other areas of the body. Metastases represent an advanced accumulation of genetic damage, which are too many mutations for conventional drugs to target at once.

"Across all cancers, over time, there is an almost exponential increase in the number of genetic mutations," says Raymond Bergan, a specialist in preventive oncology at Northwestern University medical school in Chicago. "We do a pretty good job of making a drug that can hit a target. But making one drug that will hit two different targets is very difficult."

Bergan is aiming for a target that prevents growth and metastasis of a primary tumour. His lab has been investigating genistein, a soy isoflavone that has been sold as a nutritional supplement for years. Bergan's group is putting the compound through its paces in the lab, and it is showing promise in trials for preventing — even reversing — the metastatic process of prostate cancer.

For cancer to metastasize, tumour cells must detach from their neighbours. This decreased adhesion is at least partially mediated by the enzyme focal adhesion kinase. Studies show that genistein blocks the activation of this enzyme so that prostate cancer cells remain tethered to the tumour.

Genistien also prevents mobile prostate cancer cells from invading tissue. As healthy cells grow and divide, the enzyme matrix metalloproteinase-2 (MMP-2) helps break down extracellular membrane proteins to make way for new growth. One of the proteins it degrades, however, is first in line for attack by invading cancer cells. Researchers have found that higher concentrations of MMP-2 correlate with poor prognosis. Genistein counteracts this by targeting a protein that increases MMP-2 concentrations. By binding to and inhibiting this protein, it can prevent production of MMP-2.

In phase II clinical trials, Bergan has shown that genistein can decrease MMP-2 in human prostate tissues — he's now looking at whether this prevents cancer cells from moving beyond the prostate, thereby reversing the cancer's evolution into metastatic disease. Following up on Bergan's research, Seema Khan, also at Northwestern University, is studying whether genistein — as part of a mixture of soy isoflavones — might prevent the spread of breast cancer. Her results, she says, are far less conclusive and hint that in younger women, this treatment could lead to a slight increase in cell proliferation.

Indeed, uncertainty and ambiguity are the norm in cancer prevention research. "I believe that there are enough instances where chemoprevention has been deleterious that it calls into question how much we understand," says John Potter, a senior advisor at the Fred Hutchinson Cancer Research Center in Seattle. "If I were to write a prescription for the field, I'd want to match risk and benefit."

Tailoring the right therapy to the right risk profile will help. "Most people think of cancer prevention like preventing polio: you get a one-time shot and you never have to worry about it again," says Powel Brown, chair of Clinical Cancer Prevention at MD Anderson. "But it's more likely to be akin to taking anti-cholesterol medicine for the prevention of heart disease." That is, you'll be taking the medication indefinitely — and while there may be side effects, they will be acceptable to reduce the risk of a potentially fatal disease.

Indeed, if scientists and clinicians are able to establish a cancer prevention system as well-tuned as that for heart disease, it would count as a massive success. For now, researchers in the field will have to look to the mice in Wu's lab, which are revealing one possible path to a cancer-controlled — if not cancer-free — future. ■

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