Thwarting Tumors: Nereus Trolls for Cancer Cures beneath the Sea

Wendy Wolfson
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Cancer tumors cannot grow more than a few millimeters in size, nor can they spread without developing their own network of blood vessels to supply oxygen and nutrients. Tumor blood vessels differ from normal blood vessels in that they are chaotic, leaky, and fragile. This difference makes them more susceptible to compounds that damage tumor blood vessels.

Antiangiogenic drugs such as Avastin, Nexavar, and Sutent prevent new blood vessels from forming, but a new class of drugs, called vascular disrupting agents or VDAs, damages established tumor vasculature. These can provide options to treat cancers resistant to conventional chemotherapeutics.

Using VDAs, in combination, could potentially lower the dose of toxic chemotherapeutics that must be administered to do the job and produce fewer side effects than current therapies.

San Diego-based Nereus Pharmaceuticals (http://www.nereuspharm.com) is one of several biotechs developing small molecule VDAs. Nereus's lead compound NPI-2358 disrupts the tubulin scaffolding of the endothelial cells lining tumor blood vessel walls. Rival compounds include OXiGENE's combretastatin A4 disodium phosphate and OXi4503 and Angiogene's ZD6126. Like the antiangiogenics, VDAs are selective for tumor blood vessels and spare vasculature in healthy tissue. VDAs cut off tumor blood supply, causing the tumor to die from hypoxia from the inside out. VDAs work faster than conventional chemotherapeutics and antiangiogenesis agents: in a matter of hours rather than weeks.

If VDAs are administered as a sole drug, the tumor rim remains, co-opting blood vessels in neighboring healthy tissue. The tumor tends to regrow. At this point, VDAs are slated for combination therapy with chemotherapeutics and antiangiogenic drugs that should mop up what is left of the tumor after the VDA is administered. "All cancer therapy is a combination of two, three, or four agents," said Kenneth Lloyd, Ph.D., Nereus chief scientific officer. According to Lloyd, the sequence of medication is key. A chemotherapeutic is administered first and then tumor blood vessels are knocked out with the VDA. It is hoped that using VDAs in combination could potentially lower the dose of toxic chemotherapeutics that must be administered to do the job and produce fewer side effects than current therapies.

Diving for Drugs

Nereus derives its anticancer compounds and antibiotics from marine microbes. According to Lloyd, marine microbes are an underexploited source of natural products.

Fenical, an oceanographic scientist at the Scripps Institution of Oceanography at the University of California, San Diego, spent two decades collecting marine microorganisms. If terrestrial organisms were rich sources of drugs, why not ocean microbes? In 1998, Nereus was established with research licensed from Scripps. The 32 person company moved to its present laboratory in 2002.

Nereus just received $45 million in its series D round led by BankInvest of Copenhagen, bringing its total funding to about $100 million. Nereus is backed by an international group of venture capitalists including HBM BioVentures, Alta Partners, Forward Ventures, Advent International, GIMV, InterWest Partners, and Pacific Venture Group.

The company currently has a trove of 20,000 organisms collected from oceans around the globe. Nereus grows the microbes and separates the drugs from them or synthesizes the active compound rather than going the DNA bioengineering route.

The company's VDA NPI 2358 was chosen from a series of around 200 synthetic analogs of a compound discovered in an extract of *Aspergillis*, a marine fungus. The compound is now in phase I human clinical trials for solid tumors and lymphomas. NPI 2358 is a "second generation VDA" that has a dual effect on tumor blood flow and directly induces tumor cell apoptosis, as does rival OXiGENE's OXi4503.

NPI 0052, Nereus's other lead compound, a potent proteasome inhibitor, is undergoing three phase I human clinical trials for treatment of multiple myeloma, solid tumors, lymphomas, and leukemias, respectively. NPI 0052 came straight from a marine bacteria, *Salinospora tropica*, that Fenical discovered and Nereus inlicensed. Due to its complex structure, the compound was isolated from the bacteria itself, which is grown in a fermentation tank, rather than produced through a chemical synthesis route.

According to Lloyd, the early VDAs such as those developed by Astra Zeneca and Aventis had side effects such as heart attacks and stroke. Nereus spent great effort checking the toxicity on animals. The company believes that the animal tests will be a good indication of expected side effects and toxicity, since humans can tolerate greater doses than animals. Neureus has stiff competition from

other biotechs with VDA programs, as well as their pharma sponsors.

Combrestastins Trash Blood Vessels, Suffocate Tumor Centers

Waltham, MA-based OXiGENE's (http://www.oxigene.com) small molecule compound Zybrestat induces tumor blood vessels to hemorrhage, causing tumor cells to die. OXiGENE's VDAs are analogs of combretastatins, compounds isolated from the South African tree *Combretum caffrum*. One of them, CA-4 has a high affinity for tubulin at or near the colchicine-binding site, causing tubulin polymers of the endothelial cytoskeleton to destabilize. Drugs that target tubilin were known to eliminate tumors in the early 1980s when Juliana Denekamp, Ph.D., at Gray Laboratory Cancer Research Trust, Mount Vernon Hospital, London, posited that damaging the neovasculature in tumors was a viable strategy for treating cancer. However, the drugs available to her, such as vincristine, vinblastine and colchicine, were too toxic at the doses necessary to work.

The company has completed a phase II study and now is enrolling patients in a registration study for Zybrestat as a potential treatment for anaplastic thyroid cancer (ATC), a rare, fatal cancer with an incidence of about 1000–4000 cases a year in the U.S., as well as a combination phase Ib study of Zybrestat plus Avastin in patients with advanced solid tumors. OXiGENE is further conducting a phase I trial of OXi4503, a dual-mechanism VDA in patients with advanced solid tumors that collapses tumor vessels and kills tumor cells directly. OXiGENE is focusing as well on age-related macular degeneration, ovarian cancer, and small-cell lung cancer.

VDAs Get New Attention

All eyes now are on the deal Novartis made with British company Antisoma (http://www.antisoma.com) in April 2007, worth up to US $890 million, for Antisoma's flavinoid vascular-disrupting agent ASA404 (formerly DXMAA) in phase II trials. DMXAA was discovered by Professors Bruce Baguley and William Denny and their teams at the Auckland Cancer Society Research Centre (ACSRC).

Sanofi-aventis is developing AVE-8062, a combrestatin A-4 analog and microtubule-disrupting agent for tumors, and plans to begin a phase III trial in patients with advanced sarcoma in January 2008, as well as a second phase III trial for non-small-cell lung cancer. The active compound was published by Ajinomoto.

UK biotech Angiogene (http://www.angiogene.com) reacquired the rights to its tubulin-binding VDA, ZD6126, from AstraZeneca after its phase I clinical trial. (The company has another VDA, MNO929, which it licensed to San Diego-based MediciNova.) Angiogene is currently raising money to put ZD6126 through phase II trials.

According to Peter Davis, Ph.D., Angiogene CEO, tumor angiogenesis (the formation of blood vessels to feed the tumor) can be driven by a number of factors. ''Because tumors are always very clever at evading therapies, it is quite likely that a tumor is able to escape from any therapy you give it which is blocking one pathway,'' ''It would be probably more effective to actually destroy the blood vessels already in the tumor rather than stop new blood vessels forming.'' While antiangiogenics prune the vasculature leading to the tumor to some extent, ''it is nothing like the dramatic effects we get with vascular disrupting agents,'' Davis said.

Rising Pressure

In the early clinical trials of a number of VDA companies such as Angiogene, OXiGENE, and Sanofi-aventis, hypertension as well as cardiovascular side effects appeared. ''It has been a problem in the past because of its fairly acute nature, whereas the onset of hypertension occurs quite slowly with antiangiogenic agents,'' said Peter Davis of Angiogene. ''The good thing about hypertension associated with VDAs as opposed to antiangigenic agents is that our hypertension only lasts for four hours.'' Evidence taken from animal studies and from Angiogene's competitors showed that blood pressure could be controlled during administration. Companies now exclude patients in clinical trials for pre-existing hypertension and cardiovascular problems. Davis said that patients also report tumor pain during administration but that is now treated by painkillers. ''It is not a question of or whether these compounds work,'' said Davis. ''They clearly do at well-tolerated doses. There is the question of how best to use them in combination.''

Mice Lit Up Like Christmas Trees

''These agents (VDAs) have been relatively successful.'' said Dr. Robert Kerbel, senior scientist at the Sunnybrook Research Institute and professor in the University of Toronto's Department of Medical Biophysics and Department of Laboratory Medicine and Pathobiology. ''There have been hundreds and hundreds of patients; there have been few serious adverse events.'' But in his angiogenesis research, Kerbel was confronted with a mystery. Why did the rim of the tumor grow back so robustly after being treated with VDAs, chemotherapeutics, and antiangiogenic drugs? ''The only way this could possibly happen is there must be a cell that comes from bone marrow, which incorporates itself to become [a] full-fledged, credentialed endothelial cell,'' Kerbel said. Earlier research suggested this could not be replicated by administration of antiangiogenesis drugs. ''We tried to convince ourselves that these CEPS (circulating endothelial progenitor cells) are real. These cells exist and they could be a surrogate biomarker for angiogenesis.'' Kerbel said. Kerbel, Postdoc Yuval Skaked and team irradiated mice injected with GFP protein and induced them to grow tumors. Then they dosed them with OXiGENE's Zybrestat and OXi-4503. ''The mice lit up like Christmas trees,'' Kerbel recounted. The team concluded that administration of VDAs caused acute stress, stimulating the release of CEPs from bone marrow. ''When you give a vascular-disrupting agent, there is a systemic response.'' Kerbel said. ''The VDAs cause a spike in CEPS.'' ''Supposing after that spike occurs, they migrate to the tumor rim, what would they do there?'' asked Kerbel. ''They would facilitate tumor repopulation.'' But when an antiangiogenic drug like Avastin was given in sequence before the VDA, the tumor rim size and bloodflow were reduced [1].

Researchers and clinicians strive for the ideal "therapeutic window," calibrating potent drugs up to the maximum tolerated dose to kill tumors the first time around. This kind of research can point the way to the ideal dosage and sequence timing of combinations. "The next major stage is how well are these drugs working in patients." Kerbel says. "Are they really safe? Are they really effective?"

REFERENCE

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Wendy Wolfson (wendywolfson@nasw.org) is a science technology writer based in Oakland, CA.