Research Funded by KATA

(includes partially and fully funded projects)

KATA has funded over 2 million dollars in Fanconi anemia research, including research into the causes, treatment and prevention of cancers that are particularly harmful to FA patients.

Quercetin Chemoprevention for Squamous Cell Carcinoma in Patients with Fanconi Anemia
(Dr. Parinda Mehta, MD at Cincinnati Children's Hospital)
Excessive toxicity from chemotherapy and radiation makes treatment for squamous cell carcinoma (SCC) in FA challenging and often leads to dismal outcomes. Thus, there is clearly a need for a new approach both for prevention and/or treatment that has fewer and less severe side effects. Previous studies conducted by Dr. Mehta showed that a naturally occurring antioxidant, Quercetin, is safe and well tolerated in pre-HCT patients with FA. This study will test whether Quercetin prevents the development of SCC in post-transplant FA patients. This approach is expected to work, as Quercetin has been reported to lead to prevention of SCC in mice (non-FA) and decreased tumor growth in FA head and neck cancer cell lines. In a previous clinical study by Dr. Mehta, evidence of decreased DNA damage in oral mucosa brushings was observed in patients with FA (pre-HCT) after treatment with Quercetin for one month. Based on these strong and promising data, this second study will test Quercetin treatment for 6 months (with an option of up to a total of 2 years) in 20 post-transplant patients with FA. Clinical and laboratory tests will confirm the beneficial effect of Quercetin. Expected positive impact is that success will lead to a new prevention strategy for SCC in post-HCT patients with FA that will eliminate or at least delay the development of SCC.

Development of a Safe, Completely Non-Genotoxic Anti-Kit Antibody-Based Conditioning Regimen for Hematopoietic Stem Cell Transplantation in Fanconi Anemia (Agnieszka Czechowicz, MD, PHD at Stanford University)

Bone marrow transplantation has been used for 60+ years to treat >1,000,000 patients suffering from many types of blood or immune diseases, and this therapy is the best current treatment for FA patients experiencing blood problems. This procedure relies on replacing sick blood-forming stem cells in patients with healthy ones from a donor, resulting in a completely new blood system. However, this currently requires use of irradiation and/or chemotherapy which can be extremely harmful, especially to FA patients whose cells cannot fix DNA mistakes caused by these agents. This study will test wither antibodies that can target specific cells of the body provide safe turnover of the bone marrow and eliminate bone marrow disease in FA patients without genotoxic conditioning. This type of therapy could not only be used to treat blood problems in FA patients, but also prevent them all together if used upfront in recently diagnosed patients.

Pilot Study of Metformin for Patients with Fanconi Anemia (Akikro Shimamura, MD, PhD and Elissa Furtani, MD at Harvard Medical School/Dana-Farber Cancer Institute)

Low blood counts due to bone marrow failure are a common complication of Fanconi anemia (FA) that affect health and quality of life. Metformin is an FDA-approved oral medication that has been used for many decades to treat diabetes and insulin resistance. New evidence has shown that the drug improves blood counts in mice with FA and that it protects against DNA damage. Other laboratory studies suggest that Metformin may be protective against aldehydes, which are toxic to FA mice and to FA patients.

Metformin is currently being studied in other clinical trials as an anti-aging and anti-cancer drug, but it is not known how Metformin will affect people with FA. This study will test whether Metformin improves

TGF-β Pathway Inhibitors for the Treatment of Bone Marrow Failure in Fanconi Anemia (Alan D'Andrea, MD, Dana-Farber Cancer Institute)

Fanconi anemia (FA) patients suffer from progressive bone marrow failure due to the defective hematopoietic stem cells (HSCs) in their bone marrow. The mechanisms of why the HSCs in FA patients are defective remain elusive. Recent studies suggest that DNA damage induced by physiological stress or aldehydes in HSCs may contribute to the bone marrow failure in FA. Our laboratory has recently made an important discovery that hyperactive TGFbeta pathway signaling in FA bone marrow HSCs is the cause of bone marrow failure. We have recently published a paper describing this discovery (Zhang H et al, Cell Stem Cell, 2016). Specifically, we found that inhibition of the TGFbeta pathway by specific inhibitors improves the defects in HSCs from FA mice or FA patients by promoting DNA repair. Many of the inhibitors of TGFbeta pathway are currently under clinical trials. One such drug called Galunisertib is currently under clinical trial for Myelodysplastic syndrome (MDS) which is also a bone marrow disease. Our preliminary data suggest that Galunisertib significantly reduces DNA damage in HSCs from FA mice. We therefore plan to test Galunisertib and the other drugs related to Galunisertib for their ability to improve HSC defects in FA. In Specific Aim 1, we will test the drugs in in vitro tissue culture experiments using both mouse and human FA bone marrow cells. In Specific Aim 2, we will test the selected drug in preclinical FA mouse models.

Pilot Study of Metformin for Patients with Fanconi Anemia (Akiko Shimamura, MD PhD & Elissa Furutani, MD, Dana-Farber/Boston Children's Cancer and Blood Disorders Center Project

The objective of this proposal is to conduct a pilot study of metformin to treat FA. This pilot study will explore whether Metformin is safe and efficacious in improving hematopoiesis in patients with Fanconi Anemia. We will focus on pediatric and young adult patients with FA to determine safety, tolerability, and preliminary efficacy of Metformin in Fanconi Anemia patients. This study will also integrate biological correlative studies to glean mechanistic insights which may inform future studies. The anticipate enrolling 20 patients, with a study duration of 2 years.

Field-Coverage Oral Cancer Chemoprevention via Janus Nanoparticles Susan R. Mallery, DDS, PhD & Joerg Lahann, PhD, Ohio State University

By virtue of their inability to repair a specific form of DNA damage, persons with Fanconi Anemia (FA) are appreciably more susceptible to development of certain cancers. Oral squamous cell carcinoma (OSCC), which arises from the lining cells of the mouth, occurs at >500 fold higher incidence in individuals with FA. Standard OSCC risk factors i.e. tobacco and alcohol initiate precancerous lesions and OSCC in specific carcinogen pooling areas like the floor of the mouth and the side of the tongue. For persons with FA, however, virtually every site in their mouths is at high risk for OSCC development. To address this need, FA OSCC prevention needs to provide field coverage throughout the entire mouth. A small particle capable of delivering two cancer preventing agents simultaneously (Janus nanoparticle) is well-suited for delivery of chemopreventives throughout the entire oral cavity. The purpose of this study is to optimize the nanoparticles size, shape and charge to enhance uptake by the target surface oral cavity

lining cells. Health Care Benefits for Persons with FA: For persons with Fanconi Anemia, development of oral cancer is a significant risk. A strategy to provide cancer preventing agents throughout the entire mouth without side effects has the potential to improve both quality and duration of life for persons with FA.

Targeting Lipid Metabolism in FA for the Prevention and Treatment of Squamous Cell Carcinoma
Dry and moist skin (e.g., in the mouth) plays a critical role in maintaining a barrier against environmental insults and cancer. Based on new data, this team believes that individuals with FA may have an impaired barrier and this may be why they are at risk for SCC. Specifically, data shows that FA pathway loss impairs adhesion between skin cells. FA pathway loss also stimulates motility in cells that would otherwise be stationary; this is particularly interesting, given that cancer cells have reduced attachments to their environment and tend to be highly motile. Moreover, the link between the FA pathway and changes in the skin may be due to altered lipid levels and Rac1 signaling. Fortunately, it is possible to restore lipid levels and signaling to normal with existing drugs and in this proposal it will be determined if this can normalize skin architecture to prevent and treat FA SCC.

Targeting LNK(SH2B3) to Ameliorate Hematopoietic Stem/Progenitor Defects in Fanconi Anemia.
Dr. Wei Tong, Children's Hospital Philadelphia recently discovered a novel gene called "LNK" that regulates bone marrow cell survival and growth. She found that loss of LNK in a mouse model of FA restores normal bone marrow functions and increases stem cell longevity. This new grant will allow Dr. Tong to explore the mechanisms underlying this important discovery and to develop new strategies to inhibit LNK to improve bone marrow function in Fanconi anemia. This work will also advance efforts to correct Fanconi mutations by gene therapy.

A Porcine Model of Fanconi Anemia (William Fleming, MD, PhD; Markus Grompe, MD Oregon Health & Science University

A major limitation in FA research is the absence of an animal model that faithfully recapitulates the clinical features of this disease in humans. While mice have the characteristic DNA repair defects, they do not spontaneously develop the progressive anemia or acute leukemia seen in many patients. It is thought that the short lifespan of mice provides insufficient time for the development of the progressive bone marrow failure observed in patients. This proposal is potentially very significant as it will create the first large animal model of FA. Based on the remarkable success of the pig model of cystic fibrosis, it is anticipated that our proposed FANCA deficient pig will serve as an excellent model for studying FA disease progression and even more importantly, as a pre-clinical platform to test novel therapies. **The research here will develop an animal model of cancers that occur due to mutations in FA genes, including five different breast cancer genes. The animal model can then be used to test the ability of different drugs to prevent tumor formation.**

Bridge Funding for Metformin Studies (Markus Grompe, MD, Oregon Health & Science University

The key goal of the project in this second phase is to prioritize a single drug regimen from a small list of candidates previously identified and to generate the preclinical data needed for starting a clinical trial soon. This Program Project will use a multidisciplinary approach to achieve this goal. The clinical disciplines represented include pediatrics, hematology, oncology and medical genetics. The scientific areas of expertise include molecular hematology, xenotransplantation, DNA repair, cell biology and mouse genetics. Project 1 (Grompe Lab at OHSU) will explore drugs that have already shown promise in FA mouse models such as metformin, p38 MAPK inhibitors, antioxidants and androgens, both singly and in combination. Project 2 (D'Andrea Lab at Harvard) will focus on inhibitors of the TGF- pathway for the treatment of FA. This is a new class of medications being tested by multiple pharmaceutical companies to treat inflammatory diseases. We have found that these medicines can significantly improve blood formation in FA. Project 3 (Shimamura Lab at Harvard) will use primary human cells from FA patients (bone marrow specimens) to study the compounds from projects 1 and 2. In addition, the regulatory groundwork for a clinical trial will be done. These scientific projects will be supported by technical cores which can provide expertise in DNA repair, human patient specimen procurement and testing human bone marrow cells in living mice. **Drugs may be developed from this work that help improve blood counts in individuals with different bone marrow failure disorders, including leukemia and aplastic anemia.**

A Multicenter Trial of Hematopoietic Stem cell Transplantation for the Treatment of Patients with Fanconi Anemia Lacking a Genotypically Identical Donor, Using a Chemotherapy Only Cytoreduction with Busulfan, Cyclophosphamide and Fludarabine. (Memorial Sloan-Kettering Cancer Center)

What has been given for the treatment of FA in the past is to use a combination of low doses of radiation to the whole body and low doses of the chemotherapy drugs cyclophosphamide and fludarabine before the transplant. However, the use of radiation can, later on, increase the chances of getting a second cancer of the skin, head or the neck. These chances of a second cancer are higher than normal and more devastating in patients with FA. **The purpose of this study is to find out if the doctors can use a chemotherapy drug called busulfan instead of the radiation. The goal of this study is to eliminate the short term and long term risks of the radiation.**

Prevalence of FA Gene Germline Mutation in Young Adults with Head and Neck Cancer (MD Anderson Cancer Center)

A study at MD Anderson to see if young adults (under the age of 49) previously diagnosed with head & neck cancer but never diagnosed with FA might possibly also be FA patients. **This research will help us know whether all young head and neck squamous cell cancer patients should be routinely tested for FA. If FA is diagnosed, it will help inform physicians how these patients should be treated (aggressive radiation/chemotherapy would not be appropriate).**

Potential Therapeutic Use of Resveratrol for Head and Neck Carcinogenesis of Fanconi Anemia (Robert Sclafani, PhD)

A study conducted at the University of Colorado School of Medicine by Robert Sclafani, PhD; DNA repair processes may be weak or defective in many types of cancer (for all patients, not just those with

FA). Resveratrol, a natural plant compound, has been shown to inhibit the growth of some cancer cells in vitro as well as tumor growth in animal models. When treated with this compound, head and neck squamous cancer cells and ovarian cancer cells (but not normal primary cells) died. Dr. Sclafani believes that since cancer cells lack normal DNA repair pathways, they are much more sensitive to resveratrol actions than normal cells. **The proposal seeks to extend these findings to the setting of FA and to test whether resveratrol could be a safe, non-toxic, and inexpensive treatment to prevent or treat head and neck squamous cell carcinoma.**

Fanconi Anemia and the Repair of DNA Protein Cross-Links (Harvard Medical School)

A study being conducted at Harvard Medical School by Johannes Walter, PhD. Recent studies from a laboratory in Cambridge, the United Kingdom, suggest that the inability to detoxify aldehydes may be a key problem in Fanconi anemia. Aldehydes are common carcinogens. They are constantly produced in the body as a result of cellular metabolism. Aldehydes also exist in the environment: formaldehyde and alcohol are common examples. Mice bred to have Fanconi anemia, coupled with an inability to detoxify aldehydes, developed aplastic anemia, leukemia, and developmental defects, similar to the problems experienced by FA patients. A study of FA patients in Japan showed that patients with an inability to detoxify aldehydes had a more severe form of this disease than patients without this additional defect. Dr. Johannes Walter at the Harvard Medical School is now examining how the FA pathway, when functioning normally, promotes the repair of aldehyde induced DNA damage. He hopes that understanding the function of the FA pathway in the repair process will suggest how this repair could be stimulated even in the absence of a normal FA pathway. **FARF scientists believe that this work will lay the foundation for the development of rational therapeutic approaches.**

Novel Therapeutic Agents for the Treatment of Bone Marrow Failure in Fanconi Anemia

A study being conducted by Dr. Alan D'Andrea at Harvard University and the Dana-Farber Cancer Institute. **The goal of this research is to find compounds that will help prevent bone marrow failure in individuals with FA and negate or delay the need for a bone marrow transplant.** It has been established that one component of FA bone marrow failure is an increased incidence of and hypersensitivity to DNA damage. Therefore, Dr. D'Andrea screened approximately 6000 small molecules or drugs to find ones that protected FA skin cells in the laboratory from dying after DNA damage. He has narrowed the list of candidate drugs down to 10 and will now test to see if these small molecules or drugs protect FA blood and bone marrow cells from DNA damage.

Correcting Fanconi Anemia Mutations by CRISPR/Cas9 Genome Editing to Explore a Novel Therapeutic Strategy

A recent breakthrough in the technology called "gene editing" has provided the ability to efficiently mend mutations in Fanconi anemia genes in patient's own cells. This technology, called "CRISP/Cas9", enables the prevention of bone marrow failure in FA patients by autologous bone marrow transplantation with FA-corrected cells. It is believed that CRISP/Cas9 gene editing has great potential to provide clinical solutions, in particular for FA patients without matching donors. The research in this proposal will answer key questions about gene editing in FA-deficient cells as a first essential step towards the clinical translation of this technology to treat FA patients. The technology generated from

this project could be used to correct mutated genes from a variety of different diseases, including cancer.

Screening for Therapeutics in Models of Fanconi Anemia
A multi-institution (Harvard, University of Pennsylvania, Dana-Farber Cancer Institute and the University of Oregon) project involving the testing of small molecules on cancer prone mice **to discover drugs that can delay or prevent cancers in patients with Fanconi anemia — and in all of us. (Note: the National Institutes of Health in 2011 recognized the far reaching value of this initial FA research and gave the group a 10.7 million dollar grant to continue their work).**

"Gene Editing of Induced Pluripotent Stem Cells from Patients with FA"; also titled "Correction of Human
FA-Induced Pluripotent Cells by Recombination" (Dr. Jakub Tolar M.D. Ph.D.)
A promising effort to identify an effective method of gene therapy which would safely correct the blood stem cells of FA patients. **The application to other diseases is unlimited.**

Comparative Genetic and Metastatic Potential Analyses of Head and Neck Squamous Cell Carcinomas from
Wild-type and FANCC-Deficient Mice (Dr. Laura Hays, Oregon Health and Science University)
A study of squamous cell carcinoma in normal and FA mice to determine whether the tumors are biologically and/or genetically different. Dr. Hays' research uses gene arrays to classify and quantify chromosomal rearrangements in tumor cells to determine whether the mutations found in the FA tumors are genetically more unstable and have a higher malignant potential. Additionally, she will quantify the metastatic potential by injecting the oral cavity tumors derived from the normal and FA mice into the oral cavity of immune-competent mice and measure pulmonary metastases. **This work will increase the understanding of carcinogenesis in head and neck cancer in FA patients.**