The design of GUMBOS and nanoGUMBOS

Versatile compounds with a wide range of applications

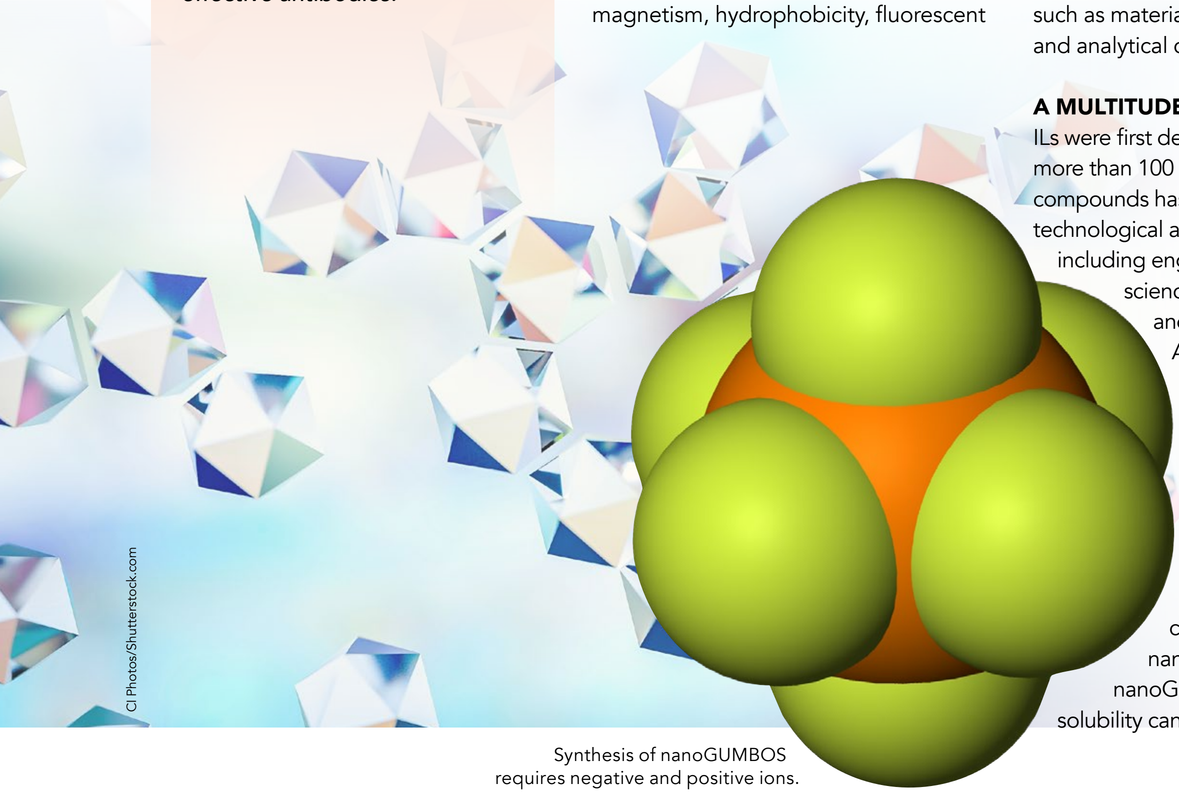
GUMBOS (Group of Uniform Materials Based on Organic Salts) are solid-phase materials with relatively low melting points of up to 250°C. Their structural design can be easily altered to obtain a final product with 'tunable' properties. Professor Isiah Warner and his team from Louisiana State University have shown that GUMBOS can be converted to nanoparticles that have a wide range of applications, including the selective treatment of cancer, the development of solar and fuel cells, the extraction of biomaterials from complex biological mixtures, and the formulation of more effective antibodies.

Unlike most ionic compounds, known for their high melting points, ionic liquids (ILs) are characterised by relatively low melting points. ILs are defined as organic salts with melting points below 100°C. The 'liquid' part of this definition is quite arbitrary, however, as organic salts with melting points between 25°C and 100°C are actually solids and sometimes referred to as 'frozen' ILs. Discovered around 100 years ago, the chemistry of these compounds has progressed to such an extent that scientists can tweak the melting point or other physical properties of the salt in question by making simple alterations to one of its counterions. Solid-phase properties which can be manipulated via this chemistry include, but are not limited to, magnetism, hydrophobicity, fluorescent properties, and cytotoxicity (the ability to kill specific cells).

Professor Isiah Warner and his colleagues from Louisiana State University have been developing solid-phase materials from organic salts whose properties can be finely tuned to achieve different functions. To emphasise the differences between these materials and ILs, they refer to them as GUMBOS: a Group of Uniform Materials Based on Organic Salts. GUMBOS are closely related to ILs but have a melting point range of 25–250°C. Warner and his team have embarked on an ongoing project to demonstrate how ionic liquid chemistry can provide unique chemistry for solid-phase applications and how GUMBOS can benefit diverse disciplines such as material science, biosciences, and analytical chemistry.

A MULTITUDE OF PURPOSES

ILs were first described in 1914. For more than 100 years, the use of these compounds has been fundamental for technological advances in many fields, including engineering, biological sciences, analytical sciences, and solvent chemistry. Although GUMBOS share properties similar to those of ILs, these solid-phase compounds would be more valuable in the development of novel biomedical applications. As an example, GUMBOS can be engineered into nanoparticles, referred to as nanoGUMBOS, whose water solubility can be altered to circumvent the toxicity problems associated with water-soluble nanomaterials.



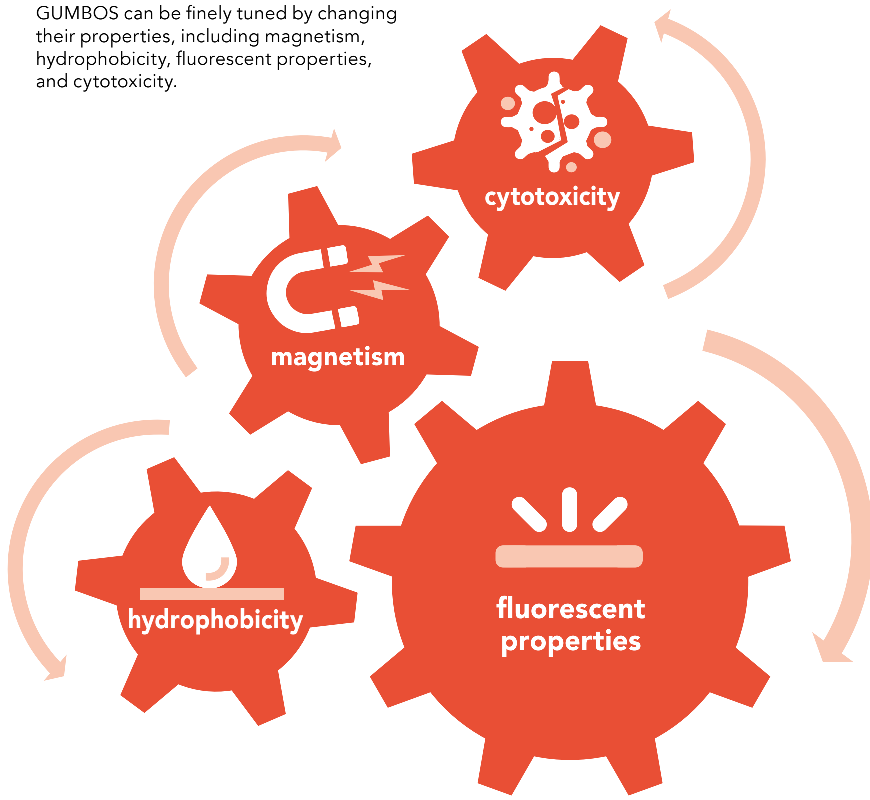
Synthesis of nanoGUMBOS requires negative and positive ions.

CI Photos/Shutterstock.com

In 2014, Warner and his collaborators published a study to demonstrate how GUMBOS and nanoGUMBOS can be designed and synthesised. The researchers showed that if the desired molecule is already a salt, then the charged ionic species of the salt deemed as essential is maintained in the final structure, while the non-essential part of the salt, known as the counter-ion, is substituted by a simple ion exchange. If the molecule is not a salt, then a charge must be added to the desired product. For example, if a positive ion is desired, then the most direct synthetic route may involve the addition of an amine group into the compound. The typical positive ions used in the synthesis of nanoGUMBOS include imidazolium, ammonium, phosphonium, and pyridinium. Hexafluorophosphate, tetrafluoroborate, or tetraphenylborate along with other bulky, negatively charged compounds are typically used as the negative ions.

Warner's team uses three primary methods for preparation of nanoGUMBOS. The simplest method involves precipitation of the GUMBOS into nanosized clusters after mixing dilute drops of the GUMBOS solution with a miscible solvent. Particles produced using this method are generally spherical. The melt–emulsion–quench method requires melting the GUMBOS, followed by emulsification, a process involving the forced dispersion of small water repellent droplets in a water-soluble medium through continuous stirring. The emulsification process is then followed by refreezing. The advantage of this approach is the possibility of additive-free preparations, making it suitable for high-purity uses such as in medical applications. A third method involves the use of porous templates such as aluminum oxide membranes with cylindrical pores, which enable the production of one-dimensional nanoGUMBOS such as rods, wires, and tubes.

GUMBOS can be finely tuned by changing their properties, including magnetism, hydrophobicity, fluorescent properties, and cytotoxicity.

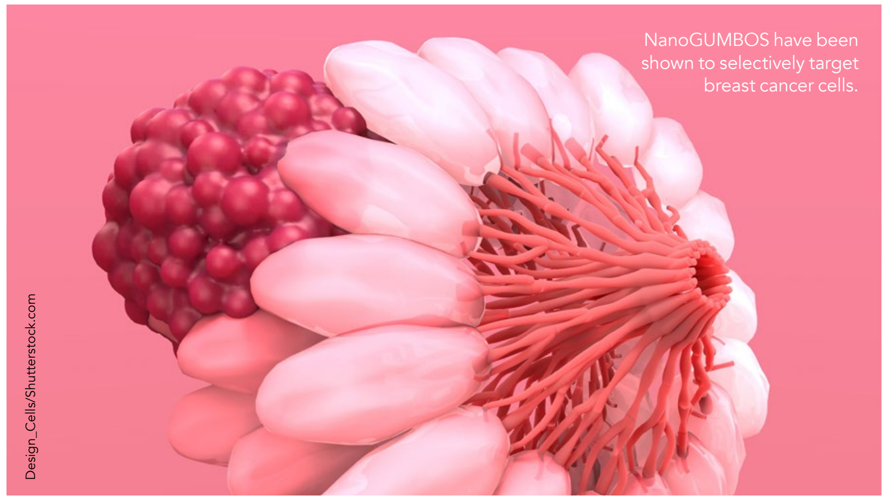


R6G-coupled nanoGUMBOS are an effective cancer treatment that targets breast cancer cells, leaving other cells unharmed.

GUMBOS can be engineered to display a wide range of solubilities and melting points. The solubility in water is largely dependent on the anion employed. Anions such as tetrafluoroborate show enhanced aqueous solubility resulting from hydrogen bonding with the surrounding water molecules. Decades of advances in IL chemistry have allowed researchers to convert hydrophilic compounds to more hydrophobic ones and vice versa by simply exchanging counter-ions with different degrees of hydrophobicity.

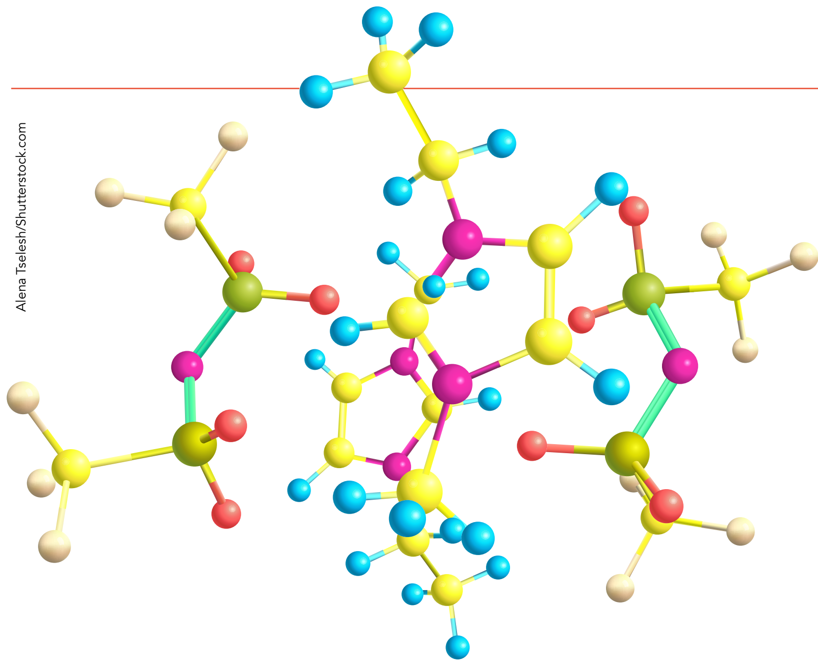
THE ANTI-CANCER POTENTIAL OF NANOGUMBOS

In contrast to conventional chemotherapy, anti-cancer treatments based on nanoscale delivery systems provide protection of the active drug from biodegradation due to metabolism and offer enhanced cellular uptake. Warner's team has developed selective chemotherapeutic agents based on



NanoGUMBOS have been shown to selectively target breast cancer cells.

Design_Cells/Shutterstock.com



Alena Tselesh/Shutterstock.com

Molecular structure of ionic liquid.

rhodamine 6G (R6G), a highly fluorescent, positively charged compound characterised by an intense red colour. By using the unique and adaptable chemistry of GUMBOS, the group started to work on R6G after investigating the correlation between hydrophobicity, charge, and chemotherapeutic toxicity of different molecules.

The group demonstrated that rhodamine 6G (R6G)-nanoGUMBOS proved to be an effective cancer treatment that is selectively toxic towards breast cancer cells, leaving normal cells unharmed. The cytotoxicity of R6G-nanoGUMBOS towards the breast cancer cell lines was dose dependent.

The study, published in 2018, showed that internalisation of the nanoGUMBOS in cancer cells resulted in dissociation of the nanomaterials within the acidic environment of the endosomal transport vesicles. This led to activation of the chemotherapeutic activity of the compound. In contrast, the R6G nanoGUMBOS are internalised in normal cells via a different mechanism that bypasses the endosomal vesicles, resulting in non-toxic behaviour in normal cells. Investigation of the therapeutic efficacy of these nanomaterials in mice demonstrated the retention of the same therapeutic properties in vivo.

MAGNETIC NANOGUMBOS FOR THE ISOLATION OF HAEMOGLOBIN

Proteins play an essential role in all living organisms, serving as structural building blocks, regulating cellular activities, mediating the transport of gases and nutrients, as well as acting as messengers for cell-to-cell communication. One of the most studied proteins is haemoglobin, which is fundamental for the transport of oxygen to all tissues of the body. Researchers are looking at ways of isolating haemoglobin to produce blood substitutes for transfusions in emergency situations.

Warner and his team have taken advantage of their extensive knowledge of ionic liquid chemistry to develop novel ways to extract haemoglobin. The team published a study in 2019, describing the development of a haemoglobin extraction biomaterial with magnetic properties, based on nanoGUMBOS. The positively charged imidazolium ion was chosen as the main cation for the organic salt for its selective affinity towards haemoglobin. The element dysprosium was also included in the structure of the biomaterial for its paramagnetic properties. Coordination of imidazolium with the iron in haemoglobin is believed to be the major driving force for selective adsorption of the protein. The novel nanoGUMBOS solid-phase extraction procedure was successfully applied to selectively isolate haemoglobin from human whole blood and proves to be a promising technique for production of synthetic blood substitutes.

NanoGUMBOS can selectively isolate haemoglobin from human whole blood and proves to be a promising technique for the production of synthetic blood substitutes.

A NEW STRATEGY TO TACKLE DRUG-RESISTANT BACTERIA

Drug-resistant bacteria represent an area of increasing concern in many clinical settings. The lack in production of new antibiotics adds to the ever-increasing incidence of infectious diseases caused by multi-drug-resistant bacteria. In a study published in 2015, Warner and his collaborators proposed novel formulations of existing antibiotics and antibacterial agents in combination with GUMBOS technology, to obtain tailor-made hydrophobic ion pairs with low toxicity and enhanced pharmacological properties. They reported that these novel formulations extend the spectra of antibacterial compounds while decreasing the concentration required to inhibit the growth of multi-drug-resistant bacteria. Future studies in animals will be needed to evaluate drug performance in vivo against resistant strains of bacteria.

WIDE-REACHING APPLICATIONS

Warner and his colleagues from Louisiana State University have developed organic salts, known as GUMBOS, that possess unique physical and chemical properties, including tunable solubility, melting point, viscosity, thermal stability, and functionality. These compounds allow the production of solid-phase materials that have a wide range of applications which include, but are not limited to, the development of nanomaterials for the selective treatment of cancer, the magnetic extraction of biomaterials from complex biological mixtures, and the formulation of more effective antibodies. GUMBOS can also be used for development of fuel cells, smart batteries, and solar cells. The Warner laboratory has accrued several patents, with some pending, for their contributions to analytical chemistry, materials chemistry, and the biomedical sciences.

Behind the Research

Professor Isiah Warner

E: iwarner@lsu.edu **T:** +1 225 578 2829 **W:** faculty.lsu.edu/iwarner
W: www.researchgate.net/profile/Isiah-Warner
W: scholar.google.ca/citations?user=bA_UIQgAAAAJ&hl=en

Research Objectives

Professor Warner and his team have developed three primary strategies for synthesising nanoGUMBOS – nanomaterials with tuneable properties that can be designed and assembled for specific uses.

Detail

Address
Louisiana State University
Department of Chemistry
434 Choppin Hall, Baton Rouge
LA 70803, USA

Bio

Dr Isiah Warner is West Professor of Chemistry and a Boyd Professor of the LSU system. He has expertise in fluorescence spectroscopy and materials chemistry. He also does research that focuses on mechanisms for enhancing student education in STEM, focusing on encouraging under-represented students to pursue terminal degrees.

Funding

The researchers gratefully acknowledge financial support through NASA cooperative agreement NNX 16AQ93A under contract number NASA/LEQSF (2016-19)-Phase 3-10, and the National Science Foundation under Grant Nos. CHE-1905105 and HRD-1736136. Any opinions, findings, and conclusions or recommendations expressed in this material are those of the author(s) and do not necessarily reflect the views of the National Science Foundation.

References

Cong, M, Pérez, RL, Chen, M, et al, (2019) Imidazolium-dysprosium-based magnetic NanoGUMBOS for isolation of hemoglobin. *Talanta*, 205, 120078. doi.org/10.1016/j.talanta.2019.06.078

Bhattarai, N, Mathis, JM, Chen, M, et al, (2018) Endocytic Selective Toxicity of Rhodamine 6G nanoGUMBOS in Breast Cancer Cells. *Molecular Pharmaceutics*, 15(9), 3837–3845. doi.org/10.1021/acs.molpharmaceut.8b00339

Cole, MR, Hobden, JA, Warner, IM, (2015) Recycling antibiotics into GUMBOS: a new combination strategy to combat multi-drug-resistant bacteria. *Molecules*, 20(4), 6466–6487. dx.doi.org/10.3390%2Fmolecules20046466

Warner, IM, El-Zahab, B, Siraj, N, (2014) Perspectives on moving ionic liquid chemistry into the solid phase. *Analytical Chemistry*, 86(15), 7184–7191. doi.org/10.1021/ac501529m

Das, S, Bwambok, D, El-Zahab, B, et al, (2010) Nontemplated Approach to Tuning the Spectral Properties of Cyanine-Based Fluorescent NanoGUMBOS. *Langmuir*, 26(15), 12867–12876. doi.org/10.1021/la101463r

LSU

Personal Response

Are there any other proteins or compounds, aside from haemoglobin, that are worth extracting with nanoGUMBOS?

" It is well known that many metal ions play key roles in numerous protein functions. For example, divalent metal cations such as Fe, Zn, and Mn have been identified as key players in many proteins. Regarding Zn, its role in the abundantly important zinc metalloproteins has been well characterised. Thus, without extensive thought, one can rationalise that it should be possible to develop schemes where many proteins can be extracted using a GUMBOS strategy. Some of these schemes may also involve magnetic extractions. "

What are the next steps in your research?

" As a result of the extensive potential applications of GUMBOS, our research has been very broadly focused. Currently, we are evaluating these broad areas to see where our research is more likely to have the greatest impact. After these evaluations, we desire to focus on areas more narrowly where we will have impact. "