OMB Number: 4040-0004 Expiration Date: 12/31/2022

Application for	Federal Assista	nce SF-424				
* 1. Type of Submiss Preapplication	ion:	* 2. Type of Ap	pplication:	* If Revision, select appropriate letter(s):		
Application		Continua	tion '	* Other (Specify):		
—	ected Application	Revision	Г	Cinci (Green,).		
	ected Application					
* 3. Date Received: Completed by Grants.gov	v upon submission.	4. Applicant Id	entifier:			
	·					
5a. Federal Entity Ide	entifier:			5b. Federal Award Identifier:		
State Use Only:						
6. Date Received by	State:	7. Sta	ate Application	Identifier: FL		
8. APPLICANT INFO	ORMATION:	•				
* a. Legal Name: T	he Florida Int	ernational	University	Board of Trustees		
* b. Employer/Taxpay	yer Identification Nur	mber (EIN/TIN):		* c. Organizational DUNS:		
65-0177616				0712988140000		
d. Address:						
* Street1:	11200 SW 8 St					
Street2:	MARC 430					
* City:	Miami					
County/Parish:	FL					
* State:	FL: Florida					
Province:						
* Country:	USA: UNITED S	TATES				
* Zip / Postal Code:	33199-0001					
e. Organizational U	Jnit:					
Department Name:				Division Name:		
Office of Res.	& Econ. Dev.					
f. Name and contac	ct information of p	erson to be co	ntacted on ma	atters involving this application:		
Prefix:			* First Name	9: Roberto		
Middle Name: M						
* Last Name: Gut	ierrez					
Suffix:						
Title: Assistant	VP for Research	ch				
Organizational Affilia	tion:					
* Telephone Number: 305-348-2494 Fax Number: 305-348-4117						
*Email: gutierrr				[111 175 175 175 1		
Linaii gullerri	_eriu.euu					

Application for Federal Assistance SF-424
* 9. Type of Applicant 1: Select Applicant Type:
H: Public/State Controlled Institution of Higher Education
Type of Applicant 2: Select Applicant Type:
Type of Applicant 3: Select Applicant Type:
* Other (specify):
* 10. Name of Federal Agency:
Environmental Protection Agency
11. Catalog of Federal Domestic Assistance Number:
66.484
CFDA Title:
South Florida Geographic Initiatives Program
* 12. Funding Opportunity Number:
EPA-R4-SFL-2021-01
* Title:
2021 SOUTH FLORIDA GEOGRAPHIC PROGRAM
13. Competition Identification Number:
Title:
14. Areas Affected by Project (Cities, Counties, States, etc.):
Add Attachment Delete Attachment View Attachment
* 15. Descriptive Title of Applicant's Project:
An examination of the risk of pharmaceuticals in South Florida: extent and pathways of exposure in a valuable recreational fishery, bonefish
a varuable recreacional rishery, ponerish
Attach supporting documents as specified in agency instructions.
Add Attachments Delete Attachments View Attachments

Application for Fe	deral Assistance SF-424				
16. Congressional Dis	stricts Of:				
* a. Applicant	026		* b. Program/P	Project FL-026	
Attach an additional list	of Program/Project Congressional Dis	stricts if needed.			
		Add Attachment	Delete Attach	View Attachment	
17. Proposed Project	:				
* a. Start Date: 01/03	1/2022		* b. End	d Date: 12/31/2024	
18. Estimated Fundin	g (\$):				
* a. Federal	292,530.	00			
* b. Applicant	0.	00			
* c. State	0.	00			
* d. Local	0.	00			
* e. Other	0.	00			
* f. Program Income	0.	00			
* g. TOTAL	292,530.	00			
* 19. Is Application Su	ubject to Review By State Under E	xecutive Order 12372 P	rocess?		
a. This application	າ was made available to the State ເ	under the Executive Orde	er 12372 Process f	for review on	
b. Program is sub	ject to E.O. 12372 but has not bee	n selected by the State for	or review.		
C. Program is not	covered by E.O. 12372.				
* 20. Is the Applicant	Delinquent On Any Federal Debt?	(If "Yes," provide expla	nation in attachn	nent.)	
Yes	No				
If "Yes", provide expla	anation and attach				
		Add Attachment	Delete Attach	nment View Attachment	
21. *By signing this application, I certify (1) to the statements contained in the list of certifications** and (2) that the statements herein are true, complete and accurate to the best of my knowledge. I also provide the required assurances** and agree to comply with any resulting terms if I accept an award. I am aware that any false, fictitious, or fraudulent statements or claims may subject me to criminal, civil, or administrative penalties. (U.S. Code, Title 218, Section 1001) ** I AGREE ** The list of certifications and assurances, or an internet site where you may obtain this list, is contained in the announcement or agency specific instructions.					
Authorized Represen	tative:				
Prefix:	*	First Name: Roberto			
Middle Name: M					
* Last Name: Gutie:	rrez				
Suffix:					
* Title: Assista	nt VP for Research				
* Telephone Number:	305-348-2494	F	ax Number: 305-	348-4117	
* Email: gutierrr@f	iu.edu				
* Signature of Authorize	d Representative: Completed by Gra	nts.gov upon submission.	* Date Signed:	Completed by Grants.gov upon submission.	

OMB Number: 2030-0020 Expiration Date: 04/30/2021

Preaward Compliance Review Report for All Applicants and Recipients Requesting EPA Financial Assistance

Note: Read Instructions before completing form.

I. A.	Applican	/Recipient (Name, Address, City, State, Zip Code)			
	Name:	The Florida International University Board of Trustees			
	Address:	11200 SW 8 St			
	City:	Miami			
	State:	FL: Florida Zip Code: 33199			
D	DUNS N	071298814			
II.	·	plicant currently receiving EPA Assistance? Yes No			
III.		vil rights lawsuits and administrative complaints pending against the applicant/recipient that all or, national origin, sex, age, or disability. (Do not include employment complaints not covered b			
		Nehme vs. Florida International University and McLaughlin vs. Florida Interst $\#04-21-2182$, OCR Complaint $\#04-20-2528$, OCR Complaint $\#04-19-2285$, and OCF			
IV.	discrimi	ivil rights lawsuits and administrative complaints decided against the applicant/recipient within that ion based on race, color, national origin, sex, age, or disability and enclose a copy of all decise a coty of all decise actions taken. (Do not include employment complaints not covered by 40 C.F.R. Parts 5 and 7	ions. Plea		
N/A		the state of the s	<i>,</i>		
٧.	of the re	ivil rights compliance reviews of the applicant/recipient conducted by any agency within the last view and any decisions, orders, or agreements based on the review. Please describe any correct . § 7.80(c)(3))			ose a copy
N/A					
VI.	Is the ap	olicant requesting EPA assistance for new construction? If no, proceed to VII; if yes, answer (a)	and/or (b)	below.	
	16.41	Yes No			
a.		nt is for new construction, will all new facilities or alterations to existing facilities be designed ar e to and usable by persons with disabilities? If yes, proceed to VII; if no, proceed to VI(b).	id constru	cted to be	readily
		Yes No			
b	-	nt is for new construction and the new facilities or alterations to existing facilities will not be reans with disabilities, explain how a regulatory exception (40 C.F.R. 7.70) applies.	dily acces	sible to a	nd usable
VII.		applicant/recipient provide initial and continuing notice that it does not discriminate on the basi olor, national origin, sex, age, or disability in its program or activities? (40 C.F.R 5.140 and 7.95)		Yes	☐ No
а	. Do the m	ethods of notice accommodate those with impaired vision or hearing?	\boxtimes	Yes	☐ No
b		tice posted in a prominent place in the applicant's offices or facilities or, for education programs ities, in appropriate periodicals and other written communications?		Yes	☐ No
С	. Does the	notice identify a designated civil rights coordinator?	\boxtimes	Yes	☐ No
VIII.		applicant/recipient maintain demographic data on the race, color, national origin, sex, age, or of the population it serves? (40 C.F.R. 7.85(a))	\boxtimes	Yes	☐ No
IX.		applicant/recipient have a policy/procedure for providing access to services for persons with	\boxtimes	Yes	☐ No

compliance with 40 C.F.R. Parts 5 and 7? Property of the designated coordinator.	rovide the name, title, position, mailing address	, e-mail address, fax number, and telephone
Elizabeth Canning, Interim Director & T: (OCRCA) 11200 SW 8 St, PC 220, Miami, FL 33199, Phone: 305-348-2785; Email: ocrca@fiu.ec		Rights Compliance & Accessibility
	activity, or has 15 or more employees, has it add at allege a violation of 40 C.F.R. Parts 5 and 7?	
https://regulations.fiu.edu/regulation=l	FIU-106	
	For the Applicant/Recipient	
I certify that the statements I have made on this forr knowingly false or misleading statement may be purwith all applicable civil rights statutes and EPA regular. A. Signature of Authorized Official	nishable by fine or imprisonment or both under app	
Completed by Grants.gov upon submission.	Assistant VP for Research	Completed by Grants.gov upon submission.
Fo	or the U.S. Environmental Protection Agency	
I have reviewed the information provided by the approximation required by 40 C.F.R. Parts provisions of 40 C.F.R. Parts 5 and 7; and that the a EPA regulations.	5 and 7; that based on the information submitted,	this application satisfies the preaward
A. *Signature of Authorized EPA Official	B. Title of Authorized Official	C. Date

If the applicant is an education program or activity, or has 15 or more employees, has it designated an employee to coordinate its

X.

* See Instructions

Instructions for EPA FORM 4700-4 (Rev. 06/2014)

General. Recipients of Federal financial assistance from the U.S. Environmental Protection Agency must comply with the following statutes and regulations.

Title VI of the Civil Rights Acts of 1964 provides that no person in the United States shall, on the grounds of race, color, or national origin, be excluded from participation in, be denied the benefits of, or be subjected to discrimination under any program or activity receiving Federal financial assistance. The Act goes on to explain that the statute shall not be construed to authorize action with respect to any employment practice of any employer, employment agency, or labor organization (except where the primary objective of the Federal financial assistance is to provide employment). Section 13 of the 1972 Amendments to the Federal Water Pollution Control Act provides that no person in the United States shall on the ground of sex, be excluded from participation in, be denied the benefits of, or be subjected to discrimination under the Federal Water Pollution Control Act, as amended. Employment discrimination on the basis of sex is prohibited in all such programs or activities. Section 504 of the Rehabilitation Act of 1973 provides that no otherwise qualified individual with a disability in the United States shall solely by reason of disability be excluded from participation in, be denied the benefits of, or be subjected to discrimination under any program or activity receiving Federal financial assistance. Employment discrimination on the basis of disability is prohibited in all such programs or activities. The Age Discrimination Act of 1975 provides that no person on the basis of age shall be excluded from participation under any program or activity receiving Federal financial assistance. Employment discrimination is not covered. Age discrimination in employment is prohibited by the Age Discrimination in Employment Act administered by the Equal Employment Opportunity Commission. Title IX of the Education Amendments of 1972 provides that no person in the United States on the basis of sex shall be excluded from participation in, be denied the benefits of, or be subjected to discrimination under any education program or activity receiving Federal financial assistance. Employment discrimination on the basis of sex is prohibited in all such education programs or activities. Note: an education program or activity is not limited to only those conducted by a formal institution. 40 C.F.R. Part 5 implements Title IX of the Education Amendments of 1972. 40 C.F.R. Part 7 implements Title VI of the Civil Rights Act of 1964, Section 13 of the 1972 Amendments to the Federal Water Pollution Control Act, and Section 504 of The Rehabilitation Act of 1973. The Executive Order 13166 (E.O. 13166) entitled; "Improving Access to Services for Persons with Limited English Proficiency" requires Federal agencies work to ensure that recipients of Federal financial assistance provide meaningful access to their LEP applicants and beneficiaries.

Items "Applicant" means any entity that files an application or unsolicited proposal or otherwise requests EPA assistance. 40 C.F.R. §§ 5.105, 7.25. "Recipient" means any entity, other than applicant, which will actually receive EPA assistance. 40 C.F.R. §§ 5.105, 7.25. "Civil rights lawsuits and administrative complaints" means any lawsuit or administrative complaint alleging discrimination on the basis of race, color, national origin, sex, age, or disability pending or decided against the applicant and/or entity which actually benefits from the grant, but excluding employment complaints not covered by 40 C.F.R. Parts 5 and 7. For example, if a city is the named applicant but the grant will actually benefit the Department of Sewage, civil rights lawsuits involving both the city and the Department of Sewage should be listed. "Civil rights compliance review" means any review assessing the applicant's and/or recipient's compliance with laws prohibiting discrimination on the basis of race, color, national origin, sex, age, or disability. Submit this form with the original and required copies of applications, requests for extensions, requests for increase of funds, etc. Updates of information are all that are required after the initial application submission. If any item is not relevant to the project for which assistance is requested, write "NA" for "Not Applicable." In the event applicant is uncertain about how to answer any questions, EPA program officials should be contacted for clarification. * Note: Signature appears in the Approval Section of the EPA Comprehensive Administrative Review For Grants/Cooperative Agreements & Continuation/Supplemental Awards form.

Project Narrative File(s)

* Mandatory Project Narrative File File	ename: Application.pdf	
Add Mandatory Project Narrative File	Delete Mandatory Project Narrative File	View Mandatory Project Narrative File

To add more Project Narrative File attachments, please use the attachment buttons below.

APPLICATION COVER PAGE

EPA-R4-SFL-2021-01

An application submitted for projects in the Florida Keys National Marine Sanctuary or Southeast Florida:

Project Title: An examination of the risk of pharmaceuticals in South Florida: extent and pathways of exposure in a valuable recreational fishery, bonefish

Principal Investigator	` '	ge, RO Santos, WR James of Environment, Florida International University
Date Submitted: Total Federal Funding		Proposed Start Date:1/1/22
_	onstitute intended acce	vent this application is accepted whole or in part, our signatures eptance of and compliance with applicable policy, rules, and tection Agency.
ENDORSEMENTS:	:	
Submitted by:		Approved by:
Principal Investigator	r	Institutional Representative
Sm/ Su/	7	Regnier Jurado cn=Regnier Jurado, o=Florida Internation University, ou=Office of Research & Economic Development, email=juradora@fiu.edu, c=US 2021.07.26 11:41:23 -04'00'
Signature		Signature
Jennifer S Rehage_		Regnier Jurado
Typed Name		Typed Name
	sor	
Title		Title
_11200 SW 8th Stree	et, AHC5 365	11200 SW 8th Street, MARC 430
Miami, FL 33199		Miami, FL 33199
Address		Address
305.348.3804	305.348.3877	305-348-2494 305-348-4117
Phone	Fax	Phone Fax
rehagej@fiu.e	edu	juradora@fiu.edu
E-mail		E-mail
For Administrative D Name: <u>Regnier Jura</u>	*	:
Address: 11200 SW	8th Street, MARC	430, Miami, FL 33199
305-348-2494	305-348-4117	juradora@fiu.edu
Phone	Fax	E-mail

PROJECT APPLICATION SUMMARY: An examination of the risk of pharmaceuticals in South Florida: extent and pathways of exposure in a valuable recreational fishery, bonefish

In recent years, pharmaceuticals and their metabolites have been recognized as an increasingly important class of emerging contaminants, given increasing evidence of adverse sublethal effects and a lack of routine monitoring. Pharmaceutical pollution in the environment results from a lack of removal by wastewater treatment of the pharmaceuticals ingested and excreted by humans. Pharmaceuticals then enter coastal environments via wastewater effluent discharge and leakage, and can be pseudo-persistent due to the continual influx from sources such as wastewater treatment plants. Upon exposure, these chemicals elicit sublethal physiological effects at low concentrations on aquatic taxa, that cascade to population, community, and ecosystem effects. For instance, in fishes, previous work shows that pharmaceutical exposure to antidepressants can change the brain chemistry of fish, and affect their activity, feeding, boldness, sociability, migratory behavior and ultimately their survival.

In South Florida, the presence and risk of pharmaceuticals is poorly understood. In fact, at present we do not have enough information to evaluate the scale of the threat, nor the contribution of pharmaceutical contamination to water quality and ecological degradation in the region. Yet, steroid hormones, personal care products, pharmaceuticals, and wastewater indicators (i.e., caffeine and sucralose) have been detected in water wherever sampled, including at levels indicative of poor water quality. Thus, there is a need for additional comprehensive monitoring of pharmaceuticals, at broader spatiotemporal scales, at higher replication, and beyond surface waters to examine internal concentrations for multiple ecosystem compartments, including biota of socio-ecological importance. Biomonitoring of internal concentrations allows for more reliable risk assessment than relying on concentrations in water, and importantly, is the starting point for examining toxicological effects. The proposed work addresses Priority Action Item 13 Impacts of Contaminants of Emerging Concern on South Florida Aquatic Ecosystems, and contributes to the larger goal of actively improving water quality throughout South Florida coastal ecosystems by enhancing monitoring and our understanding of the risk of emergent pollutants in order to enable mitigation and management strategies.

In this study, we focus on understanding the risk of pharmaceuticals to fishes, particularly species that support economically-valuable recreational fisheries in South Florida, and that because of their life history and habitat use, are excellent indicators of pharmaceutical toxicity. Our objectives are twofold: 1) Examine the spatial extent and pattern of exposure to pharmaceuticals in bonefish in order to identify hotpots, and 2) Examine the pathways of exposure to pharmaceuticals (trophic vs. inhalational) at these hotpots. To achieve these objectives, we propose: 1) To sample bonefish throughout major coastal areas in South Florida (Biscayne Bay to Dry Tortugas), and 2) To sample prey items, water and sediment compartments at identified hotspots in order to quantify the relative importance of pathways of exposure. All samples will be analyzed for 95 commonly used pharmaceuticals. Findings of this study will inform on the extent and pathways of pharmaceutical contamination across South Florida for multiple ecosystem compartments, and on pathways of exposure. Details on hotpots of exposure will provide critical information on spatial patterns of contamination and contribute to identifying contaminant sources. Data on the relative concentrations of the 95 pharmaceuticals analyzed will allow prioritization of chemicals of concern and an understanding of potential impacts on biota. In sum, our study will be the first comprehensive analysis of the risk of pharmaceuticals to South Florida biota.

1. Introduction for Addressing the Priority Action Items

a. Situation, Need & Previous Efforts: Pharmaceuticals include human and veterinary medicinal products, such as hormones, antibiotics, analgesics and antidepressants, that constitute contaminants of emerging concern, given increasing evidence of adverse sublethal effects, and a lack of routine monitoring (Kuster and Adler 2014, Fabbri and Franzellitti 2016). Pharmaceutical pollution results from a lack of removal by wastewater treatment contaminated with the more than 4,000 pharmaceuticals presently manufactured, and the 4.6 billion yearly prescriptions (in the US alone) humans ingest and excrete. Pharmaceuticals then enter coastal environments via wastewater effluent discharge and leakage, and can be pseudo-persistent due to the continual influx from sources such as wastewater treatment plants, resulting in prolonged and spatially extensive exposure to aquatic fauna (Kustler and Adler 2014, Fabbri & Franzellitti 2016). Previous work shows that groundwater discharge can be a source of wastewater runoff, and thus of persistent and frequently-used pharmaceuticals to coastal ecosystems (McKenzie et al. 2020). Despite mounting evidence of harmful effects, no legal limits exist for the control of pollution from pharmaceuticals during manufacture, use or disposal (Fabbri & Franzellitti 2016).

Upon exposure, these chemicals can elicit sublethal physiological effects at low concentrations on aquatic taxa, including fishes (Brodin et al. 2013, Hellström et al. 2016, Richmond et al 2018, Klaminder et al. 2019), and can mix with other pharmaceuticals to disrupt ecological interactions and functions at multiple levels of ecological organization-effects, which are just beginning to be studied (Cerveny et al. 2019). For example, a survey in Sweden showed that 92% of commonly-used pharmaceuticals were not readily biodegradable, 23% had the potential for bioaccumulation, and 61% were toxic to aquatic fauna (Wennmalm and Gunnarsson 2009). In fishes, previous studies show that pharmaceutical exposure (e.g., to antidepressants) can change the

brain chemistry of fish, and affect their activity, feeding, boldness, sociability, and migratory behavior, with important consequences for survival and reproduction (Brodin et al. 2013, Hellström et al. 2016, Cerveny et al. 2019, Klaminder et al. 2019). For instance, Klaminder et al. (2019) showed that salmon smolt exposed to an anxiolytic drug took longer time to initiate migration, taking more risks and having higher activity, and thus experienced greater predation.

In South Florida, the extent of pharmaceutical contamination has only been examined in a handful of studies, and thus remains poorly understood. Yet, steroid hormones, personal care products, pharmaceuticals, and wastewater indicators (i.e., caffeine and sucralose) have been detected in water wherever sampled, including at levels indicative of poor water quality (Ng et al. 2021 and references therein). Thus, there is a need for additional comprehensive monitoring of pharmaceuticals, at broader spatiotemporal scales, at higher replication, and beyond surface waters to examine internal concentrations for multiple ecosystem compartments, including biota of socio-ecological importance. Biomonitoring of internal concentrations allows for more reliable risk assessment than relying on concentrations in water (which may miss highly-sorptive compounds), and importantly, is the starting point for examining toxicological effects (Lagesson et al. 2016, Miller et al. 2021).

In this study, we **focus on understanding the risk of pharmaceuticals to fishes,** particularly species that support economically-valuable recreational fisheries in South Florida. Specifically, we selected bonefish as an indicator species for pharmaceutical toxicity. Their benthic foraging in the sediment, their low mobility and high site fidelity, their role in the food web as a secondary consumer subject to bioconcentration, their vulnerability to bioaccumulation due to their longevity (max documented=21 yrs), their recent population declines, and their habitat use of seagrass flats both far and near the mainland and land-based pollution, make

them a reliable indicator species, representative of other aquatic organisms that rely on nearshore seagrass habitats. Bonefish are also part of a popular and valuable catch-and-release flats fishery that focuses on sight fishing in shallow seagrass habitats. A recent economic assessment estimated that approximately \$465 million of the total economic impact of saltwater angling in Florida is generated by the Florida Keys flats fishery alone (Fedler 2013). Thus, the overall aim of our study is to understand the threat of pharmaceuticals to South Florida bonefish.

b. Objectives: The proposed study has 2 objectives: 1) Examine the spatial extent and pattern of exposure to pharmaceuticals in bonefish in order to identify hotpots, and 2) Quantify the pathways of exposure to pharmaceuticals (trophic vs. inhalational) at these hotpots. To achieve these objectives, we propose: 1) To sample bonefish throughout major coastal areas in South Florida (Biscayne Bay to the Dry Tortugas), and 2) To sample prey items, water and sediment compartments at these identified hotspots in order to quantify the relative importance of pathways of exposure. The proposed work addresses the following questions: a) Is there pharmaceutical exposure in South Florida recreational fisheries? b) Where are hotspots of exposure in the region? c) What is the main pathway of exposure? d) Does the relative strength of these pathways (trophic vs water/sediment) vary spatially across hotpots? We hypothesize that pharmaceutical exposure will be common in bonefish, with hotpots of exposure found in areas with the closest proximity to the mainland and the highest anthropogenic impact (i.e., Biscayne Bay), and lowest exposure in more distant, low impact areas (i.e., Dry Tortugas and Marquesas). We hypothesize that trophic transfer will be the main pathway of exposure for bonefish, primarily via benthic invertebrate prey (Miller et al. 2021). And as in previous studies (Brodin et al. 2013, Hellström et al. 2016), we expect psychoactive pharmaceuticals to show the highest concentrations and risk across all compartments examined.

c. Applications, Benefits, and Importance: Although pharmaceuticals are increasingly recognized as a contaminant of emerging concern, information on the contamination threat posed by pharmaceuticals throughout South Florida ecosystems is severely lacking. In fact, at present we do not have enough information to evaluate the scale of the threat, nor the contribution of pharmaceutical contamination to water quality and ecological degradation in the region. The anticipated results of this study will inform on the extent and pathways of pharmaceutical contamination across all of South Florida for multiple ecosystem compartments, and on the pathways of exposure. Details on hotpots of exposure will provide critical information on the spatial patterns of contamination and contribute to the identification of contaminant sources. Data on the relative concentrations of 95 pharmaceuticals will allow prioritization of chemicals of concern and an understanding of potential impacts on biota. In sum, the proposed work addresses Priority Action Item 13 Impacts of Contaminants of Emerging Concern on South Florida Aquatic Ecosystems and contributes to the larger goal of actively improving water quality throughout South Florida coastal ecosystems by enhancing monitoring and our understanding of the risk of emergent pollutants in order to enable mitigation and management strategies.

2. Methods & Approach

d. Description of Major Tasks: In order to address our 2 objectives, we propose 2 major tasks. In TASK 1, we will sample bonefish throughout South Florida to identify hotspots of exposure. Sampling will be conducted via angling (conventional tackle and fly fishing) throughout seagrass flats, and with the assistance of knowledgeable fishing guides in order to maximize sampling efficiency. We will sample a total of 125 bonefish across 5 regions of interest: 1) Biscayne Bay, 2) Upper Keys, 3) Lower Keys, 4) West of Key West (Lakes and Marquesas) and 5) Dry Tortugas (25 fish per region, Attachment 2). At each region, angling will target 2 areas of interest: a) flats

nearshore within the human-influenced shoreline halo (< 500 m), and b) flats at > 500 m from human-influenced shorelines. At each region, efforts will maximize the number of flats sampled (e.g., 6-10 flats in all regions except smaller-area Dry Tortugas). Fish will be sampled via nonlethal blood extraction, which allows comparison of plasma concentrations to human daily dosages and thus allow us to assess if fish experience either the targeted or side effect of the drug (Schulz et al. 2020). Fish will be sampled by extracting 1 to 2 mL of blood (depending on fish size to ensure extraction of < 7% of blood volume, <u>AVMA</u> recommendations) from the ventral caudal vein. Samples will be spun in a centrifuge within 6 hr of extraction, frozen and covered to prevent light degradation in the field, and stored at -80 C in the lab until analysis. Fish will be released where collected after obtaining measurements, photographs and full recovery.

All samples will be analyzed for 95 common pharmaceuticals (Attachment 3) at Dr Jerker Fick's Pharmaceutical Laboratory at the Department of Chemistry at University of Umea in Umea, Sweden. Samples will be shipped on dry ice at the end of collection and analyzed using Solid Phase Extraction High Temperature Electro-Spray Ionization Low Resolution Triple Quadrupole Liquid Chromatography Mass Spectrometer (Cerveny et al. 2020). Dr Fick leads one of the premier pharmaceutical labs in the world, with over 70 publications on pharmaceutical analysis included in journals such as Science and Nature Communications (Brodin et al. 2013, Hellström et al. 2016, Richmond et al. 2018 and see reference list for additional publications produced by Dr Fick), and has analyzed our previously collected bonefish pharmaceutical samples (Attachments 3-5 & Section 11). Pharmaceutical concentrations will be analyzed using multivariate statistics that quantify similarity among groupings of pharmaceuticals detected (and for their concentrations) across regions and areas by building similarity matrices and ordinations to visualize relationships among regions/areas (in Primer®, Attachment 5B-C). We will compare the number of

pharmaceuticals detected across regions and areas using generalized linear models in R (Attachment 5A). Concentrations will be compared to human daily therapeutic concentrations to determine which pharmaceuticals are detected at concentrations that may have effects (either targeted or side effects), and the occurrence of therapeutic concentrations will be compared across regions/areas. We will conduct hotspot analysis using ArcGIS to identify locations of hot (clusters of high values) and cold (clusters of low values) spots in the occurrence and concentration data.

In TASK 2, we will sample bonefish prey, water and sediment to identify pathways of exposure at the hotspots identified in TASK 1. This sampling will allow us to quantify the relative importance of trophic vs. inhalational (via water and sediment) pathways of exposure. While previous work has shown that water can be the primary mode of uptake (Du et al. 2014), dietary uptake of pharmaceutical contaminants can also play a key role in exposure to higher-order consumers. Sediment can also have the highest contaminant burden relative to biota and water, and can thus be a major exposure route for benthic-dwelling organisms (Miller et al. 2021), such as bonefish and their benthic prey. Thus, there is a need to examine multiple trophic compartments that experience different routes of exposure, and thus varying degrees of bioaccumulation relative to non-trophic compartments (Lagesson et al. 2016, Richmond et al. 2018, Miller et al. 2021). Understanding the relative role of the trophic pathway is critical since pharmaceuticals can remain bioavailable in biota for weeks to months, and can re-enter the food web at later time through the consumption of contaminated prey, despite a dissipation of water concentrations (Lagesson et al. 2016). The feeding strategy and habitat use of the prey (i.e., pelagic vs. benthic) can be an important determinant of their role in exposure, with studies showing higher concentrations in both benthicdwelling (Lagesson et al. 2016, Miller et al. 2021), and filter-feeder prey that consume suspended fine particulate matter (Richmond et al. 2018).

Sampling of prey, water and sediment will be conducted at a maximum of 5 hotspots identified in TASK 1. We will select 1 hotpot per region (based on the most concerning occurrence or concentration) in order to have spatial representation (Attachment 2). At each hotspot, we will collect 5 sediment samples (2 g), 10 water samples (500 mL) and 20 prey samples (2 g; 35 samples x 5 hotspots = 175 total samples). For the water samples, 5 will be collected at the hotspot site, and 5 will be collected at any nearby potential mainland sources if present (e.g., canals; else additional samples will be collected at the hotspot). For the 20 prey samples, sampling will be assigned to 4 potential trophic channels of exposure (5 replicates each): benthic vertebrate, benthic invertebrate, pelagic vertebrate and pelagic invertebrate. Candidate species for each channel will include dominant bonefish prey items (i.e., toadfish, snapping shrimp, mud and swimming crabs, snails and polychaetes, Crabtree et al. 1998). All samples will be analyzed for the same 95 pharmaceuticals described in **TASK 1**. Samples will be homogenized for analysis (Miller et al. 2021, Cerveny et al. 2020), and shipped on dry ice at the end of collection. We will use multivariate statistics to compare pharmaceutical groupings across hotspots as done with the bonefish plasma samples. We will compare the number of pharmaceuticals and occurrence of therapeutic concentrations across pathways (prey, sediment, water) and prey types (nested within pathway) using generalized linear models in R.

e. Environmental Impact: Impacts by the proposed study are minimal. Sample collection of bonefish in year 1 is nonlethal, and will be conducted using standard angling methods and best practices for catch -and-release and AVMA fish handling and sampling standards. Sample collection of prey species in year 2 is only for a small number of species and individuals, and uses a small seine, which has minimal impact on the submerged aquatic vegetation sampled.

<u>f. Future Efforts:</u> The proposed 2-year study is not part of a larger project, but is a stand-alone effort

aimed at understanding the threat of pharmaceuticals to South Florida ecosystems. Although future efforts are not needed for the success of the project, outcomes and outputs of the proposed work are informative to future efforts: 1) to understand the magnitude of the problem posed by emerging pollutants in South Florida, 2) to determine which ecosystem components are most vulnerable to exposure, 3) to examine sources of pollution in South Florida waters, 4) to expand monitoring needs to achieve protection, and 5) to identify best practices for reducing pharmaceuticals entering coastal environments.

3. Project Management

g. Administration: **Dr. JS Rehage** is an Associate Professor in the Institute of Environment at Florida International University (FIU) and will be lead investigator in the project. Dr. Rehage has over 15 yrs. of experience working on coastal fishes and the recreational fisheries they support throughout South Florida, with 65 peer-reviewed publications in the field. Dr. Rehage will coordinate and oversee all aspects of the project, including field and laboratory activities, data analyses, the writing of reports, scientific and popular publications, and the organization of presentations and the stakeholder workshop at the end of the study.

h. Roles & Responsibilities: **Dr. RO Santos** is an Assistant Professor at FIU, and a seascape ecologist with expertise in spatial processes in coastal ecosystems. Dr. Santos will lead the spatial analysis of the bonefish data to identify hotspots (**TASK 1**), and will aid in the production of reports and publications. Dr. Santos will contribute 2.5 weeks of time to the project. **Dr. WR James** is a Postdoctoral Fellow and coastal ecologist at FIU with expertise in trophic ecology and quantitative data analyses. Dr. James will assist with field work, lead the analysis of the trophic vs. nontrophic pathways of exposure, and lead the organization of the stakeholder workshop. Dr. James will contribute 2 months of his time to the project. A **full-time field technician** to be hired will lead all

field activities, sample processing, and shipment of samples to Sweden, as well as data entry, qa/qc and management and assist with data analyses and report/manuscript/popular article/presentation production. The technician will contribute 100% of the time to the project.

4. Support Requirements & Conditions

<u>i. Permit or cooperation required</u> - Research permits issued by Biscayne National Park, Dry Tortugas NP and the FKNMS will be needed for sampling activities. Estimated time for permits are 30 days for FKNMS, and 90 days for NPS. Applications for both will be submitted as soon as the award is processed in order to avoid delays in project timeline.

j. Data or facility access: Access to the Bonefish and Tarpon Trust vessel will be needed for all field activities (see Sections 10-11 & Attachment 1).

PROJECT SCHEDULE		Year 1				Year 2			
Deliverables & Milestones	Q1	Q2	Q3	Q4	Q1	Q2	Q3	Q4	
Task 1: SPATIAL EXTENT EXPOSURE									
Obtain research permits for sampling									
Identify guides to assist in bonefish sample collection									
Sampling of bonefish plasma via vessel-based angling									
Plasma sample preparation & shipment to Sweden									
Analysis of samples at Umea University Lab									
Data analysis to identify hotspots of exposure									
Check-in meetings with FIU team to track progress									
Project progress meetings with team & partner (BTT)									
Submit semi-annual reports									
Presentation at a scientific/stakeholder conference									
Production of manuscript 1 & popular articles									
Task 2: PATHWAYS OF EXPOSURE									
Renew research permits for sampling									
Sample collection of prey, water & sediment at hotspots									
Sample processing for analysis									
Sample preparation & shipment to Sweden									
Analysis of samples at Umea University Lab									
Data analysis to examine relative importance of pathways									
Check-in meetings with FIU team to track progress									
Project progress meetings with team & partner (BTT)									
Submit semi-annual reports									
Presentation at a scientific/stakeholder conference									
Final meeting with project team to develop final report									
Production of manuscript 1 & popular articles									
Organize stakeholder workshop for information transfer									
Submit & edit final report with Project Officer feedback									

Q1 = January-March, Q2 = April-June, Q3 = July-September, Q4 = October-December

5. Project Schedule

k. Milestone & deliverable schedule: The project will last 2 years and will consist of **2 TASKS**. The table above describes the timing of the milestones and deliverables for the **2 TASKS**.

6. Environmental Results - Outputs & Outcomes

1. Outputs: The table below describes all outputs and outcomes for the project. Our outputs and outcomes are directly linked to EPA's Strategic Plan's Objective 1.2 Provide for Clean and Safe Water, Objective 2.1 Enhance Shared Accountability, Objective 2.2. Increase Transparency and Public Participation and Objective 3.3 Prioritize Robust Science. The goal of our study is to better understand the threat of pharmaceuticals to South Florida, which aligns with Objective 1.2. Our information transfer workshop aligns with Objective 2.2., our partnership with Bonefish and Tarpon Trust aligns with Objective 2.1, and our efforts to study substances with high uncertainty with the goal of influencing policy aligns with Objective 3.3.

OUTPUTS

- Data on concentrations of 110 pharmaceuticals in bonefish plasma, georeferenced from Biscayne Bay to Dry Tortugas
- GIS maps showing hotpots of pharmaceutical exposure for bonefish from Biscayne Bay to Dry Tortugas
- Data on concentrations of 95 pharmaceuticals in bonefish prey, water & sediment samples at hotspots of exposure
- 4. Analysis of the relative importance of trophic vs respiration exposure pathways
- Workshop to disseminate project findings with managers & stakeholders
- 6.Publications in peer-reviewed scientific journals describing all project findings
- 7. Publications in popular fishing magazines, news outlets and social media describing all project findings
- 8.Presentations of findings at regional and national conferences

OUTCOMES

- Improved understanding of pharmaceutical risk & water quality issues in South Florida ecosystems
- Better comprehension of the pathways of pharmaceutical exposure to components of the ecosystem
- Insight into hotspots of pharmaceutical exposure that informs on contamination sources
- Identification of monitoring & water quality improvements needed to achieve protection of South Florida ecosystems
- Increase partnership among academic, nonprofit organizations and federal institutions
- 6. Enhanced communication of water quality issues throughout South Florida stakeholders & resource managers

m. Tracking outputs & outcomes: We will evaluate progress toward outputs and outcomes via 3 criteria: project schedule, project costs, and quality of data products. First, we will assess if

deliverables and milestones are being completed per the proposed timeline. We will monitor project progress with quarterly check-in progress meetings (see Timeline Table), and we will forecast and update the project schedule as needed in order to effectively complete planned tasks semi-annually (by update reports). Second, we will conduct a similar procedure for monitoring project performance financially, and ensure that project funds are being spent appropriately across semi-annually. Third and most important, we will monitor and evaluate the quality of data products at a semi-annual time step, and make adjustments to the allocation of effort as needed to achieve and maintain the high quality of outputs and outcomes we aim for in the project.

7. Programmatic Capability & Past Performance

- **1.** E Gaiser (PI), J Fourqurean, K Grove, **JS Rehage (coPI)** & J Kominoski. 2018-2024. Effects of pulse amplification on the resilience of the Florida Coastal Everglades. National Science Foundation LTER Program, Florida Coastal Everglades, \$ 7,004,800 (\$792,674 to Rehage). https://fcelter.fiu.edu/ All project goals are being achieved on time, and the project is being managed successfully. All yearly reports submitted on time and approved by NSF, and 134 scientific papers published to date.
- **2. JS Rehage (PI), RO Santos (coPI)**, CR Kelble, MO Hall, JA Nelson. 2019-2023. Effects of freshwater inflows and seagrass die-off on recreational fisheries: A trophic & movement ecology approach, Everglades National Park, \$ 747,300. All project goals are being achieved on time, and the project is being managed successfully. All quarterly and yearly reports submitted on time and approved by NPS, and 1 scientific paper published and 1 in revision.
- **3. JS Rehage (PI).** 2008-2022. Role of marsh-mangrove habitats as dry-down refuges for marsh fishes. RECOVER (Restoration Coordination & Verification), Comprehensive Everglades Restoration Plan, US Army Corps of Engineers, \$ 1,209,662. All project objectives met and continue to be met on time, project has been managed successfully for 14 years and continues to be renewed every 5 years, all quarterly and yearly reports submitted on time and approved by ACOE, 22 scientific papers published (and 1 in revision and 3 in preparation).
- **4. Rehage (PI), RO Santos (coPI)**, RE Boucek, P Frezza, JJ Lorenz, P Stevens, J Ley, K Flaherty-Wallia, M Matheson. 2017-19. Assessing the effects of closure of Joe Bay on recreational fisheries. Everglades National Park, \$ 378,141 (\$258,141 to JS Rehage & RO Santos). All project objectives were met on time and we completed the work successfully providing management recommendations to NPS. All quarterly and yearly reports were submitted on time and approved by NPS, and 1 scientific paper in preparation and 1 popular article published.
- **5.** Sukop, R Jaffe, **JS Rehage (coPI)**, M Bhat, & P Mozumder. 2013-2018. Robust decision-making for south Florida water resources by ecosystem service valuation, hydro-economic optimization,

and conflict resolution modeling. National Science Foundation, Water Sustainability and Climate Change (WSC-Category 2), \$ 4,589,000 (\$ 1,443,863 to FIU, \$ 281,735 to JS Rehage). All interdisciplinary goals were achieved on time and all planned work was completed. Quarterly and yearly reports were submitted on time and approved by NSF, and 27 scientific articles were produced (4 by Rehage).

Rehage has close to 15 years of experience managing successfully and leading federal, state and NGO projects throughout South Florida totaling \$ 5,341,783 in funding awarded directly to Rehage and an additional \$ 10,673,746 awarded as coPI. Santos has over 13 years of experience and coleading research projects in South Florida.

8. Budget summary

a. Personnel. For PIs Rehage and Santos, 20% of their summer salary is requested (2.4 weeks), totaling \$12,716 in year 1 (Y1), and \$13,097 in year 2 (Y2). For postdoc James, 15% of his annual salary is being requested (1.8 months), totaling \$8,700 in Y1, and \$8,961 in Y2. Funds are requested for a full-time technician, \$35,000 in Y1, and \$36,750 in Y2. An annualized 3% increase applies to the PI salaries and a 5% annualized increase applies to the technician. **b. Fringe benefits.** A fringe benefit rate of 36.13% applies to the partial salaries requested for Rehage, Santos and James totaling \$7,738 for Y1 and \$7,970 for Y2, and a 4.09% Other OPS and Temporary Faculty fringe benefit rate applies to the full-time technician (\$1,432 for Y1 and \$1,503 for Y2). c. Travel. We are requesting funds for field travel totaling \$9,000 in Y1 and \$6,000 in Y2. In Y1, these funds will be used to collect bonefish plasma samples, and in Y2, funds will be used to collect prey, water and sediment samples. Funds will cover costs associated with field vehicles obtained from FIU, lodging, per diem (subsistence), and field boat expenses (gas). Travel costs to collect samples in the Dry Tortugas will include costs for the Yankee Freedom Ferry for travel to the site and camping fees. Field vehicles have a daily rate of \$73 + mileage/gas, research.fiu.edu/facilities/recharge/serc-fieldrates/). In Y1, funds will also compensate fishing guides for assistance in collecting bonefish samples using their own vessels (at a rate of \$300/day). We are requesting \$4,000 per year to cover

statistical training for James for project data analysis, in conference travel for Santos, Rehage and James to obtain critical knowledge related to the project at ecotoxicological conferences (e.g., SETAC), to present project findings to the scientific community, and to organize a workshop to present findings to regional stakeholders at the end of the study. d. Equipment. No equipment is being requested. e. Supplies. Supplies total \$3,600 in Y1, and \$2,700 in Y2. In Y1, expendable supplies include fishing gear and bait for bonefish collection, needles, vials, pipettes, 2 9-V centrifuges, cooler, foil, field notebooks. In Y2, expendable supplies include items needed to collect prey, water and sediment samples (small seine nets, dip nets, snorkeling gear, cooler, sample cups, vials and bags, foil, coring device, supplies for sample homogenization) and in both years data storage (external hard drives and cloud-based storage yearly fee). f. Contractual costs. All samples will be analyzed at Jerker Fick's Pharmaceutical Laboratory at the Department of Chemistry at University of Umea in Umea, Sweden at a rate of \$210 per sample, and will total \$26,250 in Y1 (125 bonefish plasma samples) and \$36,750 in Y2 (for 175 water, prey and sediment samples). h. Other. Funds totaling \$1,800 in Y1 and \$4,200 in Y2 are being requested for shipment of samples to Umea University in Sweden for analysis. Samples are shipped using World Courier on dry ice in order to maintain sample integrity and higher costs in Y2 are due to greater number and weight of water samples. i. Total direct costs. A total of \$110,236 in Y1 and \$121,931 in Y2 are requested in direct costs, with a grand project total of \$232,167 in direct costs. **i. Indirect costs**. An Off Campus indirect cost rate of 26% applies to all direct costs included in the project since more than 50% of the project will be performed off-campus. Indirect costs requested for Y1 total \$28,661, and for Y2 total \$31,702 k. Total costs. Total project costs are \$292,530, of which \$138,897 are requested in Y1, and \$153,633 are requested in Y2. Leveraged resources. Project partner Bonefish and Tarpon Trust (BTT) will provide support for the study in the manner of: a) field assistance for

sample collection, b) a research vessel to be used in sampling, and c) contacts with fishing guides to assist in bonefish sampling in Y1 (see Sections 10-11 & Attachment 1).

9. Distinguishing between subawards v/s contracts

There are no subawards in this study. A contract with the Pharmaceutical Lab at the University of Umea (Umea, Sweden) will provide analysis of all pharmaceutical samples.

10. Voluntary cost share/match and other leveraged funds

We have leveraged field assistance and a research vessel through our partnership with Bonefish and Tarpon Trust (BTT, see Sections 8, 11 & Attachment 1). The field assistance and research vessel will be critical to sample collection over the 2 years of the study, and will result in maximized sample efficiency that relies on BTT and fishing guide expertise and cost savings in both labor and field travel expenses.

11. Partnerships with other entities

We are partnering with BTT for this project. BTT is a science-based organization whose primary goal is to conserve and restore flats fisheries (bonefish, tarpon and permit) and their habitats through research stewardship, education and advocacy. BTT is the premier non-government organization working to understand and conserve socio-economically valuable recreational fisheries in South Florida and Caribbean Basin. In 2019-20, BTT funded a Caribbean study of pharmaceutical presence in bonefish, comparing the risk of contaminants across 4 bonefish populations (Key West, Mexico, Belize, Bahamas and Puerto Rico). The study showed that in 57 bonefish plasma samples collected, all bonefish had detectable pharmaceuticals, with an average of 5 pharmaceuticals per fish (Attachments 3-5). In Key West, the antidepressant Venlafaxine, the opioid Codeine, the betablocker Atenolol, and the opioid antagonist Naloxone were the most common pharmaceuticals detected in 15 samples, with a fish collected in Man Key having the highest number of

pharmaceuticals detected (16 compounds). Although these data provide a small number of samples for South Florida, the detections suggest that pharmaceuticals may be a concern to South Florida ecosystems and warrant further standardized sampling to better characterize risk as proposed in this study.

12. Information Transfer

We will disseminate project results (outputs/outcomes) and lessons learned through four venues: 1) articles published in peer-reviewed articles, 2) popular articles in local outlets (e.g., Florida Keys Free Press newspaper and WRLN Radio environmental news), fishing magazine articles and social media stories (i.e., Florida Sportsman, Fly Fishing Magazine, FlyLords), 3) presentations at local scientific conferences, particularly the Coastal and Estuarine Research Federation and the Greater Everglades Ecosystem Restoration meetings, and to regional working groups focused or linked on water quality (e.g., FKNMS Water Quality Protection Program, REstoration COordination & VERification's (RECOVER), Southern Coastal Systems workgroup), and 4) through a workshop we will organize to disseminate findings and identify next steps in both the research and management fronts stemming from our project. The workshop will be held at the end of year 2 and will include relevant stakeholders and resource managers (FKNMS, NOAA, RECOVER, ACOE, SFWMD, NPS, Biscayne Bay Task Force, FWC, BTT, Miami WaterKeeper, Captains for Clean Water, Upper Florida Keys Guide Association, Lower Keys Fishing Guide Association) and will include a presentation of study findings, presentations by project partner BTT, invited speakers with relevant data to our study and a discussion of next steps.

13. Literature Cited

(**Bold**=Papers produced by the Umea Lab to be used for sample analysis in this study)

Brodin, T et al. Dilute concentrations of a psychiatric drug alter behavior of fish from natural populations. *Science* 339.6121 (2013): 814-815.

Cerveny, D, et al. Bioconcentration and behavioral effects of four benzodiazepines and their environmentally relevant mixture in wild fish. *Sci Total Environ* 702 (2020): 134780.

Crabtree, RE, et al. Feeding habits of bonefish, Albula vulpes, from the waters of the Florida Keys. *Fishery Bull* 96.4 (1998): 754-766.

Du, B et al. Bioaccumulation and trophic dilution of pharmaceuticals across trophic positions of an effluent-dependent stream. *Philos Trans R Soc Lond B Biol Sci* 369.1656 (2014): 20140058.

Fabbri, E & S Franzellitti. Human pharmaceuticals in the marine environment: focus on exposure and biological effects in animal species. *Environ Toxicol Chem* 35.4 (2016): 799-812.

Fedler A. Economic impact of the Florida Keys flats fishery. Report to Bonefish & Tarpon Trust (2013).

Hellström, G et al. GABAergic anxiolytic drug in water increases migration behaviour in salmon. *Nature Communications* 7.1 (2016): 1-7.

Klaminder, J et al. Less anxious salmon smolt become easy prey during downstream migration." Sci Total Environ 687 (2019): 488-493.

Küster, A and N Adler. Pharmaceuticals in the environment: scientific evidence of risks and its regulation. *Philos Trans R Soc Lond B Biol Sci* 369.1656 (2014): 20130587.

Lagesson, A et al. Bioaccumulation of five pharmaceuticals at multiple trophic levels in an aquatic food web-Insights from a field experiment. *Sci Total Environ* 568 (2016): 208-215.

McKenzie, T et al. Submarine groundwater discharge: A previously undocumented source of contaminants of emerging concern to the coastal ocean. *Mar Pollut Bull* 160 (2020): 111519.

Miller, TH et al. Multicompartment and cross-species monitoring of contaminants of emerging concern in an estuarine habitat. *Environ Pollut* 270 (2021): 116300.

Ng, B et al. Understanding the occurrence and distribution of emerging pollutants and endocrine disruptors in sensitive coastal South Florida Ecosystems. *Sci Total Environ* 757 (2021): 143720.

Richmond, EK et al. A diverse suite of pharmaceuticals contaminates stream and riparian food webs. *Nature Communications* 9.1 (2018): 1-9.

Schulz, M et al. Revisited: Therapeutic and toxic blood concentrations of more than 1100 drugs and other xenobiotics. *Critical Care* 24.1 (2020): 1-4.

Wennmalm, Å & B Gunnarsson. Pharmaceutical management through environmental product labeling in Sweden. *Environment International* 35.5 (2009): 775-777.

Attachment 1



July 23, 2021

RE: Letter of support for 2021 SOUTH FLORIDA GEOGRAPHIC PROGRAM, FON: EPA-R4-SFL-2021-01

Dear EPA panel reviewers,

On behalf of Bonefish and Tarpon Trust, I am pleased to support the EPA Region 4 proposal titled *An examination of the risk of pharmaceuticals in South Florida: extent and pathways of exposure in a valuable recreational fishery, bonefish* by principal investigators Rehage, Santos and James at Florida International University.

The project aims to examine: 1) the spatial extent and pattern of exposure to pharmaceuticals in bonefish and identify hot spots of exposure; 2) the pathways of exposure to pharmaceuticals (trophic vs. respiration) at those hotpots. The study builds on a study by FIU that we funded, and we are excited to see the initial survey of pharmaceutical exposure in bonefish expanded to a larger study. As a partner in the study, we will provide leveraged resources that will assist FIU in the collection of samples, reduce project costs, and ensure the success of the study. Specifically, we will provide a research vessel for sample collection, field assistance in sampling bonefish and prey, and facilitate interactions with fishing guides that will assist in the collection of samples.

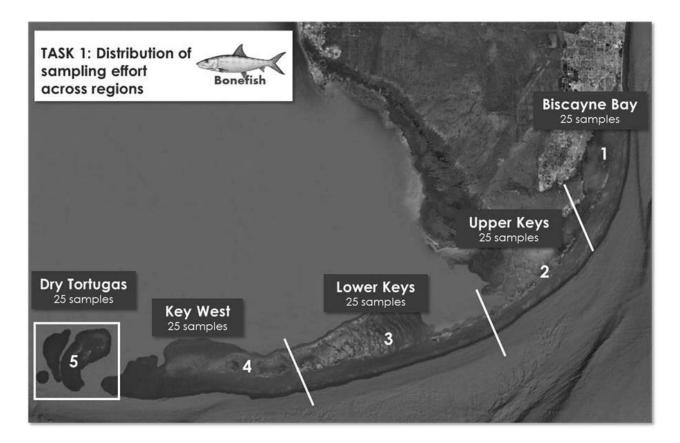
We believe this project is of incredible value to informing water quality issues in South Florida and thus our is a project that we strongly support. Pharmaceuticals are emergent contaminants that are not part of water quality monitoring programs in Florida Keys, Florida Bay & Biscayne Bay, and little is known about their presence even though the data from the initial study shows that the potential for pharmaceuticals to be a threat to bonefish and other biota is very high.

Sincerely,

Aaron Adams, Ph.D.

Director of Science and Conservation

ATTACHMENT 2. Map showing distribution of bonefish to be sampled across the 5 regions of interest: 1) Biscayne Bay, 2) Upper Keys, 3) Lower Keys, 4) West of Key West (Lakes and Marquesas) and 5) Dry Tortugas (25 fish per region). At each region, angling will target 2 areas of interest: a) flats nearshore within the human-influenced shoreline halo (< 500 m), and b) flats at > 500 m from human-influenced mainlands.



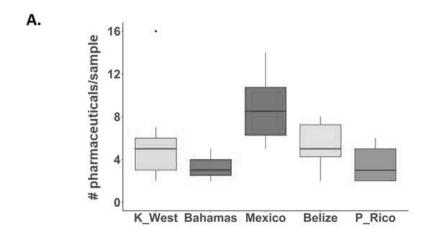
ATTACHMENT 3. Table showing the 95 pharmaceuticals to be analyzed in the study, shown by drug class in alphabetical order (28 total classes).

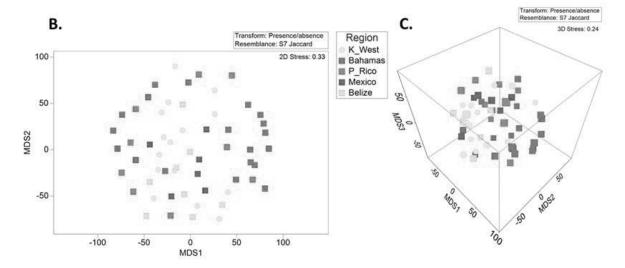
Drug Class	Pharmaceutical	Drug Class	Pharmaceutical
ACE inhibitor	Cilazapril	Bronchodilator	Terbutalin
Adrenergic_Agent	Alfuzosin	Cholesterol treatment	Atorvastatin
Adrenergic_Agent	Finasteride	Cholesterol treatment	Ezetimibe
Adrenergic_Agent	Flutamid	Cholesterol treatment	Fenofibrate
Alzheimer's treatment	Donepezil	Cholesterol treatment	Rosuvastatin
Alzheimer's treatment	Memantin	Diabetes	Glibenclamide
Antacid	Ranitidine	Diabetes	Glimepiride
Antiarrhythmic	Amiodiarone	Diabetes	Repaglinide
Antiarrhythmic	Flecainide	Diuretic	Furosemide
Antiarrhythmic	Sotalol	Motion Sickness	Meclozine
Antibiotic	Azithromycine	Muscle Relaxant	Atracurium
Antibiotic	Ciprofloxacin	Opioid	Buprenorphin
Antibiotic	Clarithromycine	Opioid	Codeine
Antibiotic	Clindamycine	Opioid	Tramadol
Antibiotic	Erythromycine	Opioid_Antagonist	Naloxon
Antibiotic	Norfloxacin	Parkinson's treatment	Biperiden
Antibiotic	Ofloxacin	Parkinson's treatment	Bromocriptin
Antibiotic	Oxytetracycline	Parkinson's treatment	Orphenadrin
Antibiotic	Roxithromycine	Parkinson's treatment	Trihexyphenidyl
Antibiotic	Sulfamethoxazol	Psychoactive	Alprazolam
Antibiotic	Tetracycline	Psychoactive	Amitryptiline
Antibiotic	Trimetoprim	Psychoactive	Bupropion
Anticholinergic	Dicycloverin	Psychoactive	Carbamazepin
Anticoagulant	Dipyridamol	Psychoactive	Chloprothixe n
Antidiarrhea	Loperamide	Psychoactive	Chlorpromazine
Antifungal	Clotrimazol	Psychoactive	Citalopram
Antifungal	Fluconazole	Psychoactive	Clomipramine
antifungal	Ketoconazole	Psychoactive	Clonazepam
Antifungal	Miconazole	Psychoactive	Cyproheptadine
Antihistamine	Azelastine	Psychoactive Psychoactive	Duloxetin
Antihistamine	Clemastine	Psychoactive Psychoactive	Flunitrazepam
Antihistamine	Desloratidin	Psychoactive	Fluoxetin
Antihistamine	Diphenhydramin	Psychoactive Psychoactive	Flupetixol
Antihistamine	Fexofenadine	Psychoactive Psychoactive	Fluphenazine
Antihistamine Antihistamine	Promethazin	Psychoactive Psychoactive	Haloperidol
Antihypertensive	Diltiazem	Psychoactive Psychoactive	
			Hydroxyzine
Antihypertensive	Eprosartan Eolodinino	Psychoactive Psychoactive	Maprotilin
Antihypertensive	Felodipine	Psychoactive	Mianserin
Antihypertensive	Ibersartan	Psychoactive Psychoactive	Mirtazapine
Antihypertensive	Telmisartan	Psychoactive Psychoactive	Oxazepam
Antihypertensive	Verapamil	Psychoactive Psychoactive	Paroxetin
Anti-inflammatory	Diclofenac	Psychoactive	Perphenazine piantifa a
Anti-migraine	Dihydroergotamin	Psychoactive	Pizotifen
Asthma treatment	Budesonide	Psychoactive	Risperidone
Betablocker	Atenolol	Psychoactive	Sertraline
Betablocker	Bisoprolol	Psychoactive	Venlavafaxin
Betablocker	Metoprolol	Psychoactive	Zolpidem
Breast Cancer Treatment	Tamoxifen		

ATTACHMENT 4. Table of detections in BTT study (see Section 11), showing pharmaceuticals detected across the 5 regions: Key West, Puerto Rico, Bahamas, Belize & Mexico. Key West had the most detections across 15 bonefish sampled compared to other regions. Only pharmaceuticals for which there was at least one detection in a sample are shown (44 pharmaceuticals detected in 18 drug classes out of 95 pharmaceuticals analyzed). The table is sorted by number of detections in Key West bonefish. A psychoactive drug, the opiod Codeine, and a betablocker were most commonly detected pharmaceuticals in the 15 samples analyzed from Key West.

Pharmaceutical	Drug Class	Key West Detections (n=15)	Puerto Rico Detections (n=9)	Bahamas Detections (n=19)	Belize Detections (n=8)	Mexico Detections (n=6)
Venlavafaxin	Psychoactive	13	1	5	6	9
Codeine	Opioid	10	2	5	4	3
Atenolol	Betablocker	8	3	3	5	5
Naloxon	Opioid Antagonist	6	4	4	5	5
Fluconazole	Antifungal	4	2	4	5	2
Alfuzosin	Adrenergic Agent	4	4	3	1	2
Mianserin	Psychoactive	3	0	0	1	1
Biperiden	Parkinsons	3	0	1	0	2
Atracurium	Muscle Relaxant	3	2	0	1	3
Diphenhydramin	Antihistamine	3	0	2	0	2
Bisoprolol	Betablocker	2	2	2	2	0
Clotrimazol	Antifungal	2	3	0	2	2
Trimetoprim	Antibiotic	2	2	11	1	3
Risperidone	Psychoactive	1	2	0	0	1
Hydroxyzine	Psychoactive	1	0	0	4	4
Paroxetin	Psychoactive	1	0	1	0	0
Chlorpromazine	Psychoactive	1	0	0	0	0
Flupetixol	Psychoactive	1	0	0	0	0
Fluphenazine	Psychoactive	1	0	0	0	0
Oxazepam	Psychoactive	1	0	0	0	0
Perphenazine	Psychoactive	1	0	0	0	0
Sertraline	Psychoactive	1	0	0	0	0
Trihexyphenidyl	Parkinsons	1	1	0	0	1
Diclofenac	NSAID	1	1	1	0	1
Atorvastatin	Lipid	1	0	0	0	0
Ibersartan	Antihypertensive	1	0	0	0	0
Clemastine	Antihistamine	1	0	0	2	0
Dicycloverin	Anticholinergic	1	0	0	0	0
Flecainide	Antiarrhythmic	1	0	1	0	2
Memantin	Alzheimers	1	0	3	1	1
Carbamazepin	Psychoactive	0	0	0	0	1
Haloperidol	Psychoactive	0	0	1	0	0
Duloxetin	Psychoactive	0	0	1	0	0
Pizotifen	Psychoactive	0	0	1	0	0
Orphenadrin	Parkinsons	0	0	1	0	0
Metoprolol	Betablocker	0	1	2	1	2
Eprosartan	Antihypertensive	0	0	0	0	3
Telmisartan	Antihypertensive	0	0	1	0	0
Diltiazem	Antihypertensive	0	0	0	0	0
Azelastine	Antihistamine	0	3	0	1	0
Desloratidin	Antihistamine	0	0	3	0	0
Clindamycine	Antibiotic	0	0	1	0	0
Tetracycline	Antibiotic	0	0	0	0	1
Sotalol	Antiarrhythmic	0	0	3	0	0
	16 Unique Pharma Classes	80 Total Detections	33 Total Detections	60 Total Detections	42 Total Detections	53 Total Detections

ATTACHMENT 5. Details of the BTT study showing A) the mean number of pharmaceuticals per plasma sample (+/-SE) across 5 areas of study: Key West, Bahamas, Mexico, Belize & Puerto Rico. Horizontal lines are means, boxes are SEs, & vertical lines denote minimum and maximum values. On average, 9 pharmaceuticals/sample were detected in Mexico (the highest value), compared to 5 pharmaceutical/bonefish in Key West. Non-metric multidimensional scaling (nMDS) shows similarity of pharmaceutical detections across all bonefish plasma samples using a presence absence matrix in B) two dimensions and C) three dimensions (due to high stress in 2D). There are no distinct groupings across regions shown by the mixing of colors. This suggests that the same types of pharmaceuticals are being detected across multiple regions in the Caribbean (with Belize/Mexico showing the least spread in pharmaceuticals, and Bahamas showing the most variance in the identity of pharmaceuticals detected across samples).





JS Rehage, PhD

Associate professor, Earth & Environment, Institute of Environment, Florida International University, Miami, FL, rehagei@fiu.edu, 305.348.3804

(a) PROFESSIONAL PREPARATION							
INSTITUTION	LOCATION	MAJOR / AREA OF STUDY	DEGREE	YEAR			
Florida International Univ	Miami, FL	Environmental Studies	BS	1995			
University of Kentucky	Lexington, KY	Ecology	PHD	2003			
USGS	Homestead, FL	Ecology of fishes	Postdoc	2004-05			

(b) APPOINTMENTS

- 2014 Present Associate Professor, Earth & Environment Dept, Institute of Water & Environment, Florida International University, Miami, FL
- 2008 2014 Assistant Professor, Earth & Environment Dept, Southeast Environmental Research Center, Florida International University, Miami, FL
- 2006 2008 Assistant Professor, Oceanographic Center & Farquhar College of Arts & Sciences, Nova Southeastern University, Dania, FL

(c) PRODUCTS (Graduate students, †postdocs)

- Gervasi, CL, JS Rehage, RO Santos, RJ Rezek†, WR James†, RE Boucek, CR Bradshaw, C Kavanagh, J Osborne. *In press.* Bottom-up conservation: Using stakeholder knowledge to inform conservation priorities for an unregulated and recreationally valued fish species. Canadian Journal of Fisheries and Aquatic Sciences.
- Baker, R, MD Taylor, WR James †, R. Rezek †, **JS Rehage**, and 20 others. 2020. Letter to Science: Fisheries rely on threatened salt marshes. Science 370: 670-6710 doi: 10.1126/science.abe9332.
- <u>Brown CE</u>, MG Bhat, **JS Rehage**. 2020. Valuing ecosystem services under climate risk: a case of recreation in the Florida Everglades. Journal of Water Resources Planning & Management 146: 04020089. https://doi.org/10.1061/(ASCE)WR.1943-5452.0001290
- Rastetter EB, MD Ohman, KJ Elliott, **JS Rehage**, VH Rivera-Monroy, <u>RE Boucek</u>, E Castañeda-Moya, TM Danielson, L Gough, P Groffman, CR Jackson, CF Miniat, GR Shaver. 2021. Future trajectories for ecosystems in the U.S. Long-term Term Ecological Research Network: The importance of time lags. Ecosphere. DOI: 10.1002/ecs2.3431
- **Rehage JS**, RO Santos†, <u>EKN Kroloff</u>, JE Heinen, Q Lai, B Black, <u>RE Boucek</u>, AJ Adams. 2019. How has the quality of bonefishing changed? Using local ecological knowledge to assess temporal patterns in the South Florida Flats Fishery. Environmental Biology of Fishes 102: 285-298. https://doi.org/10.1007/s10641-018-0831-2
- Adams JA, **JS Rehage**, SJ Cooke. 2019. A multi-methods approach is essential for effective management and conservation of the ecotourism-based recreational flats fishery (*Introductory paper to special issue on Fishes of the Flats*). Environmental Biology of Fishes 102: 105-115. https://doi.org/10.1007/s10641-018-0840-1
- Mirchi, A, DW Watkins, V Engel, MC Sukop, J Czajkowski, M Bhat, **JS Rehage**, D Letson, Y Taktsuka, R Weisskoff. 2018. A hydro-economic model of South Florida water resources system. Science of the Total Environment 628-629:1531-1541.
- Santos RO†, **JS Rehage**, <u>E Kroloff</u>, AJ Adams, JE Osborne. 2017. Quantitative assessment of a data-limited recreational bonefish fishery using a timeseries of fishing guides reports. PLoS ONE 12(9): e0184776. https://doi.org/10.1371/journal.pone.0184776

(d) SYNERGISTIC ACTIVITIES

Associate Editor: Env Biol of Fishes, L&O Letters; Contributor to Report Card on Everglades & System Status Reports (SSR) to US Congress on progress of Everglades Restoration; expert technical evalution for Comprehensive Everglades Restoration Plan; research featured on nonscience media: Watermen, SilverKings & Sportfishing TV fishing shows, Florida Sportsman, Fishsens, Coastal Angler & National Geographic online magazines, Miami Herald, Patagonia Fish Activist & Jeff Corwin's Ocean Mysteries.

Rolando O. Santos Corujo, Ph.D.

Assistant Professor – Florida International University, Dept. of Biological Sciences / Institute of Environment 3000 NE 151 St, MSB 351, Miami, Florida 33181 Email: rsantosc@fiu.edu – Cellphone: (787) 462-8565



Education:

Ph.D. - Marine Biology and Fisheries. **December 2014**. University of Miami, Rosenstiel School of Marine and Atmospheric Science, FL

Dual M.S. - Marine Biology and Coastal Management. 2010. Nova Southeastern University, FL

Dual B.S. - Environmental Science and Geography. **2004**. University of Puerto Rico - Río Piedras, SJ, Puerto Rico

Academic positions:

Assistant Professor (2020-present) Department of Biological Sciences, Florida International University, Miami, FL

Postdoctoral Research Associate (2018-2020) CASE Distinguished Postdoctoral Fellowship, Florida International University, Miami, FL

Postdoctoral Research Associate (2015-2018), Earth & Environment Department & Rehage Lab Bonefish and Tarpon Trust-funded project, Florida International University, Miami, FL

Selected Peer-reviewed Publications:

Graduate students

Santos RO, <u>James WR</u>, Nelson J, Rehage JS, Pittman S, Serafy J, and Lirman D (*in review*) Isotopic niche variation of an omnivorous fish as a function of spatial characteristics of submerged aquatic vegetation seascapes. Ecosphere

<u>James WR, Topor ZM, Santos RO</u> (2020) Seascape structure influences the community structure of marsh nekton. Estuaries and Coasts https://doi.org/10.1007/s12237-020-00853-7

Koval G, Rivas N, D'Alessandro, Hesley D, **Santos RO**, Lirman D (2020) Fish predation hinders the success of coral restoration efforts using microfragmented massive corals. Peer J DOI 10.7717/peerj.9978

<u>Stipek C</u>, **Santos RO**, Lirman D, Babcock E (2020) Modelling the resilience of seagrass communities exposed to pulsed freshwater discharges: A seascape approach. PLOS ONE https://doi.org/10.1371/journal.pone.0229147

Santos RO, Lirman D (2012) Using habitat suitability models to predict changes in seagrass distribution caused by water management practices. Canadian Journal of Fisheries and Aquatic Sciences 69:1-9 https://doi.org/10.1139/f2012-018

Lirman D, Thyberg T, Herlan J, Hill C, Young-Lahiff C, Schopmeyer S, Huntington B, **Santos RO**, and Drury C (2010) Propagation of the threatened staghorn coral Acropora cervicornis: methods to minimize the impacts of fragment collection and maximize production. Coral Reefs 29:729-735 https://doi.org/10.1007/s00338-010-0621-6

W. Ryan James, PhD

Postdoctoral Research Associate Florida International University Institute of the Environment 11200 SW 8th St, Miami, FL 33199

Professional Preparation

University of Louisiana Lafayette	Environ and Evo Biology	PhD 2020
University of Alabama at Birmingham	Biology	MS 2016
University of Alabama	Biological Sciences	BS 2013

Selected Publications

Nelson JA, Harris JM, Lesser JS, **James WR**, Suir SM, & WP Broussard III (2020) New mapping metrics to test functional response of food webs to coastal restoration. *Food webs* https://doi.org/10.1016/j.fooweb.2020.e00179

Harris JM[†], **James WR**[†], Lesser JS[†], Doerr JC, & JA Nelson (2020) Foundation species shift alters the energetic landscape of marsh nekton. *Estuaries & Coasts* doi: 10.1007/s12237-020-00852-8

Lesser JS, **James WR**, Stallings CD, Wilson RM, & JA Nelson (2020) Trophic niche size and overlap decrease with increasing ecosystem productivity. *Oikos* doi: 10.1111/oik.07026

Jones SF, Stagg CL, Yando ES, **James WR**, Buffington KJ, & MW Hester (2020) Stress gradients interact with disturbance to reveal alternative states in salt marsh: Multivariate resilience at the landscape scale. *Journal of Ecology* https://doi.org/10.1111/1365-2745.13552

James WR, Santos RO, Rehage JS, Doerr JC, & JA Nelson (2020) *E*-scape: consumer specific landscapes of energetic resources derived from stable isotope analysis and remote sensing. *bioXriv* doi: https://doi.org/10.1101/2020.08.03.234781

James WR, JS Lesser, SY Litvin, & JA Nelson (2019) Assessment of food web recovery following restoration using resource use metrics. *Science of the Total Environment* doi: 10.1016/j.scitotenv.2019.134801.

Kimball ME, Connolly RM, Alford SB, Colombano DD, **James WR**, Kenworth MD, Norris GS, Ollerhead J, Ramsden S, Rehage JS, Sparks EL, Waltham NJ, Worthington TA, & Taylor MD (2021) Novel applications of technology for advancing tidal marsh ecology. *Estuaries & Coasts* doi: 10.1007/s12237-021-00939-w

Pittman SJ, Yates KL, Bouchet PJ,... James WR... (34 additional authors) (2021) Seascape ecology: Identifying research priorities for an emerging ocean sustainability science. *Marine Ecology Progress* Series DOI: https://doi.org/10.3354/meps13661

Baker R, Taylor MD, Beck MW, Cebrian J, Colombano DD, Connolly RM, Currin C, Deegan LA, Feller IC, Gilby BL, Kimball ME, Minello TJ, Rozas LP, Simenstad C, Turner RE, Waltham NJ, Weinstein MP, Ziegler SL, zu Ermgassen PSE, ... James WR... (29 additional authors) (2020) Fisheries rely on threatened salt marshes. *Science* DOI: 10.1126/science.abe9332

James WR, Topor ZM, & RO Santos (2020) Seascape structure influences the community structure of marsh nekton. *Estuaries & Coasts* doi: 10.1007/s12237-020-00853-7

OMB Number: 2030-0020 Expiration Date: 04/30/2021

EPA KEY CONTACTS FORM

Authorized Representative: Original awards and amendments will be sent to this individual for review and acceptance, unless otherwise indicated.

Name: Prefix: Mr.			First Name: Roberto						Middle Name:	М				
	Last	Name:	Gutierrez						Suffix:					
Title:	Assi	stant	VP for Res	earch										
Comple	te Ad	dress:												
Street	t1: [11200	SW 8 St											
Street	:2 :	MARC 4	:30											
City:		Miami				State:	FL: Florid	la						
Zip / F	Postal	Code:	33199			Country:	USA: UNI	FED STATES	3					
Phone I	Numb	er:	305-348-249	94			Fax Numl	ber:	05-348-4117					
E-mail A	Addre	ss:	gutierrr@fi	u.edu										
Payee:	Indivi	dual au	thorized to a	ccept paymen	ts.									
Name:	Prefix	x: Ms.		First Name:	Donna				Middle Name:					
		Name:	Kiley						Suffix:					
Title:			Post Award											
Comple														
Street	. г		SW 8 St											
Street	:2 :	MARC 4	:30											
City:	[Miami				State:	FL: Florida	a						
Zip / F	ostal	Code:	33199			Country:	USA: UNIT	ED STATES						
Phone I	Numb	er:	305-348-249	94			Fax Numb	oer: 30	305-348-4117					
E-mail A	Addre	ss:	dkiley@fiu.											
			-											
			ntact: Indivi	-	nsored Prog	grams Offic	ce to contact	concerning	g administrati	ve matters (i.e., indii	rect cost			
Name:	Prefix	x: Mr.		First Name:	Roberto			ı	/liddle Name:	М				
	Last	Name:	Gutierrez						Suffix:					
Title:	Assi	istant	VP for Res	earch										
Comple	te Ad	dress:												
Street	:1: [11200	SW 8 St											
Street	:2 :	MARC 4	:30											
City:		Miami				State:	FL: Florida	a						
Zip / F	Postal	Code:	33199			Country:	USA: UNIT	TED STATES						
Phone I	Numb	er:	305-348-249	94			Fax Numb	oer: 30	5-348-4117					
E-mail A	E-mail Address:		gutierrr@fi	u.edu										

EPA Form 5700-54 (Rev 4-02)

EPA KEY CONTACTS FORM

Project Manager: *Individual responsible for the technical completion of the proposed work.*

Name:	Prefix:		First Name:	Jennifer					Middle N	Name:	S		
	Last Name:	Rehage								Suffix:			
Title:	Associate	Professor											
Comple	te Address	<u>!</u>											
Stree	t1: 11200	SW 8 St											
Stree	t2 : AHC5	365											
City:	Miami				State:	FI	ı: Florida	a					
Zip / I	Postal Code:	33199			Count	ry:	USA: UNIT	ED STATE	ES				
Phone I	Number:	305-348-38	04				Fax Numb	er:					
E-mail A	Address:	rehagej@fi	u.edu										

BUDGET INFORMATION - Non-Construction Programs

OMB Number: 4040-0006 Expiration Date: 02/28/2022

SECTION A - BUDGET SUMMARY

	Grant Program Function or	Catalog of Federal Domestic Assistance	Estimated Unob	ligated Funds	New or Revised Budget							
	Activity	Number	Federal	Non-Federal		Federal	Non-Federal	Total				
(a)		(b)	(c)	(d)		(e)	(f)	(g)				
1.	2021 South Florida Geographic Program	66.484	\$	\$	\$	292,530.00	\$	\$ 292,530.00				
2.												
3.												
4.												
5.	Totals		\$	\$	\$	292,530.00	\$	\$ 292,530.00				

Standard Form 424A (Rev. 7- 97) Prescribed by OMB (Circular A -102) Page 1

SECTION B - BUDGET CATEGORIES

6 Object Class Categories				GRANT PROGRAM. F	FUN	NCTION OR ACTIVITY			Total	
6. Object Class Categories		(1))	(4)	(5)		
		2021 South Florida			1					
		Geographic Program								
]					
a. Personnel	\$	115,224.00	\$		\$		\$	\$	115,224.00	
					1					
b. Fringe Benefits		18,643.00							18,643.00	
		23,000.00			1				23,000.00	
c. Travel		23,000.00			1				23,000.00	
d. Equipment					1					
u. Equipment					4					
e. Supplies		6,300.00							6,300.00	
f. Contractual		63,000.00							63,000.00	
					1					
g. Construction]					
					1					
h. Other		6,000.00							6,000.00	
i. Total Direct Charges (sum of 6a-6h)		232,167.00			1			\$	232,167.00	
i. Total Direct Charges (sum of 6a-6n)		232,167.00			1			'	232,167.00	
j. Indirect Charges		60,363.00][\$	60,363.00	
, manoot onargos					1			*	,	
k. TOTALS (sum of 6i and 6j)	\$	292,530.00	\$		\$		\$	\$	292,530.00	
in 101ALO (Sum of of und of)					1					
			l		ı			1		
7. Program Income	\$		\$		\$		\$	\$		
7. 1 Togram income			Ĺ		<u>'Ľ</u>					

Authorized for Local Reproduction

Standard Form 424A (Rev. 7- 97) Prescribed by OMB (Circular A -102) Page 1A

		SECTION	C -	NON-FEDERAL RESO	UR	CES				
	(a) Grant Program			(b) Applicant	(c) State			(d) Other Sources	(e)TOTALS	
8. 2021 South Florida Geographic Program			\$	0.00	\$	0.00	\$	0.00	\$	0.00
9.]			
10.										
11.										
12. TOTAL (sum of lines 8-11)				0.00	\$	0.00	\$	0.00	\$	0.00
		SECTION	D -	FORECASTED CASH	NE	EDS				
		Total for 1st Year		1st Quarter	,	2nd Quarter		3rd Quarter		4th Quarter
13.	Federal	\$ 138,897.00	\$	34,724.00	\$	34,724.00	\$	34,724.00	\$	34,725.00
14.	Non-Federal	\$								
15.	TOTAL (sum of lines 13 and 14)	\$ 138,897.00	\$	34,724.00	\$	34,724.00	\$	34,724.00	\$	34,725.00
	SECTION E - BUD	GET ESTIMATES OF FE	DE	RAL FUNDS NEEDED	FO	R BALANCE OF THE	PR	OJECT		
	(a) Grant Program		FUTURE FUNDING PERIODS (YEARS)							
			1	(b)First		(c) Second	_	(d) Third		(e) Fourth
16.	2021 South Florida Geographic Program		\$	38,408.00	\$	38,408.00	\$	38,408.00	\$	38,409.00
17.										
18.										
19.										
20.	TOTAL (sum of lines 16 - 19)	38,408.00	\$	38,408.00	\$	38,408.00	\$	38,409.00		
		SECTION F	- C	THER BUDGET INFOR	RMA	ATION				
21.	Direct Charges: \$232,167			22. Indirect	Cha	arges: MTDC 26.0% Off	Ca	mpus Rate (\$60,363)		
23.	Remarks:									

Authorized for Local Reproduction

Standard Form 424A (Rev. 7- 97) Prescribed by OMB (Circular A -102) Page 2

Other Attachment File(s)

* Mandatory Other Attachment Filename:	DHHS FA	Rate	AGM	033121	.pdf
Add Mandatory Other Attachment Delete	Mandator	y Other	Attac	chment	View Mandatory Other Attachment

To add more "Other Attachment" attachments, please use the attachment buttons below.

Add Optional Other Attachment Delete Optional Other Attachment View Optional Other Attachment

The following attachment is not included in the view since it is not a read-only PDF file.

Upon submission, this file will be transmitted to the Grantor without any data loss.

DHHS FA Rate AGM 033121.pdf