BIOCENTURY
Innovations
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TOOLS & TECHNIQUES

SHARED BURDEN

By Karen Tkach Tuzman, Associate Editor

Having learned their lesson with PD-L1, companies are collaborating on assay standards for tumor mutation burden — a likely contender for the next major biomarker in immuno-oncology. A Friends of Cancer Research-led consortium of pharmas and diagnostics companies is driving the charge and plans to publish a white paper with its recommendations this year.

The coalition has been gathering members and momentum since it began meeting in September to create a framework for comparing tests on tumor mutation burden (TMB).

So far, discussions have included representatives from Friends, FDA, six test developers and six pharmas — AstraZeneca plc, Bristol-Myers Squibb Co., the Genentech Inc. unit of Roche, the EMD Serono Inc. subsidiary of Merck KGaA, Merck & Co. Inc. and Pfizer Inc.

The partners are aiming to avoid the problems created for drug developers by the lack of harmony over assays for PD-L1. Four immunohistochemistry (IHC) assays, each with its own scoring algorithm and method of measuring cells, were used to support trials for at least five Phase III mAbs against PD-1 or PD-L1. That has made it nearly impossible to compare results across the class and determine what level of PD-L1 expression predicts patient benefit.

In 2015, a group of 10 companies, regulatory agencies and cancer associations launched the Blueprint Project to compare PD-L1 assays, standardize scoring and recommend best practices. The project published the first phase of its analysis last year in the *Journal of Thoracic Oncology*.

Since TMB could upstage PD-1/PD-L1 for stratifying patients in cancer immunology, stakeholders are pooling heads now to define common assay parameters for the new biomarker.

"We saw this play out with the PD-L1 IHC story, and we wanted to get ahead of the problem," said David Fabrizio, leader of cancer immunotherapy at Foundation Medicine Inc., a member of the consortium.

Evidence has been growing in the literature that TMB, a sign of how immunogenic a tumor is, could be a better predictor than PD-L1 of how a patient will respond to an immunotherapy.

BMS acted on that evidence, opting to switch from PD-L1 status to TMB level for a subset of patients in its Phase III CheckMate -227 trial.

THINKSTOCK

On Feb. 5, BMS announced Opdivo nivolumab plus Yervoy ipilimumab met the progression-free survival (PFS) endpoint vs. chemotherapy in first-line non-small cell lung cancer (NSCLC) patients with high TMB.

The decision followed BMS's 2016 high profile Phase III miss in CheckMate -026. In that trial, Opdivo monotherapy missed the endpoint of improving PFS vs. chemotherapy in first-line NSCLC patients whose tumors expressed PD-L1 at ≥5%. Post-hoc whole exome sequencing suggested the compound would have met in TMB-high patients.

TMB is a measure of the number of somatic mutations in a tumor genome, which correlates with the number of neoantigens on tumor cell surfaces. Because T cells recognize neoantigens, which are not found in normal cells, the more neoantigens are present, the higher the likelihood and potential potency of a T cell-mediated antitumor response.

"I think the jury is still out from a prospective perspective, but there is enough data now from us and others that suggests it may be a reasonable biomarker to identify patients who derive benefit from this class of agents," said Priti Hegde, director and senior scientist for oncology biomarker development at Genentech. The company is including a prospectively defined TMB-high cohort in a Phase II/III study of its anti-PD-L1 mAb Tecentriq atezolizumab.

Unlike IHC, TMB assays aren't tied to the subjectivity of a pathologist's eye. But the science is still in flux on questions such as how to weigh different mutations and how to extrapolate mutation counts between gene panels and whole exome sequencing.

The coalition's goal is to create a centralized understanding of best practices and make it easier to compare results across assays. "We want to generate alignment against a universal set of standards," said Fabrizio.

COMMON GROUND

Friends of Cancer Research President and CEO Jeff Allen told BioCentury the TMB coalition emerged from a forum on analytical performance standards for next-generation sequencing (NGS) testing co-sponsored by Friends and Alexandria Real Estate Equities Inc.

Allen said companies have been very willing to work with competitors to enable comparison across assays. "There's a recognition that the way that PD-L1 expression was moving forward, with multiple tests and multiple drugs, was becoming quite complicated and probably not advantageous to anyone."

"The idea is to try to get out in front of it this time," said Eric Rubin, SVP of Oncology Early Development at Merck & Co. "The Blueprint Project arose when folks had already filed. In this case it's a little bit earlier, so there's an opportunity for some alignment before labels get out there."

Allen said the coalition hopes to formalize protocols that will allow companies and academics to determine the degree to which they'd like to participate.

The goal is to "unify the approach to measuring" TMB. "It's not that any one approach is the right way; we just want to better understand how they relate to one another," said Allen. "I hope this analytical collaboration will allow different test manufacturers to be able to say, here's how our test relates to diagnostic company A. That may make the drugs and the testing available to more patients."

To enable comparison across tests, the partners hope to generate shared reference standards based on whole exome sequencing from common cell lines or databases.

They also want consensus on the relationship between the frequency of mutations per megabase as calculated via targeted sequencing panels, and the total number of mutations in the exome, said John Simmons, director of translational science and diagnostics at Personal Genome Diagnostics Inc. (PGDx). He said 10 mutations per megabase roughly corresponds to 200 distinct mutations in an exome.

The coalition also hopes to align companies on protocol issues such as determining which mutations to include.

"In some instances, different mutations may be weighed more heavily than others," said Allen. "We're trying to allow that type of flexibility as the understanding behind TMB continues to evolve."

He said agreeing on cutoffs for different cancers is outside the scope of the coalition for now, although a feasible aim is to "get to the point where we're able to discuss different common ranges, in terms of what constitutes high levels of mutational burden, versus intermediate and low."

MEASURING MUTATIONS

Even before launching the consortium, consensus had been emerging across assay developers that sampling a few hundred genes could replace sequencing the whole exome.

TMB calculations from whole exome sequencing correlate well with Foundation's 324-gene panel (R^2 = 0.95), according to Fabrizio, and with Illumina Inc.'s TruSight Tumor 170 assay (R^2 = 0.91), according to the product datasheet.

At least eight diagnostic companies and one non-profit organization are commercializing NGS-based tests to measure TMB. All are commercializing tests using panels of 170 to 592 genes (see "Weighing the Burden").

Simmons said PGDx shifted from whole exome sequencing to targeted sequencing panels because they are more economical, scalable and accessible to patients. In addition, the panels are better able to identify chromosomal rearrangements, and have a clearer regulatory path, he said.

WEIGHING THE BURDEN

At least eight companies and one not-for-profit organization are developing or marketing next-generation sequencing (NGS) assays to measure tumor mutation burden (TMB). Most are including TMB analysis as part of a larger profile of tumor tissue or circulating tumor DNA. Some, like FoundationOne CDx from **Foundation Medicine Inc.** (NASDAQ:FMI) and MSK-IMPACT from **Memorial Sloan Kettering Cancer Center** (MSKCC), are FDA-approved as *in vitro* diagnostics, but most are currently for research use only. (A) MSKCC and five companies — Foundation Medicine, **Illumina Inc.** (NASDAQ:ILMN), **Personal Genome Diagnostics Inc.**, **Qiagen N.V.** (Xetra:QIA; NYSE:QGEN) and **Thermo Fisher Scientific Inc.** (NYSE:TMO) — are participating in a coalition led by **Friends of Cancer Research** to harmonize assay standards. *Source: ClinicalTrials.gov; company websites*

COMPANY	TEST NAME	TEST DESCRIPTION
Caris Life Sciences Inc.	Caris Molecular Intelligence CGP+	Assay profiling mutations in tumor tissue using a 592-gene panel
Foundation Medicine Inc. (NASDAQ:FMI) (A)	FoundationOne CDx	Assay profiling mutations in tumor tissue using a 324-gene panel
Foundation Medicine Inc.; **Roche** (SIX:ROG; OTCQX:RHHBY)	bTMB assay	Assay profiling mutations in cell-free DNA in plasma using a 394-gene panel
Illumina Inc. (NASDAQ:ILMN) (A)	TruSight Tumor 170	Assay profiling mutations in tumor tissue using a 170-gene panel
KEW Group Inc.	Cancerplex	Assay profiling mutations in tumor tissue using a >400-gene panel
Memorial Sloan Kettering Cancer Center (A)	MSK-IMPACT	Assay profiling mutations in tumor tissue using a 468-gene panel
NeoGenomics Inc. (NASDAQ:NEO)	NeoTYPE Discovery Profile	Assay combining NGS testing of 315 molecular markers and Tumor Mutation Burden (TMB) analysis
Personal Genome Diagnostics Inc. (A)	Unnamed panel	Assay profiling mutations in tumor tissue using a >500 gene panel
Qiagen N.V. (Xetra:QIA; NYSE:QGEN) (A)	GeneRead DNAseq Mix-n-Match Panels	Customizable assay profiling mutations using 570 primer sets
Thermo Fisher Scientific Inc. (NYSE:TMO) (A)	Ion Torrent Oncomine Tumor Mutational Load Assay	Assay profiling mutations in tumor tissue using a 409-gene panel

"We know that for a lot of the thresholds that are in range, you can make a smaller panel to deliver that," he said.

The relationship between panel size and sensitivity for detecting low rates of TMB is also important, said Fabrizio. While panel sizes are limited by the cost of sequencing at depths required to maintain accuracy, "a smaller panel will only have accuracy down to a high TMB cutoff."

told BioCentury it plans to use data from CheckMate - 227 to support addition of TMB to its label.

POPULATING PANELS

Companies are designing panels of genes that serve two primary purposes: acting as surrogates for neoantigen load, and matching patients with targeted therapies.

> "We saw this play out with the PD-L1 IHC story, and we wanted to get ahead of the problem."
>
> David Fabrizio, Foundation Medicine

According to Simmons, PGDx performed retrospective studies with undisclosed pharmas and discovered TMB thresholds as low as six or eight mutations per megabase might be needed to identify patients likely to benefit from certain combinations or specific monotherapies.

"We needed an assay that had at least a megabase of coding sequence included to have accuracy across that dynamic range," he said.

The companies differ on whether and how they define high TMB status.

For example, Caris Life Sciences Inc. defines high TMB status for its assay as 17 mutations per megabase or more; Foundation Medicine and PGDx leave that decision to the pharmas.

"They're the ones with the clinical data and the statistical power to decide those cutoffs," which will vary by cancer type and line of therapy, said Simmons.

Steven Averbuch, head of precision medicine at BMS, said the pharma plans to publish how it selected a prospective cutoff of 10 mutations per megabase for CheckMate -227 based on retrospective analyses. At this year's meeting of the American Association for Cancer Research (AACR), BMS will present data on identification of a TMB cutoff from CheckMate -568, a Phase II trial on Opdivo plus Yervoy in stage IV NSCLC.

The pharma employed Foundation's 324-gene FoundationOne CDx assay to calculate TMB for the trial. FoundationOne CDx is FDA approved as a companion diagnostic test for 17 targeted therapies used to treat solid tumors. In February, the company

But the analysis methods for the two purposes are different, said Simmons. For TMB, he said PGDx and others de-prioritize resistance mutations that are enriched after treatment with tumor-targeted therapies, and "hotspot" regions of the genome that are prone to mutations in many patients.

"Hotspot mutations don't differentiate between a high mutation burden cancer and low mutation burden cancer," said Simmons. "It doesn't really capture the biology of neoantigens that are really what's driving the T cell response."

Fabrizio said Foundation's panel includes genes in the DNA damage response pathway whose mutant forms are likely to drive accumulation of additional mutations.

The panels also often include genes important for cancer biology, that don't yet have a clear link to drug response or resistance.

"There aren't 500 clinically relevant genes," said Simmons. His company looks at pathways commonly involved in cancer and drug resistance, and at how prevalent alterations are across the exome, "to make sure that we're covering the cancer genome."

Another consideration is whether to use reference genomes to calculate TMB, or to match tumor sequences to normal tissue from the same patient.

Foundation and PGDx are using the former. "We want to make sure we're making a test that is deployable and has all the efficiencies needed to enter routine clinical practice," said Simmons.

Fabrizio acknowledged public reference genomes don't fully account for patient diversity. Foundation supplements them with

private databases of rare germline variants and computational methods to find out whether mutations are germline or somatic; the latter is more likely to be specific to the tumor.

> "It's not that any one approach is the right way; we just want to better understand how they relate to one another."
>
> Jeff Allen, Friends of Cancer Research

FUTURE BURDEN

BMS's Averbuch believes TMB will be most effective when combined with other biomarkers, such as markers of host inflammation or resistance.

"Probably composite biomarker approaches will more precisely define opportunities for response, especially when you're thinking about combination immunotherapies with different mechanisms of action," he said.

He thinks TMB and PD-L1 are "very complementary" biomarkers. "They don't correlate with one another; they're telling us about different aspects about the tumor-host environment and the immunobiology."

But combining different diagnostic technologies like IHC and NGS-based panels with rapid turnaround for patient decision-making will be challenging, said Averbuch. "We put a lot of effort into collaborations with the diagnostic industry to help us sort through that."

BMS is also exploring the predictive value of additional host, tumor and microbial factors, both internally and through a five-year collaboration with Johns Hopkins University launched in 2016.

An open question is whether directly counting neoantigens will enable better predictions than TMB.

"It's very plausible that the quality of the neoantigens will be very important, especially in tumors where there is low TMB," Averbuch said. BMS is investigating this as well, internally and through collaborations.

Merck's Rubin said that while the link between TMB and neoantigen load is the leading explanation for TMB's predictive value, there are competing theories he thinks are viable.

"There are competing theories that have more to do with other changes that happen in the cell in response to high mutation rates. For example, there are sensor pathways like STING that recognize disturbed DNA," Rubin said. "We're still sorting through that."

COMPANIES AND INSTITUTIONS MENTIONED

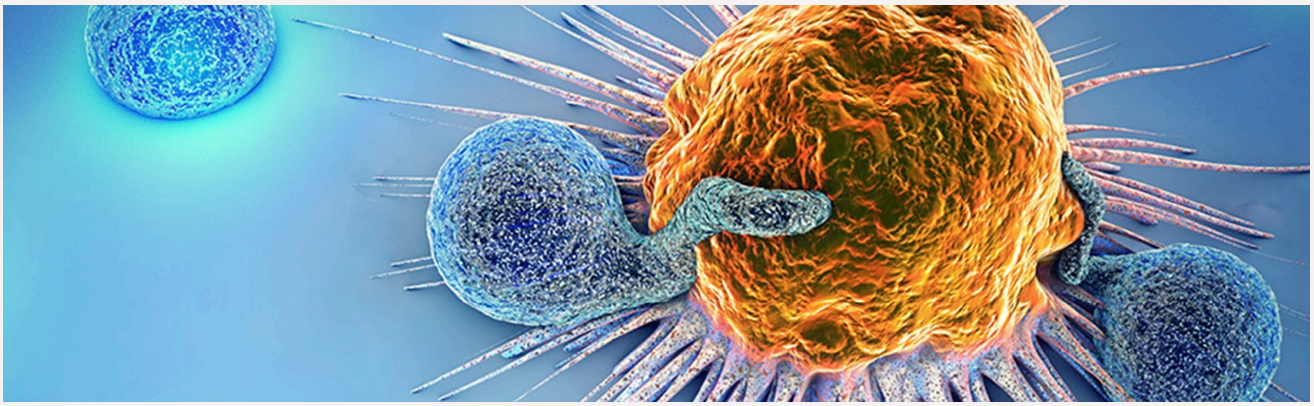
American Association for Cancer Research (AACR), Philadelphia, Pa.
AstraZeneca plc (LSE:AZN; NYSE:AZN), London, U.K.
Bristol-Myers Squibb Co. (NYSE:BMY), New York, N.Y.
Caris Life Sciences Inc., Irving, Texas
Foundation Medicine Inc. (NASDAQ: FMI), Cambridge, Mass.
Friends of Cancer Research, Washington, D.C.
Genentech Inc., South San Francisco, Calif.
Illumina Inc. (NASDAQ: ILMN), San Diego, Calif.
Johns Hopkins University, Baltimore, Md.
Merck & Co. Inc. (NYSE:MRK), Kenilworth, N.J.
Merck KGaA (Xetra:MRK), Darmstadt, Germany
Personal Genome Diagnostics Inc., Baltimore, Md.
Pfizer Inc. (NYSE:PFE), New York, N.Y.
Roche (SIX:ROG; OTCQX:RHHBY), Basel, Switzerland
U.S. Food and Drug Administration (FDA), Silver Spring, Md.

TARGETS AND COMPOUNDS

PD-1 (PDCD1; CD279) - Programmed cell death 1
PD-L1 (B7-H1; CD274) - Programmed cell death 1 ligand 1
STING (TMEM173) - Transmembrane protein 173

REFERENCES

Hirsch, F., et al. "PD-L1 immunohistochemistry assays for lung cancer: Results from Phase 1 of the Blueprint PD-L1 IHC Assay Comparison Project." *Journal of Thoracic Oncology* **(2017)**
McCallister, E. "Necessary adjustment." *BioCentury* **(2018)**
Hansen, S. "The devil is in the assay." *BioCentury* **(2016)**
Tkach, K. "Unmarked territory." *BioCentury Innovations* **(2016)**
Tuzman, K. "Quality over quantity." *BioCentury Innovations* **(2018)**



THINKSTOCK

PRODUCT R&D

ENGINEERED TO KILL

By Lauren Martz, Senior Writer

Nkarta Inc. is betting that boosting NK cells' intrinsic ability to discriminate between cancer and healthy cells will increase their efficacy as anti-cancer agents and lower their risk of toxicity compared with the more common tactic of adding in chimeric antigen receptors.

The company was formed to develop technology from scientific founder Dario Campana, and has been incubating in Johnson & Johnson's South San Francisco JLABS site since 2015, with undisclosed funding from SR One, New Enterprise Associates and Novo Ventures.

Last month, Paul Hastings joined as president and CEO. Hastings was previously chairman, president and CEO of cancer company OncoMed Pharmaceuticals Inc.

Campana is professor of pediatrics and director of the Division of Immunopathology and Cell Therapy at the National University of Singapore. He's also a founder of T cell therapy company Unum Therapeutics Inc.

Nkarta joins a field of at least nine other companies developing NK cell therapies as cytotoxic agents for cancer (see "NK Cells Across Generations").

The first wave of NK cell products aimed to use the cells as therapeutic infusions in their unmodified, but often expanded and activated, state to treat various cancers.

However, the efficacy has disappointed in the clinic. Most companies are now engineering in CARs against specific cancer antigens to increase the cells' ability to target tumors.

According to Nkarta SVP of R&D James Trager, that approach might make NK cells liable to the same toxicities as CAR T cells, in particular the risk of triggering cytokine release syndrome.

Nkarta's goal is to preserve as much of the cells' safety profile as possible, by engineering in molecules that can increase the cells' tumor-killing potency by augmenting an endogenous signaling pathway and increasing persistence *in vivo*.

"CARs work well with NK cells, but we do feel there's an advantage to riding the natural biology of NK cells to maintain the good margin of safety," he said.

In addition, the company has a technology for expanding the cells that bypasses an initial purification step and cuts up to two-thirds of the standard time.

INNATE MAGNIFICATION

As innate immune cells, NK cells don't target a single antigen, but integrate a variety of surface clues on a target cell to determine if it is stressed and should be killed.

Trager said the requirement to sum multiple signals acts as a safeguard against toxicity in normal tissues. In addition, the cells' ability to recognize multiple distress signals in various

combinations means they should be less susceptible than T cells to suppression by the tumor microenvironment, he said.

"A T cell is guided towards a single antigen and kills the cell in front of it, but the T cell effect can be down-regulated when the tumor loses the antigen," he said. By contrast, "NK cells read out a variety of positive and negative signals that help them recognize stressed cells in one way or another, making it less likely for a cancer cell to evolve evasion mechanisms."

He thinks adding CARs to NK cells could make them vulnerable to such "antigen escape," which would then render them no more effective than unmodified NK cell therapies.

The first CAR-NK cell programs have entered the clinic. Last month, NantKwest Inc. and University Hospital Frankfurt started a Phase I trial of a HER2-targeted CAR-NK cell in glioblastoma patients. PersonGen Biomedicine (Suzhou) Co. Ltd. has a CAR-NK cell therapy against multiple targets in Phase II studies in China.

NK CELLS ACROSS GENERATIONS

While **Nkarta Inc.** and two other companies have skipped to next-generation NK cell technologies, the majority of the competitors have first-generation products in the clinic. NK cell therapy companies initially went to the clinic with non-engineered versions of the innate immune cells, but many are now working on next-generation technologies. At least 10 companies have disclosed clinical or preclinical NK cell programs. Among the field's nine clinical candidates, seven involve non-engineered versions of the cells. Next-generation therapies coming down the line typically incorporate genetic modifications to add new functionalities to the cells. Six out of the eight companies that have disclosed next-gen technologies are adding CAR constructs to enhance tumor targeting, and two are adding receptors that bind therapeutic cancer antibodies. Nkarta Inc. stands alone in its approach of engineering the cells to overexpress an endogenous receptor complex plus a membrane-bound growth factor. (A) Partnered with **Oxford BioMedica plc** (LSE:OXB); *Source: BCIQ: BioCentury Online Intelligence; company websites and interviews*

COMPANY	FIRST-GENERATION PRODUCT	DESCRIPTION	STATUS	INDICATION	NEXT-GENERATION TECHNOLOGY
Green Cross Corp. (KSE:006280)	MG4101	Expanded allogeneic NK cells	Phase II	Hepatocellular carcinoma; acute myelogenous leukemia (AML)	Unspecified gene modifications (A)
NantKwest Inc. (NASDAQ:NK)	Activated Natural Killer cells (aNK cells)	Allogeneic activated NK cells	Phase II	Merkel cell carcinoma	CARs; antibody receptors
PersonGen Biomedicine (Suzhou) Co. Ltd.	Anti-CD19 CAR-NK cells	NK cells engineered with CARs against CD19	Phase I/II	CD19+ leukemia and lymphoma	CARs
	Anti-CD33 CAR-NK cells	NK cells engineered with CARs against CD33	Phase I/II	CD33+ AML	
CellProtect Nordic Pharmaceuticals AB	CellProtect	Autologous expanded and activated NK cells	Phase I/II	Multiple myeloma (MM)	None disclosed
Celularity Inc.	PNK-007	Umbilical cord blood-derived expanded NK cells	Phase I	AML; MM	CARs
Fate Therapeutics Inc. (NASDAQ:FATE)	Fate-NK100	Memory NK cells	Phase I	AML; ovarian cancer; solid tumors	CARs; antibody receptors
Gamida Cell Ltd.	NAM-NK cells	Nicotinamide (NAM)-expanded haploidentical or mismatched related donor NK cells	Phase I	B cell lymphoma; MM	None disclosed
Glycostem Therapeutics	oNKord NK cells	Umbilical cord blood-derived NK cells	Phase I	AML	Modifications including CARs
Nkarta Inc.	NA	NA	NA	NA	Activating receptor complexes; growth factors
Ziopharm Oncology Inc. (NASDAQ:ZIOP)	NA	NA	NA	NA	Modifications including CARs

NantKwest and Fate Therapeutics Inc. are also engineering NK cells to express receptors that bind antitumor antibodies.

Nkarta is the only company aiming to boost NK cell efficacy by exploiting the cells' natural tumor targeting mechanisms.

One of its approaches, described in a 2013 *Cancer Research* paper, involves overexpression of an endogenous receptor complex involved in NK cell activation, cellular cytotoxicity and cytokine production.

To produce high levels of the complex, the team transduced NK cells with genes encoding its constituents: the NKG2D receptor; DAP10, an adapter protein that stabilizes NKG2D on the cell surface; and the intracellular domain of CD3ζ, which initiates a cytotoxic signaling pathway in the cells upon receptor activation.

Campana's lab showed that when the receptor was engaged via a tool antibody, the transduced cells secreted higher levels of proinflammatory molecules and cytotoxic granules, and in a xenograft mouse model of osteosarcoma, the transduced cells decreased tumor burden, compared with control NK cells.

A second approach involves expressing a membrane-bound version of IL-15 to improve *in vivo* persistence.

"One of the differences between NK cells and T cells is that NKs are shorter lived. That's not a terrible thing in general, but we need them around long enough to kill the tumors," said Trager.

NK cells naturally express IL-15 receptors. According to Trager, because IL-15 is the main growth factor for NK cells, engineering cells to express the ligand on the cell surface should ensure higher, consistent levels of receptor engagement. "Essentially the ligand is on a long protein hinge, which gives it enough distance from the surface and flexibility to bind its receptors," he said.

In a 2014 *Blood* paper, Campana showed introducing membrane-bound IL-15 into NK cells increased survival in culture and increased *in vivo* expansion in mice.

Trager isn't aware of other companies that have engineered NK cells to express IL-15. Ziopharm Oncology Inc. has engineered T cells to express the cytokine.

Nkarta plans to test cells modified to express both the NKG2D complex and IL-15 first in hematological malignancies since those cancers have already been successfully treated with engineered T cells. He would not disclose the specific indications the company will test first or when trials will start.

"Should we detect a clinical signal, we will want to move quickly into solid tumors," he added.

BIOCENTURY PRODUCT PROFILE	
INNOVATION STAGE	
Product	NK cells overexpressing the NKG2D receptor complex and membrane-bound IL-15
Concept	NKG2D complex increases reactivity of NK cells to cancer signals; membrane-bound IL-15 increases the cells' *in vivo* persistence
Disease	Cancer
Competition	Unmodified NK cells; NK cells engineered to express CARs; other cell therapies
Differentiation	Increased potency and persistence; avoids CAR-associated toxicities
Administration	IV
Risks	Unknown toxicity related to excessive NK cell activity or persistence
Development status	Preclinical
Patents	Patent applications filed
Company; lead investigator	Nkarta Inc.; Dario Campana, National University of Singapore

He told BioCentury Nkarta is interested in partnering some of its candidates, and intends to continue developing its internal pipeline.

PEBLS FOR THOUGHT

Last month at the Immuno-Oncology 360º conference in New York, Campana presented data on another way to drive NK cell activation, via Protein Expression Blockers (PEBLs), which work by blocking the cells' inhibitory receptors.

Trager would not disclose whether Nkarta has licensed rights to the PEBL technology, although he said the company has licensed a portfolio of patent applications covering Campana's work from the National University of Singapore and St. Jude Children's Research Hospital — Campana's previous affiliation — and will likely continue to license IP covering his new technologies.

The original goal was to enable use of CAR T cells in T cell malignancies — a challenge because antigens expressed on cancerous T cells are also present on healthy T cells, which means CAR T cells carrying those antigens would kill each other.

In a *Blood Advances* study published in November, Campana's team used a PEBL molecule comprising an anti-CD7 antibody scFv domain linked to an endoplasmic reticulum-binding amino acid sequence that anchors the domain inside the cell.

The group introduced the construct into CAR T cells targeting the T cell antigen CD7 to prevent the therapeutic cells from expressing CD7 on their surfaces.

In a mouse model of T-cell acute lymphoblastic leukemia (T-ALL), the PEBL CAR Ts eliminated leukemic cells, and in mice with T-ALL patient-derived xenografts they decreased leukemia burden to undetectable levels. Control CD7-targeted CAR Ts without the PEBL decreased leukemic cell burden by 68% in the T-ALL mouse model.

At last month's conference, Campana presented unpublished data showing a PEBL designed to prevent surface expression of NKG2A — an inhibitory receptor on NK cells — decreased cancer burden in mice and extended survival compared with mock transduced NK cells.

> "We do feel there's an advantage to riding the natural biology of NK cells to maintain the good margin of safety."
>
> James Trager, Nkarta

Trager said the company is keeping an open eye on how the technology develops, and whether it might lead to any safety signals.

"We're enthusiastic about the technology and Dario has really pioneered this way to knock off cell surface receptors. But this work is still preliminary and there are some unanswered questions. When we start to knock down the inhibitors, we have to be cognizant of the risks," said Trager.

SPEEDY EXPANSION

Nkarta also has rights to a cell expansion technology from Campana that Trager said is at the "heart" of all its programs, whose primary benefit is its speed.

"We can get up to a target dose within 10-12 days with very reliable, consistent expansion that results in activated NK cells in a good state to go back into the patient," he said. The process yields an autologous product of at least 95% NK cells and less than 5% T cells.

He said techniques used by other companies generally take two weeks to over a month; few take less than 10 days.

"Patients are waiting for therapies and don't have a month to wait," said Trager.

According to Trager, Nkarta's process is quicker because it doesn't require purification of the cells before they are expanded.

In a 2009 *Cancer Research* paper, Campana described the process, which involves collecting peripheral blood mononuclear cells (PBMCs) from a patient, and partially separating out, but not fully purifying, the NK cells.

The cells are then expanded in co-culture with a feeder cell line engineered to express NK stimulatory molecules. The culture is also exposed to a low dose of the activating cytokine IL-2.

The culture conditions favor growth of NK cells over other immune cells, said Trager. "We typically expand the cells for a few days because we want them to outgrow the T cells and other cells in culture, then we engineer them. It is easier to engineer them once they are up and growing," he said.

The NK cells are further expanded in a high dose of IL-2 for several days before preconditioning the patient via lymphodepletion and then re-infusing the therapeutic cells.

Campana's group has treated patients with non-engineered NK cells expanded via the technique, but none of the studies have read out yet. While the initial studies don't provide proof of concept, "they are encouraging and speak to the ability to put these rapidly expanded cells into the clinic," said Trager. /

COMPANIES AND INSTITUTIONS MENTIONED

Fate Therapeutics Inc. (NASDAQ:FATE), San Diego, Calif.
Johnson & Johnson (NYSE:JNJ), New Brunswick, N.J.
National University of Singapore, Singapore
NantKwest Inc. (NASDAQ:NK), San Diego, Calif.
Nkarta Inc., South San Francisco, Calif.
OncoMed Pharmaceuticals Inc. (NASDAQ:OMED), Redwood City, Calif.
PersonGen Biomedicine (Suzhou) Co. Ltd., Suzhou, China
St. Jude Children's Research Hospital, Memphis, Tenn.
University Hospital Frankfurt, Frankfurt, Germany
Unum Therapeutics Inc., Cambridge, Mass.
Ziopharm Oncology Inc. (NASDAQ:ZIOP), Boston, Mass.

TARGETS AND COMPOUNDS

CD3ζ (CD247)
CD7 - CD7 molecule
DAP10 (HCST) - DNAX-activation protein 10
HER2 (EGFR2; ErbB2; neu) - Epidermal growth factor receptor 2
NKG2A (CD159a; KLRC1) - Killer cell lectin-like receptor subfamily C member 1
NKG2D (KLRK1; CD314) - Killer cell lectin-like receptor subfamily K member 1

REFERENCES

Chang, Y.-H., et al. "A chimeric receptor with NKG2D specificity enhances natural killer cell activation and killing of tumor cells." *Cancer Research* (2013)

Fujisaki, H., et al. "Expansion of highly cytotoxic human natural killer cells for cancer cell therapy." *Cancer Research* (2009)

Imamura, M., et al. "Autonomous growth and increased cytotoxicity of natural killer cells expressing membrane-bound interleukin-15." *Blood* (2014)

Martz, L. "Innate harmony." *BioCentury Innovations* (2016)

Png, Y., et al. "Blockade of CD7 expression in T cells for effective chimeric antigen receptor targeting of T-cell malignancies." *Blood Advances* (2017)

EMERGING COMPANY PROFILE

STABILIZING DAMAGED GOODS

By Karen Tkach Tuzman, Associate Editor

Methuselah Health Inc. aims to treat aging-related diseases by stabilizing proteins that have adopted dysfunctional conformations as a result of genomic or environmental damage. By studying damaged proteins instead of DNA, the newco can find disease-driving targets that genomics studies miss.

"We started Methuselah in response to the realization that genome-wide association studies (GWAS) were not going to be the answer to target identification for the bulk of common degenerative diseases of middle and old age," CEO and Medicxi co-founder and Partner David Grainger told BioCentury. "The core of the Methuselah hypothesis is that it's proteome instability, rather than DNA-encoded loss of function" that causes late-onset diseases like Type II diabetes, Alzheimer's disease and osteoarthritis.

Methuselah focuses on proteins with post-translational modifications (PTMs) caused by oxidation, cross-linking, methylation and other chemical reactions in response to UV light exposure and other stressors. Grainger said these "unregulated PTMs" can lock proteins into dysfunctional, disease-driving states, particularly if the protein is mutated and thus prone to misfolding.

To identify targets, the newco analyzes blood or tissue samples from patients, healthy volunteers or animal models on its mass spectrometry platform to look for proteins with damaging PTMs, then homes in on damaged proteins that are present at high levels in the disease state.

Grainger that while others have used mass spectrometry to study specific types of PTMs, "what had not been solved was the specific challenges that let you look at all possible proteomic damage all at the same time, in an unbiased, non-hypothesis-driven sense," adding that Methuselah's platform simultaneously detects all possible PTMs across the whole proteome.

The newco validates the targets' role in disease by generating destabilizing mutations in the corresponding genes of animals and looking for signs of disease.

After pinpointing a target, Methuselah screens its small molecule libraries for compounds that protect target proteins from damage under high-stress conditions. "You change that equilibrium, so it spends more of its time in the stable conformation that isn't susceptible to" damaging PTMs, Grainger said. "That small molecule will, over time, cause the depletion of the damaged protein" and natural clearance processes will gradually put the tissue to right.

Methuselah has identified an undisclosed target for dyslipidemia in Type II diabetes and is screening for small molecules that stabilize it.

Grainger said the company is negotiating exclusive partnerships with undisclosed pharmas in other disease areas, including neurodegeneration and autoimmunity, to finance development of its internal pipeline without having to raise further equity.

METHUSELAH HEALTH INC., Wilmington, Del.
Technology: Proteomic platform to identify targets with disease-associated protein damage and small molecules that stabilize those targets
Disease focus: Endocrine/metabolic
Clinical status: Preclinical
Founded: 2015 by David Grainger, David Mosedale, Miroslav Radman
University collaborators: Mediterranean Institute for Life Sciences
Corporate partners: None
Number of employees: 3
Funds raised: $4 million
Investors: Medicxi
CEO: David Grainger
Patents: None issued

Methuselah has raised $4 million from Medicxi, which Grainger expects will give the company runway through 2019. The company has not disclosed whether it has filed any patent applications.

At least one other company, ViewPoint Therapeutics Inc., is developing compounds that stabilize protein conformations in aging-related diseases. Its lead candidate VP1-001, a small molecule stabilizer of α-crystallin, is in preclinical testing for cataracts.

Takeda Pharmaceutical Co. Ltd. markets the PPARγ agonist Actos pioglitazone for dyslipidemia in Type II diabetes. At least two other companies, Genfit S.A. and GW Pharmaceuticals plc, have products in the clinic for the indication.

Marketed TZDs like Actos have black box warnings for congestive heart failure and edema. Methuselah did not respond in time for publication to say how its compounds compare with TZDs or other products .

COMPANIES AND INSTITUTIONS MENTIONED

Genfit S.A. (Euronext:GNFT), Loos, France
GW Pharmaceuticals plc (NASDAQ: GWPH), Cambridge, U.K.
Methuselah Health Inc., Wilmington, Del.
Takeda Pharmaceutical Co. Ltd. (Tokyo:4502), Osaka, Japan
ViewPoint Therapeutics Inc., San Francisco, Calif.

TARGETS

PPARα - Peroxisome proliferation activated receptor α

TRANSLATION IN BRIEF

RAISING KRAS

By Mary Romeo, Staff Writer

For the first time, a joint research team from Wellspring Biosciences LLC and the Janssen Research & Development unit of Johnson & Johnson (NYSE:JNJ) showed a small molecule K-Ras (KRAS) inhibitor worked in animal models of cancer. The results could pave the way for clinical development of a therapy against the target, a member of the notoriously "undruggable" Ras family of proteins.

> "All signs are a go to get this into humans."
>
> Kevan Shokat, UCSF

Kevan Shokat, co-founder of drug discovery incubator Wellspring and professor at the University of California San Francisco, told BioCentury the latest study means "all signs are a go to get this into humans."

Mutations in Ras are found in about 30% of cancers and lock the GTP-binding proteins in a constitutively active state. The companies are targeting one of the most common mutations, a G12C substitution that occurs in 11-16% of lung cancers and in 1-4% of pancreatic and colon cancers.

Because Ras is highly dynamic and has few binding pockets, it has been hard to find compounds that bind with high affinity.

In 2016 the J&J and Wellspring team developed small molecules that covalently bind the mutant cysteine and showed the compounds allosterically prevented KRAS from binding GTP, trapping it in its GDP-bound inactive state (see "p53 and RAS: Back from the Dead." *BioCentury Innovations* (Dec. 15, 2016)).

Shokat told BioCentury the challenge was to get enough of the small molecule to maintain sufficient exposure and duration *in vivo* to bind KRAS. "We showed the crystal structure and *in vitro* effects, but no effect on tumors themselves," he said.

In the new study, published in *Cell*, the team optimized the small molecules and selected one, dubbed ARS-1620, that showed a tenfold improvement in potency compared with the parent compound. In cancer cell lines harboring the G12C mutation, ARS-1620 increased target engagement, decreased Ras signaling, and decreased cell growth compared with an inactive analog.

In mice, ARS-1620 had oral bioavailability of 60% and average peak tumor concentrations of 1.5-5.5 μM, with a G12C target occupancy of 75-90%.

In patient-derived xenograft mouse models of cancer with the G12C mutation, oral ARS-1620 decreased tumor growth compared with vehicle. ARS-1620 did not show effects on tumors in models lacking the G12C mutation, suggesting the small molecule is mutant-specific. Shokat said this "wild-type sparing property is a big advantage for humans," as it is unlikely to inhibit the KRAS pathway in normal tissues.

In 2013, Wellspring spinout Araxes Pharma LLC formed a partnership with Janssen to develop the small molecule KRAS inhibitors. Araxes is responsible for development through Phase I proof of concept, and Janssen is responsible for further development and will have exclusive, worldwide rights to commercialize any products.

Lead author Yi Liu told BioCentury the team is continuing to optimize the G12C inhibitor, but declined to provide a development timeline for entering the clinic. Liu is CSO at Wellspring and former CSO of Kura Oncology Inc. (NASDAQ:KURA).

Shokat said the results suggest the strategy could go beyond G12C and work against other KRAS mutations. He said his group at UCSF is attempting to develop a molecule targeting G12D-KRAS, which is found in pancreatic cancer.

Shokat added that his group has additional data suggesting a small molecule inhibitor could target KRAS in its active, GTP-bound state by binding in a deeper groove in the same pocket and allosterically modulating the protein. That could be relevant for targeting mutations like G12V and G12D — another KRAS form that Shokat is focused on. *Janes, M., et al. "Targeting KRAS mutant cancers with a covalent G12C-specific inhibitor." Cell (2018)*

FILLING THE MYELOID VOID

By Chris Lieu, Staff Writer

A Genentech study in *Cell Reports* cranked up the resolution on neuroinflammatory states in the brain by identifying gene expression patterns in microglia that are associated with various types of CNS disease. The findings pave the way for identifying unique inflammatory mechanisms in each disease and determining whether those mechanisms are harmful or helpful.

Microglia, the myeloid cells of the brain, are thought both to contribute to and protect against many CNS diseases, but little is known about how the cells compare to their peripheral counterparts.

The dogma has been that activated microglia adopt one of two states: a proinflammatory state (M1) or a phagocytic state involved in tissue repair or restoration (M2). But this view, which rests largely on histology and a handful of genetic markers, is breaking down as more sophisticated tools for assessing cellular phenotypes become available.

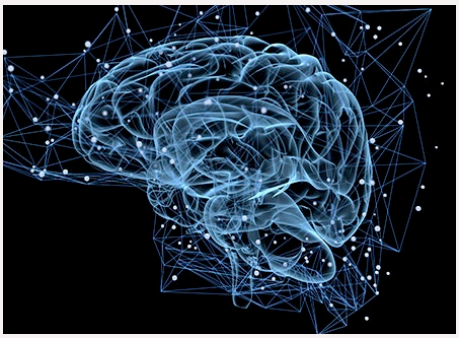
Many of the markers that researchers have used are not reliable because they may not be expressed in any brain myeloid cell populations, said David Hansen, author on the January study and a scientist at the Genentech Inc. unit of Roche (SIX:ROG; OTCQX:RHHBY).

"Many authors still focus on a single gene, or a small number of genes, when analyzing microglial activation," added author Brad Friedman, also a Genentech scientist.

In its study, the team conducted a meta-analysis of CNS myeloid transcriptional profiles in mouse models of various CNS diseases, including neurodegenerative diseases, cancers, infections and inflammatory conditions. The analysis identified co-regulated sets of genes — dubbed "modules" — that were up-regulated in activated microglia, distinct for each group of diseases, and more diverse than previously thought.

For example, the neurodegeneration-related module, consisting of 134 genes, occurred in models of Alzheimer's disease, amyotrophic lateral sclerosis and multiple sclerosis, but not in models of lipopolysaccharide (LPS)-induced neuroinflammation or viral infection of the CNS.

> "Integrating these gene modules with human genetics results may help to further prioritize which genes could be good drug targets."
>
> Brad Friedman, Genentech



THINKSTOCK

The team also evaluated how the modules were induced within individual cell subpopulations, including resting microglia and disease-associated microglia, from single-cell RNA-seq data in the 5XFAD mouse model of AD. The subsets of interferon-activated and proliferating microglia each expressed a module that was distinct from the neurodegeneration-related module expressed by the disease-associated microglia.

In addition, the neurodegeneration-related module was induced in brain tissue samples from both AD patients and AD mouse models, whereas the neutrophil-monocyte and LPS-related modules were elevated only in patient tissue. Those findings suggested the human disease may involve more inflammatory signaling or peripheral immune cell infiltration than mouse models, the authors wrote in the paper.

Hansen added that the relevance of microglial activation states in mouse models to human disease, and which modules are best at approximating responses in human disease, are unclear.

He said the group's next steps include studying expression profiles of microglia, both as populations and single cells, from patients with CNS diseases.

In the paper, the authors wrote it is unclear whether the modules play a role in progression of their respective diseases. However, Friedman said, "integrating these gene modules with human genetics results may help to further prioritize which genes could be good drug targets."

Genentech has not patented its *Cell Reports* findings, and the data sets used to conduct the meta-analysis are available through a searchable gene expression database, The Myeloid Landscape. *Friedman, B., et al. "Diverse brain myeloid expression profiles reveal distinct microglial activation states and aspects of Alzheimer's disease not evident in mouse models." Cell Reports (2018)*

MOA THE SAME

By Jaime de León, Staff Writer

An ETH Zurich-led team has developed a high throughput method of comparative metabolomics that can predict MOAs for antibiotics, and used it to suss out how a library of previously uncharacterized GSK compounds killed bacteria.

While the team's *Science Translational Medicine* study showed proof of concept in mycobacterial culture, the method could predict MOAs of compounds against any pathogenic bacteria, bypassing the need to generate a library of bacterial mutants to determine a compound's target.

The team began by generating a set of metabolomics profiles for *Mycobacterium smegmatis* treated with 62 reference compounds that had 17 known MOAs, including inhibition of bacterial DNA replication, protein synthesis, cell wall synthesis and folate synthesis. The team used a chromatography-free mass spectrometry method that takes about a minute to collect data — 40-50 times faster than conventional chromatography-based mass spectrometry, according to author Mattia Zampieri, a researcher at ETH Zurich.

"These compounds actually are likely to have modes of action similar to already known antibiotics."

Mattia Zampieri, ETH Zurich

Abundances of over 1,000 bacterial metabolites, collected at six time points following compound exposures, were used to create dynamic profiles for each compound that were indicative of underlying MOAs.

"Once you have this reference profile, you can actually use any molecule you want to try to assess whether the metabolic profile of the molecule is significantly similar to any of the reference compounds," said Zampieri.

The team then collected metabolic profiles of *M. smegmatis* in culture treated with 212 compounds from GlaxoSmithKline plc (LSE:GSK; NYSE:GSK) that phenotypic screening had shown killed the related species, *M. tuberculosis*, with low toxicity to human cells but whose MOAs were otherwise unknown.

Based on similarities between metabolic response profiles of the reference and GSK compounds, Zampieri said, "We found that more than 70% of these compounds actually are likely to have modes of action similar to already known antibiotics. And this despite the fact that they have chemical structures that are very different from any known antibiotic."

However, the authors wrote in the paper that the method alone could not be used to determine whether the compounds modulated known antibiotic targets directly or via indirect mechanisms, such as binding to other proteins involved in the same cellular process.

To validate the metabolomic analysis, the study authors used an *in vitro* enzyme assay to show that four of the GSK compounds predicted to have a mode of action similar to trimethoprim, which non-competitively inhibits bacterial dihydrofolate reductase (DHFR), did indeed directly inhibit the target.

Three more GSK compounds induced metabolic profiles in *M. smegmatis* similar to that of quinolones, a result the team confirmed by showing the compounds enhanced recombinase A (recA) promoter activity in *E. coli* — a known effect of quinolone-induced DNA damage. By contrast, three GSK compounds the method predicted to have MOAs unlike quinolones did not increase recA promoter activity.

Sixteen other compounds had metabolic signatures unlike any of the reference compounds, suggesting they had novel MOAs, the team wrote in the paper.

The team also wrote that extending the method to the prediction of compound MOAs in other non-mycobacterial species would require building a new set of reference profiles in that species or a related one.

Zampieri added the method can extend to compound classes other than antimicrobials, including those that alter the bacterial metabolome but do not affect cell growth. The team is using the method in the latter way to predict MOAs for non-lethal agents that could be used in combination with lethal antibiotics to boost efficacy. He said the team has no plans to patent or license the method.

GSK did not respond to requests for comment. *Zampieri, M., et al. "High-throughput metabolomic analysis predicts mode of action of uncharacterized antimicrobial compounds." Science Translational Medicine (2018)*

DISTILLERY

THE DISTILLERY brings you this week's most essential scientific findings in therapeutics, distilled by *BioCentury Innovations* editors from a weekly review of more than 400 papers in 41 of the highest-impact journals in the fields of biotechnology, the life sciences and chemistry. The Distillery goes beyond the abstracts to explain the commercial relevance of featured research, including licensing status and companies working in the field, where applicable. This week in therapeutics includes important research findings on targets and compounds, grouped first by disease class and then alphabetically by indication.

THERAPEUTICS

CANCER

INDICATION: Acute myelogenous leukemia (AML)

In vitro, cell culture and mouse studies identified a pyrazolo-tetrahydroquinolinone-based GSK3A inhibitor that could help treat AML. Chemical synthesis, *in vitro* activity and HEK cell-based binding assays of pyrazolo-tetrahydroquinolinone analogs yielded a compound that bound GSK3A with a K_d of 4.8 μM and inhibited the enzyme with an IC_{50} of 66 nM. In six human AML cell lines and in blasts from five AML patients, the compound decreased colony formation compared with vehicle, whereas in normal human hematopoietic stem cells, the compound had no effect. In a syngeneic and two xenograft mouse models of AML, the compound increased survival and decreased disease progression. Next steps could include testing the compound in additional models of AML.

TARGET/MARKER/PATHWAY: Glycogen synthase kinase 3 α (GSK3A)
LICENSING STATUS: Patent and licensing status unavailable
PUBLICATION DETAILS: Wagner, F. et al. Sci. Transl. Med.; published online March 7, 2018
doi:10.1126/scitranslmed.aam8460
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INDICATION: Breast cancer

Studies in mice suggest a peptide-based EPHA2 agonist conjugated to paclitaxel could help treat triple-negative breast cancer (TNBC). Conjugation of an EPHA2 agonist peptide to the generic chemotherapy paclitaxel and *in vitro* binding assays yielded a peptide-drug conjugate that bound EPHA2 with a K_d of 4.9 μM. In a xenograft mouse model of TNBC, the conjugate decreased the number of lung metastases compared with vehicle or Abraxane nab-paclitaxel. In another xenograft mouse model of TNBC, the conjugate decreased numbers of cancer cells in the blood compared with Abraxane. Next steps could include testing the peptide-drug conjugate in additional models of TNBC.

Celgene Corp., BeiGene Ltd., Biocon Ltd., Green Cross Corp., Specialised Therapeutics Australia Pty. Ltd. and Taiho Pharmaceutical Co. Ltd. market Abraxane, an albumin-stabilized nanoparticle formulation of paclitaxel, to treat breast cancer. Celgene, BeiGene, Specialised Therapeutics Australia and Taiho Pharmaceutical also market the compound to treat non-small cell lung cancer (NSCLC), gastric and pancreatic cancers and have the compound in Phase III testing to treat melanoma and Phase I/II testing to treat esophageal cancer.

TARGET/MARKER/PATHWAY: EPH receptor A2 (EPHA2)
LICENSING STATUS: Patent and licensing status unavailable
PUBLICATION DETAILS: Salem, A. et al. *J. Med. Chem;* published online, Feb. 22, 2017.
doi:10.1021/acs.jmedchem.7b01837
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THERAPEUTICS

CANCER

INDICATION: Cancer; breast cancer; chronic lymphocytic leukemia (CLL); lymphoma; neuroendocrine tumors

Patient sample, cell culture and mouse studies suggest inhibiting XBP1 or its activator IRE1 could help treat MYC- or MYCN-driven cancers. In breast cancer patient tumor samples, high levels of IRE1 were associated with high levels of MYC. In a MYCN-driven human neuroblastoma cell line, shRNA targeting XBP1 decreased viability compared with scrambled shRNA. In five MYC-driven human Burkitt's lymphoma cell lines and a MYC-driven CLL cell line, a tool compound IRE1 inhibitor decreased proliferation and viability compared with vehicle. In xenograft mouse models of neuroblastoma and MYC-driven Burkitt's lymphoma, tumor-specific XBP1 knockdown and the IRE1 inhibitor decreased tumor growth compared with normal XBP1 expression and vehicle, respectively. In two patient-derived xenograft (PDX) mouse models of MYC-driven breast cancer, a second tool compound IRE1 inhibitor decreased tumor growth and increased survival compared with vehicle, and in one of the PDX models, the inhibitor plus the generic chemotherapy docetaxel decreased tumor growth and increased survival compared with either agent alone. Next steps could include identifying and testing inhibitors of XBP1 in the models.

TARGET/MARKER/PATHWAY: X-box binding protein 1 (XBP1); endoplasmic reticulum to nucleus signaling 1 (IRE1; ERN1; IRE1A); v-myc myelocytomatosis viral oncogene homolog (MYC; c-Myc); v-myc myelocytomatosis viral related oncogene neuroblastoma derived (MYCN; NMYC)
LICENSING STATUS: Patent and licensing status unavailable
PUBLICATION DETAILS: Zhao, N. et al. *J. Clin. Invest.*; published online Feb. 26, 2018
doi:10.1172/JCI95873
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email: Xi.Chen@bcm.edu
LICENSING STATUS: Patented; available for licensing and partnering
PUBLICATION DETAILS: Xie, H. et al. *J. Clin. Invest.*; published online Feb. 26, 2018
doi:10.1172/JCI95864
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INDICATION: Breast cancer; liver cancer

Mouse studies suggest inhibiting the USP2a-splicing isoform of USP2 could help treat metastatic breast and liver cancers. In a xenograft mouse model of metastatic liver cancer, a tool compound that inhibits USP2a increased survival compared with vehicle. In a second mouse model of metastatic breast cancer, the compound decreased metastasis to the lung. Next steps could include identifying and testing USP2a inhibitors in models of breast or liver cancer.

TARGET/MARKER/PATHWAY: Ubiquitin specific peptidase 2 (USP2; USP9)
LICENSING STATUS: Patent and licensing status unavailable
PUBLICATION DETAILS: Zhao, Y. et al. *Cell Rep.*; published online Feb. 27, 2018
doi:10.1016/j.celrep.2018.02.007
CONTACT: Bo Zhong, Wuhan University, Wuhan, China
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INDICATION: Cancer; cervical cancer; liver cancer; melanoma

In vitro, cell culture and mouse studies identified an imidazole-carbazole conjugate-based MYC inhibitor that could help treat MYC-driven cancer. Chemical synthesis and *in vitro* binding assays on imidazole-carbazole conjugate analogs yielded a compound that bound the G-quadruplex DNA region of the MYC gene with a K_d of 0.1μM. In MYC-driven human cervical squamous cell carcinoma (SCC), HeLa, hepatocellular carcinoma and melanoma cell lines, the compound was cytotoxic with IC_{50} values of 2.1-4.2 μM. In the MYC-driven human cervical SCC cell line, the compound increased apoptosis compared with no treatment. In a xenograft mouse model of MYC-driven cervical SCC, the compound decreased tumor growth compared with vehicle. Next steps could include testing the compound in additional models of MYC-driven cancer.

TARGET/MARKER/PATHWAY: v-myc myelocytomatosis viral oncogene homolog (MYC; c-Myc)
LICENSING STATUS: Patent and licensing status unavailable
PUBLICATION DETAILS: Hu, M.-H. et al. *J. Med. Chem.*; published online Feb. 23, 2018
doi:10.1021/acs.jmedchem.7b01697
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THERAPEUTICS

CANCER

INDICATION: Melanoma

Mouse studies identified a commensal bacteria-derived adenine analog that could help treat melanoma. In a mouse model of the disease, an adenine analog isolated from the commensal bacterial species *Staphylococcus epidermidis* decreased tumor size compared with vehicle. In another mouse model of melanoma, topical administration of the *S. epidermidis* strain that produces the analog decreased the number of tumors compared with an *S. epidermidis* strain that did not secrete the analog. Next steps could include testing the analog in models of other skin cancers.

TARGET/MARKER/PATHWAY: An undetermined target

LICENSING STATUS: Patent application filed; licensed to MatriSys Bioscience Inc.

PUBLICATION DETAILS: Nakatsuji, T. et al. *Sci. Adv.*; published online Feb. 28, 2018

doi:10.1126/sciadv.aao4502

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INDICATION: Melanoma

Studies in patient samples, cell culture and mice suggest a miR-378a-3p variant or inhibiting its target PARVA could help treat melanoma. In patients, high tumor levels of PARVA were associated with poor survival. In a human melanoma cell line, shRNA targeting PARVA decreased invasiveness compared with non-specific shRNA. In a xenograft mouse model of melanoma, a variant of miR-378a-3p containing an adenosine (A) to inosine (I) substitution generated by RNA editing — a post-transcriptional modification down-regulated in melanoma — or tumor-specific shRNA-mediated knockdown of PARVA decreased primary tumor volume and numbers of lung metastases compared with an unedited version of miR-378a-3p or non-specific shRNA. Next steps could include identifying and testing PARVA inhibitors in models of melanoma.

TARGET/MARKER/PATHWAY: MicroRNA-378a-3p (miR-378a-3p); parvin α (PARVA)

LICENSING STATUS: Patent and licensing status unavailable

PUBLICATION DETAILS: Velazquez-Torres, G. et al *Nat. Commun.*; published online Jan. 31, 2018

doi:10.1038/s41467-018-02851-7

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INDICATION: Non-small cell lung cancer (NSCLC)

Patient sample and mouse studies suggest inhibiting SHP-2 could help treat ALK inhibitor-resistant NSCLC. In four ALK inhibitor-resistant patient-derived NSCLC cell lines, the small molecule ALK inhibitor Zykadia ceritinib plus either shRNA targeting SHP-2 or the selective SHP-2 inhibitor SHP099 decreased colony formation compared with any of the agents alone. In three xenograft mouse models of ALK inhibitor-resistant NSCLC, Zykadia plus SHP099 decreased tumor growth compared with Zykadia alone. Next steps by Novartis AG and Tango Therapeutics Inc. could include testing the combination therapy in models of other ALK inhibitor-resistant cancers.

Novartis and Helsinn Healthcare S.A. market Zykadia to treat NSCLC and have the compound in Phase II testing to treat solid tumors.

Novartis has SHP099 in preclinical testing to treat cancer.

Tango Therapeutics has undisclosed programs in discovery for cancer.

TARGET/MARKER/PATHWAY: Src homology protein tyrosine phosphatase 2 (SHP-2; SHPTP2; PTPN11); anaplastic lymphoma kinase (ALK)

LICENSING STATUS: Patent and licensing status unavailable

PUBLICATION DETAILS: Dardaei, L. et al. *Nat. Med.*; published online March 5, 2018

doi:10.1038/nm.4497

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THERAPEUTICS

CANCER

INDICATION: Prostate cancer

Cell culture and mouse studies suggest inhibiting PDHA1 could help treat prostate cancer. In tumor samples from patients, levels of PDHA1 were higher than in normal prostate tissue. In three human prostate cancer cell lines, shRNA targeting PDHA1 decreased proliferation and tumorsphere formation compared with scrambled shRNA. In a mouse model of prostate cancer, prostate-specific knockout of PDHA1 decreased tumor growth and invasiveness and cell proliferation in tumors compared with normal PDHA1 expression. In the mouse model and in three xenograft mouse models of prostate cancer, a tool compound PDHA1 inhibitor decreased tumor growth compared with vehicle. Next steps could include identifying and testing PDHA1 inhibitors in additional models of prostate cancer.

TARGET/MARKER/PATHWAY: Pyruvate dehydrogenase E1 α 1 subunit (PDHA1)
LICENSING STATUS: Patent and licensing status unavailable
PUBLICATION DETAILS: Chen, J. et al. *Nat. Genet.*; published online Jan. 15, 2018
doi:10.1038/s41588-017-0026-3
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CARDIOVASCULAR

INDICATION: Block coagulation

Mouse studies suggest that ANGPT1 could help treat disseminated intravascular coagulation (DIC) associated with sepsis. In lipopolysaccharide (LPS)-treated human umbilical vein endothelial cells (HUVECs), ANGPT1 decreased levels of clotting factors compared with no treatment. In a mouse model of sepsis-associated DIC, IV injection of ANGPT1 encoded by an adenoviral vector decreased clotting and serum levels of clotting factors compared with GFP. Next steps could include testing ANGPT1 in additional models of sepsis-associated DIC.

TARGET/MARKER/PATHWAY: Angiopoietin 1 (ANG1; ANGPT1)
LICENSING STATUS: Patent application filed; licensing status unavailable
PUBLICATION DETAILS: Higgins, S. et al. *J. Clin. Invest.*; published online March 5, 2018
doi:10.1172/JCI97488
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INFECTIOUS DISEASE

INDICATION: Candida; fungal infection

Cell culture studies identified a glucan-based NKp30 activator that could help treat *Cryptococcus neoformans* and *Candida albicans* infections. *In vitro*, a *C. neoformans* and *C. albicans* cell wall glucan bound NKp30 with higher affinity than a structurally related protein. In a human NK cell line co-cultured with *C. neoformans* or *C. albicans*, the compound increased NK cell-mediated killing of fungal cells compared with no treatment. In primary NK cells from healthy or HIV-infected patients cocultured with *C. neoformans*, the compound increased NK cell-mediated killing of fungal cells compared with no treatment. Next steps could include testing the compound in animal models of fungal infection.

TARGET/MARKER/PATHWAY: Natural killer p30 receptor (NKp30; NCR3; CD337)
LICENSING STATUS: Patent and licensing status unavailable
PUBLICATION DETAILS: Li, S. et al. *Nat. Commun.*; published online Feb. 21, 2018
doi:10.1038/s41467-018-03014-4
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THERAPEUTICS

INFECTIOUS DISEASE

INDICATION: Zika virus

Mouse and studies suggest inhibiting *A. aegypti* LTRIN could help treat Zika infection. LTRIN is a mosquito salivary protein transmitted to a host with the virus that suppresses host immunity. In a mouse model of Zika infection, an antibody against *A. aegypti* LTRIN decreased brain viral load compared with a control antibody. Next steps could include testing the antibody in mouse models of established Zika infection.

TARGET/MARKER/PATHWAY: Aedes aegypti LTRIN (A. *aegypti* LTRIN)
LICENSING STATUS: Patent and licensing status unavailable
PUBLICATION DETAILS: Jin, L. et al. *Nat. Immunol.*; published online March 5, 2018
doi:10.1038/s41590-018-0063-9
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MUSCULOSKELETAL

INDICATION: Bone repair

Mouse studies suggest inhibiting TGFB1 could help treat heterotopic ossification, an ectopic formation of bone in extraskeletal tissues. In a mouse model of heterotopic ossification, knockout of TGFB1 in immune cells decreased bone volume in the Achilles tendon compared with normal TGFB1 expression. In two other mouse models of the disease, a TGFB1-neutralizing antibody decreased osteoblast numbers in the bone marrow, and bone formation in the Achilles tendon and hamstrings compared with an isotype-matched control antibody. Next steps include identifying and testing small molecule TGFB inhibitors in animal models of heterotopic ossification.

TARGET/MARKER/PATHWAY: Transforming growth factor β 1 (TGFB1)
LICENSING STATUS: Patent application filed; available for licensing
PUBLICATION DETAILS: Wang, X. et al. *Nature Commun.*; published online Feb. 7, 2018
doi:10.1038/s41467-018-02988-5
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NEUROLOGY

INDICATION: Spinal muscular atrophy (SMA)

Fruit fly and mouse studies suggest allosteric modulation of the AF2 domain of the androgen receptor could help treat spinal bulbar muscular atrophy (SBMA). In a transgenic fruit fly model of SBMA expressing the mutant human androgen receptor that causes the disease, tool compounds that allosterically modulate AF2 — which is involved in binding co-regulatory proteins — plus dihydrotestosterone (DHT) decreased neuromuscular junction defects and increased locomotor function and survival compared with vehicle. In a mouse model of SBMA, one of the AF2 modulators decreased spinal cord and skeletal muscle degeneration and increased body weight, grip strength and other markers of functional recovery. Next steps include seeking partners for clinical testing of AF2 modulators in SBMA.

TARGET/MARKER/PATHWAY: Androgen receptor
LICENSING STATUS: Patent application filed; available for licensing or partnering
PUBLICATION DETAILS: Badders, N. et al. *Nat. Med.*; published online March 5, 2018
doi:10.1038/nm.4500
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THERAPEUTICS

OPHTHALMIC DISEASE

INDICATION: Retinal detachment

Dog studies suggest BEST1 gene therapy could help treat bestrophinopathies and other degenerative retinal diseases caused by BEST1 mutations. In a dog model of bestrophinopathy, subretinal injection of human BEST1 encoded in an adeno-associated viral (AAV) vector decreased subretinal lesions and retinal microdetachments compared with vehicle. Next steps could include testing the gene therapy in other models of BEST1-mutant retinal diseases.

TARGET/MARKER/PATHWAY: Bestrophin 1 (BEST1)

LICENSING STATUS: Patent and licensing status unavailable

PUBLICATION DETAILS: Guziewicz, K. et al. *Proc. Natl. Acad. Sci. USA*; published online March 5, 2018

doi:10.1073/pnas.1720662115

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TECHNIQUES

ASSAYS AND SCREENS

TECHNOLOGY: Binding assays; cellular assays

A cell-based method for screening binding interactions could identify ligands of glycan-binding proteins as leads for drug development. The method utilizes a glycan array that consists of several sugar analogs modified with a library of azide functional groups, displayed on Lec2-mutant Chinese hamster ovary (CHO) cells in multiwell plates by linking to the cells' native surface glycans. Proteins of interest are screened against the array and analogs that bind are identified using flow cytometry-based detection. In proof-of-concept experiments using the osteoclast differentiating protein sialic acid binding Ig-like lectin 15 (SIGLEC15), the method identified three glycan analogs with high bind affinities for SIGLEC15. In a human osteoprogenitor cell-based model of osteoclast formation, co-culture with Lec2-mutant CHO cells expressing one of the three analogs decreased osteoclast formation compared with unmodified Lec2-mutant CHO cells. Next steps could include using the method to identify high-affinity ligands of other SIGLEC proteins.

DESCRIPTION: Cell-based glycan array for identifying ligands of glycan-binding proteins for drug development leads

LICENSING STATUS: Unpatented; licensing status not applicable

PUBLICATION DETAILS: Briard, J. et al. *Nat. Commun.*; published online Feb. 28, 2018

doi:10.1038/s41467-018-03245-5

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TECHNOLOGY: Cellular assays; bioinformatics screen

A yeast-based method coupled with bioinformatics could help identify tumor antigens that stimulate orphan TCRs for use in cancer T cell therapies and vaccines. The method involves screening TCRs derived from cytotoxic tumor infiltrating lymphocytes (TILs) in patient tumor samples against a library of 400 million synthetic antigens that are covalently linked to a human leukocyte antigen (HLA) molecule tethered to yeast. An algorithm based on exome- and proteome-sequencing data of patient-derived tumor samples then predicts human counterparts of the synthetic antigens enriched in the library by the yeast screen. Screening of 20 TCRs expressed by TILs in colorectal tumors against the antigen library yielded four TCRs, for which the algorithm predicted 56 cognate human antigens. In co-culture, T cells engineered to express the four TCRs were stimulated by a human antigen-presenting cell line pretreated with the antigens. Next steps could include using the results from the colorectal cancer screen to develop a T cell therapy or vaccine, and using the screening method to identify tumor antigens for TCRs in other tumor types.

DESCRIPTION: Yeast- and algorithm-based method to identify tumor antigens of orphan T cell receptors (TCRs) for use in cancer T cell therapies and vaccines

LICENSING STATUS: Patent and licensing status unavailable

PUBLICATION DETAILS: Gee, M. et al. *Cell*; published online Dec. 21, 2017

doi:10.1016/j.cell.2017.11.043

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TECHNIQUES

BIOMARKERS

TECHNOLOGY: Tissue markers

Tumor levels of PHLDA1 could help predict resistance to RTK inhibitors in endometrial, breast and other cancers. In tumor samples from patients with metastatic breast cancer or locally advanced non-metastatic renal cancer treated with various RTK inhibitors, low levels of PHLDA1 protein were associated with acquired drug resistance. Transcriptomic profiling of human endometrial cancer cell lines harboring activating mutations in keratinocyte growth factor receptor (KGFR; FGFR2; CD332) identified an association between low expression levels of PHDLA1 mRNA and resistance to inhibitors of fibroblast growth factor receptor (FGFR) or FGFR1 (CD331). In one of the corresponding wild-type cancer cell lines, shRNA targeting PHLDA1 decreased sensitivity to the FGFR inhibitor AZD4547 compared with a scrambled shRNA, and culturing for 14 days in 1 µM AZD4547 induced drug resistance comparable to that observed in PHLDA1-knockdown cells, as measured by proliferation rates. In two epidermal growth factor receptor 2 (HER2; EGFR2; ErbB2; neu)-positive breast cancer cell lines, induction of drug resistance with the HER1/HER2 inhibitor Tykerb lapatinib or the anti-HER2 antibody Herceptin trastuzumab decreased PHLDA1 protein levels compared with vehicle or an IgG control. In a xenograft mouse model of HER2-positive breast cancer, Herceptin decreased tumor levels of PHLDA1 mRNA compared with the control IgG. Next steps include using PHLDA1 expression to predict which patients would respond to drug therapies.

AstraZeneca plc has AZD4547 in Phase II testing to treat solid tumors.

Roche, its Genentech Inc. unit and Chugai Pharmaceutical Co. Ltd. market Herceptin for HER2-positive breast and gastric cancers.

Novartis AG and Eddingpharm Inc. market Tykerb for advanced or metastatic breast cancer and have the compound in clinical testing for multiple other cancers.

DESCRIPTION: Tumor cell levels of pleckstrin homology like domain family A member 1 (PHLDA1) to predict resistance to receptor tyrosine kinase (RTK) inhibitors in endometrial and breast cancers

LICENSING STATUS: Patent and licensing status unavailable

PUBLICATION DETAILS: Fearon, A. et al. *Cell Rep.*; published online Feb. 27, 2018

doi:10.1016/j.celrep.2018.02.028

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DISEASE MODELS

TECHNOLOGY: Transgenics and knockouts

BAP1-knockout, transgenic mice with melanocyte-specific expression of mutant human GNA11 could be used to screen therapies for metastatic uveal melanoma. Mice with systemic BAP1 knockout and tamoxifen-induced expression of the human GNA11 Q209L mutation in melanocytes recapitulated the high proliferation of metastatic tumors in the skin observed in metastatic uveal melanoma patients. Next steps could include using the model to screen metastatic uveal melanoma therapies.

DESCRIPTION: Mice with BRCA1-associated protein 1 (BAP1) knockout expressing mutant human G protein subunit α 11 (GNA11) as models of metastatic uveal melanoma

LICENSING STATUS: Patent and licensing status unavailable

PUBLICATION DETAILS: Moore, A. et al. *Cell Rep.*; published online Feb. 27, 2018

doi:10.1016/j.celrep.2018.01.081

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TECHNIQUES

DRUG DELIVERY

TECHNOLOGY: Nanoparticles

Photothermal-activated, tumor-targeted nanoparticles could be used to deliver cancer therapies. The nanoparticles consist of β-cyclodextrin-based polyrotaxane and the photothermal agent perylene diimide to enable release of drug cargo in response to near-infrared (NIR) laser irradiation, linked to a tumor-targeting cyclic peptide. In xenograft mouse models of breast, cervical and lung cancers, nanoparticles targeting integrin $\alpha_v\beta_3$ (CD51/CD61) and loaded with the topoisomerase inhibitor camptothecin or paclitaxel, plus NIR laser irradiation at the tumor site, decreased tumor growth and increased survival compared with the drug-loaded nanoparticles alone. Next steps could include testing the drug-loaded nanoparticles targeting other cancer antigens in additional models of cancer.

DESCRIPTION: Photothermal-activated, tumor-targeted nanoparticles to deliver therapies for cancer

LICENSING STATUS: Patent and licensing status unavailable

PUBLICATION DETAILS: Yu, G et al. *Nat. Commun.*; published online Feb. 22, 2018

doi:10.1038/s41467-018-03119-w

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