BIOGRAPHICAL SKETCH

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NAME: BRITO, Gerly Anne de Castro

eRA COMMONS USER NAME (credential, e.g., agency login): GABNIH

POSITION TITLE: Full Professor of Medical Histology and Embryology – PROPAP/UFC

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable. Add/delete rows as necessary.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
Federal University of Ceará, Brazil	MD	08/1987	Medicine
Federal University of Ceará, Brazil	Residency	02/1990	Internal Medicine
Federal University of Ceará, Brazil	MS	09/1995	Pharmacology
Federal University of Ceará, Brazil	PhD	12/1997	Pharmacology
University of Virginia, Charlottesville, VA, USA	Postdoctoral Training	02/2002	Infectious Disease

A. Personal Statement

I have a strong research background in inflammatory responses, more focused in the gastrointestinal tract, from mouth to the intestines. I have specific expertise in bench research involving *Clostridioides difficile (C. difficile)* infection, more concentrated in its pathogenesis. I have led research around the effects of *C. difficile* toxins on epithelial cell apoptosis, neutrophil morphology and function, the Wnt/beta catenin pathway, adenosine/adenosine deaminase system, modulation with glutamine alanyl-glutamine, and on the role of enteric glial cells. My research group have ongoing collaborations with national and international leaders in the field, including Dr. Cirle A. Warren (12 publications in collaboration) and Dr. Richard Guerrant (20 articles published in collaboration), both from University of Virginia where I did my postdoctoral training, investigating the mechanisms associated with cell death in response to *C. difficile* toxin A. In cooperation with Dr. Vivaldo Moura Neto from Instituto Estadual do Cerébro Paulo Niemeyer, we investigated the role of enteric glial cells in inflammatory bowel conditions (Nogueira et al., 2017; Costa et al, 2019). The last article provides evidence that 5-Fluorouracil, an anticancer agent induces reactive gliosis and reduction of enteric neurons in a S100B/RAGE/NFκB-dependent manner, since pentamidine, a S100B inhibitor, prevented 5-FU-induced neuronal loss, enteric glia activation, intestinal inflammation, oxidative stress and histological injury. In addition, the collaboration with Dr. Moura-Neto resulted in a review article on glial cell function (Coelho-Aguiar et al., 2015) and a book chapter (Coelho-Aguiar et al., 2020). I also carry out a pioneer and translational research, investigating the *Clostridium difficile* incidence in Northeast of Brazil, in collaboration with Prof. Carlos Quezada Gómez and Esteban Chaves-Olarte, both from Costa Rica University. I'm an author of over 203 international publications, H index 36 Researcher ID: C8597-2013, and I have large experience as a PI on several Brazilian-funded grants (30 grants to date), including six grants on *C. difficile* research. I have mentored the 10 post-doctoral (2 current, 8 concluded) and 24 PhD student (4 current, 20 concluded) and 19 MS (2 current and 17 concluded). Together, these activities brought me the ability and experience to be a PI at this proposed grant.

Nogueira, L. T., Costa, D. V., Gomes, A. S., Martins, C. S., Silva, A. M., Coelho-Aguiar, J. M., Castelucci, P., Lima-Júnior, R. C., Leitão, R. F., Moura-Neto, V., & **Brito, G. A**. (2017). The involvement of mast cells in the irinotecan-induced enteric neurons loss and reactive gliosis. Journal of neuroinflammation, 14(1), 79. https://doi.org/10.1186/s12974-017-0854-1

Costa, D., Bon-Frauches, A. C., Silva, A., Lima-Júnior, R., Martins, C. S., Leitão, R., Freitas, G. B., Castelucci, P., Bolick, D. T., Guerrant, R. L., Warren, C. A., Moura-Neto, V., & **Brito, G. A**. (2019). 5-Fluorouracil Induces Enteric Neuron Death and Glial Activation During Intestinal Mucositis via a S100B-RAGE-NFκB-Dependent Pathway. Scientific reports, 9(1), 665. https://doi.org/10.1038/s41598-018-36878-z

Coelho-Aguiar, J., Bon-Frauches, A. C., Gomes, A. L., Veríssimo, C. P., Aguiar, D. P., Matias, D., Thomasi, B. B., Gomes, A. S., **Brito, G. A**., & Moura-Neto, V. (2015). The enteric glia: identity and functions. Glia, 63(6), 921–935. https://doi.org/10.1002/glia.22795

Coelho-Aguiar, Juliana; Pires Veríssimo, Carla; Viana da Silva Costa, Deiziane; Bastos de Moraes Thomasi, Beatriz; Carina Bon Frauches, Ana; Pereira Ribeiro, Fabiana ; Lucia Tavares Gomes, Ana ; **Brito G.A.**; MOURA-NETO, VIVALDO . The Enteric Glial Network Acts in the Maintenance of Intestinal Homeostasis and in Intestinal Disorders. Glia in Health and Disease. 1ed.: IntechOpen, 2020, v. 1, p. 35-62. DOI: 10.5772/intechopen.89170

B. Positions and Honors

Positions and Employment

1990-1990	Primary Care Physician, City Health Department
1991-1997	Assistant Professor, Department of Morphology, Universidad Federal do Ceará, Brazil
1997- 2011	Associate Professor, Department of Morphology, Universidade Federal do Ceará, Brazil
2000-2002	NIH-ITREID Research Post-Doctoral Fellow, Division of Infectious Disease, University of Virginia School of Medicine, Charlottesville, VA, USA
2011-	Full Professor, Department of Morphology, Universidade Federal do Ceará, Brazil/PROPAP
2010-2016	Member of National Committee for evaluation of proposals for grants in the Morphology and Cellular Biology, Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), Brasília, DF
2007-2015	Member of State Committee for evaluation of proposals for grants in the Cellular and Environmental Biology, Fundação Cearense de Apoio a Pesquisa (FUNCAP), Fortaleza, CE
2009-2014	Member of Administrative Committee of Instituto de Biomedicina do Semiárido Brasileiro (IBISAB)
2019-	Member of National Committee for evaluation of proposals for grants in the Morphology and Cellular Biology, Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), Brasília, DF

Other Experience and Professional Memberships

2013-	Member, Brazilian Society of Cellular Biology
2005-	Member, Brazilian Society of Anatomy
2003-	CNPq Peer Review Committee: Morphology, ad hoc reviewer

Honors

1995	Outstanding Young Investigator Award, Serra Negra, SP, Brazil
2002	Celso Werneck Ribeiro Award, Brazilian Society of Cancer
2015	Joseph Lister Award – Boston, Mass, USA
2020	Selected between 250 Brazilian researchers of all fields highlighted by Gender and Number/ Serrapilheira Institute / Open Box Science platform.

C. Contribution to Science

My initial contribution on *C. difficile* pathogenesis was the demonstration that TcdA alters *in vitro*-adherent human neutrophil morphology and function through TcdA–induced glucosylation of the signaling small-size guanine 50-triphosphate–binding proteins of the Rho family. At the same time, we showed that TcdA induces apoptosis of human intestinal epithelial cells (T84) by a mechanism dependent on inactivation of Rho, activation of caspases 3, 6, 8, and 9 and Bid, and mitochondrial damage followed by cytochrome c release. These findings provided important information on basic biological responses to injury and suggested that TcdA–induced changes in human neutrophil shape and function and apoptosis in human intestinal epithelial cells, which play a role in the pathogenesis of *C. difficile* colitis. We also demonstrated the roles of glutamine and alanyl-glutamine in inhibiting cell apoptosis and damage by blocking caspase 8 activation and reducing TcdA-induced intestinal secretion and mucosal disruption. These results suggest the potential of glutamine and alanyl-glutamine as adjuvant therapeutic measures in *C. difficile* induced disease. I served as the principal investigator on three of these studies, corresponding author in one, and was co-investigator in another one, resulting in the following publications:

Brito GA, Sullivan GW, Ciesla WP Jr, Carper HT, Mandell GL, Guerrant RL. (2002) *Clostridium difficile* toxin A alters in vitro-adherent neutrophil morphology and function. J Infec tDis. May 1;185(9):1297-306.

Brito GA, Fujji J, Carneiro-Filho BA, Lima AA, Obrig T, Guerrant RL. (2002) Mechanism of *Clostridium difficile* toxin A-induced apoptosis in T84 cells. J Infect Dis. Nov 15;186(10):1438-47.

Brito GA, Carneiro-Filho B, Oriá RB, Destura RV, Lima AA, Guerrant RL. (2005) Clostridium difficile toxin A induces intestinal epithelial cell apoptosis and damage: role of Gln and Ala-Gln in toxin A effects. Dig Dis Sci. Jul;50(7):1271-8.

Carneiro BA, Fujii J, **Brito GA**, Alcantara C, Oriá RB, Lima AA, Obrig T, Guerrant RL. (2006) Caspase and bid involvement in Clostridium difficile toxin A-induced apoptosis and modulation of toxin A effects by glutamine and alanyl-glutamine in vivo and in vitro. Infect Immun. Jan;74(1):81-7.

Santos AA, Braga-Neto MB, Oliveira MR, Freire RS, Barros EB, Santiago TM, Rebelo LM, Mermelstein C, Warren CA, Guerrant RL, **Brito GA.**(2013).Glutamine and alanyl-glutamine increase RhoA expression and reduce *Clostridium difficile* toxin-A-induced intestinal epithelial cell damage. Biomed Res Int.:152052. doi: 10.1155/2013/152052.

I continued my studies showing that glutamine depletion potentiates acute inflammation induced by carrageenan or *C. difficile*, possibly by increasing neutrophil migration through resident cell activation and production of IL-1β and TNF-α and that glutamine supplementation reverses these effects and may be useful during inflammatory catabolic stress induced by this infection. Reinforcing the importance of neutrophil infiltration on TcdA effects, we showed that fucoidin, an inhibitor of leukocyte rolling through L-selectin blockage, reduced TcdA-induced secretion, edema, mucosal disruption, inflammatory-cell infiltration and adenosine deaminase (ADA) activity. This was the first demonstration that TcdA induces increase in the activity of the adenosine catabolizing enzyme, suggesting a role for adenosine in *C. difficile* pathogenesis. Thus, the next study demonstrated an important effect of the A2A adenosine receptor agonist, ATL 313, on toxin A-induced mucosal disruption, cell death, ADA activity, and inflammatory cell infiltration. Following this data, we showed that the inhibition of adenosine deaminase by EHNA prevent TcdA-induced damage and inflammation possibly through the A2A adenosine receptor, suggesting that the modulation of adenosine/adenosine deaminase may represents an important tool in the management of *C. difficile*-induced disease. The results were described in the following scientific publications, in which I served as the corresponding author.

Nascimento SB, Sousa RB, Martins MJ, Souza Gomes A, Souza MH, Guerrant RL, Cunha FQ, Ribeiro RA, **Brito GA**. (2005) Glutamine depletion potentiates leucocyte-dependent inflammatory events induced by carrageenan or *Clostridium difficile* toxin A in rats. Immunology. Nov;116(3):328-36.

Barreto AR, Cavalcante IC, Castro MV, Junqueira AF, Vale MR, Ribeiro RA, Souza MH, **Brito GA**. Fucoidin prevents Clostridium difficile toxin-A-induced ileal enteritis in mice. (2008) Dig Dis Sci. Apr;53(4):990-6.

Cavalcante IC, Castro MV, Barreto AR, Sullivan GW, Vale M, Almeida PR, Linden J, Rieger JM, Cunha FQ, Guerrant RL, Ribeiro RA, **Brito GA**. (2006) Effect of novel A2Aadenosine receptor agonist ATL 313 on Clostridium difficile toxin A-induced murine ileal enteritis. Infect Immun. May;74(5):2606-12.

de Araújo Junqueira AF, Dias AA, Vale ML, Spilborghs GM, Bossa AS, Lima BB, Carvalho AF, Guerrant RL, Ribeiro RA, **Brito GA**. (2011) Adenosine deaminase inhibition prevents Clostridium difficile toxin A-induced enteritis in mice. Infect Immun.Feb;79(2):653-62. doi: 10.1128/IAI.01159-10. PMCID: PMC3028843

FOSCHETTI, D.A. ; BRAGA-NETO, M.B. ; BOLICK, D. ; MOORE, J. ; ALVES, LA. ; MARTINS, CS. ; BOMFIN, LE. ; SANTOS, AAQA. ; LEITÃO, RFC. ; BRITO, GAC. ; WARREN, CA. . Clostridium difficile toxins or infection induce upregulation of adenosine receptors and IL-6 with early pro-inflammatory and late anti-inflammatory pattern. Brazilian Journal of Medical and Biological Research, v. 53, p. 9877, 2020.

In addition to the contributions described above, in association with a team of collaborators, I also investigated the participation of Angiotensin II subtype 1 receptor in TcdA-induced intestinal secretion, providing evidence that AT_1 receptor inhibition, is a potentially novel approach to control *C. difficile* colitis and diarrhea. Furthermore, we demonstrated that haem oxygenase-1/carbon monoxide pathway exerts a protective role in the TcdA-induced enteritis, and that retinol protects epithelial cell damage induced by TcdA. The demonstration of modulators of TcdA damage may be important for the management of the *C. difficile*-associated disease. Another important contribution of our group was the first demonstration that TcdA inhibits Wnt/β-catenin signaling in cultured intestinal epithelial cells, affects β-catenin protein levels, and prevents c-MYC expression induced by Wnt3a independently of GSK3β inhibition. I served as the corresponding author on two of these articles.

Alcantara CS, Jin XH, **Brito GA**, Carneiro-Filho BA, Barrett LJ, Carey RM, Guerrant RL. (2005) Angiotensin II subtype 1 receptor blockade inhibits *Clostridium difficile* toxin A-induced intestinal secretion in a rabbit model. J Infect Dis.Jun 15;191(12):2090-6.

Medeiros CA, Warren CA, Freire R, Vieira CA, Lima BB, Vale ML, Ribeiro RA, Souza MH, **Brito GA**. (2011) Role of the haem oxygenase/carbon monoxide pathway in Clostridium difficile toxin A-induced enteritis in mice. J Med Microbiol. Aug;60(Pt 8):1146-54. doi: 10.1099/jmm.0.028910-0.

Maciel AA, Oriá RB, Braga-Neto MB, Braga AB, Carvalho EB, Lucena HB, **Brito GA**, Guerrant RL, Lima AA. (2007) Role of retinol in protecting epithelial cell damage induced by Clostridium difficile toxin A. Toxicon. Dec 15;50(8):1027-40.

Bezerra Lima B, Faria Fonseca B, da Graça Amado N, Moreira Lima D, Albuquerque Ribeiro R, Garcia Abreu J, **de Castro Brito GA** (2014) *Clostridium difficile* toxin A attenuates Wnt/β-catenin signaling in intestinal epithelial cells. InfectImmun. Jul;82(7):2680-7. doi: 10.1128/IAI.00567-13.

MARTINS, CONCEIÇÃO S. ; COSTA, DEIZIANE V. S. ; LIMA, BRUNO B. ; LEITÄO, RENATA F. C. ; FREIRE, GILDÊNIO E. ; SILVA, GUILHERME F. M. ; PACÍFICO, DVISON M. ; ABREU, JOSÉ G. ; Brito, Gerly A. C. . Clostridioides difficile Toxin A-Induced Wnt/b-Catenin Pathway Inhibition Is Mediated by Rac1 Glucosylation. Frontiers in Microbiology, v. 11, p. 1998, 2020.

Since 2013 we have been engaged in epidemiological study about the incidence of *C. difficile* infection in four major hospital (3 of than public): Hospital at Institute of Cancer of Ceara – Haroldo Juaçaba Hospital, Children Hospital Albert Sabin, University Hospital Walter Cantídio and Infectious Disease Hospital São José. This research has relevancy for public health, since there are no other laboratory performing isolation and strain characterization of *C. difficile* infection in the Ceara State and neither in Northeast of Brazil. Another important contribution of this research is the creation and publication of a guide, written by our group, for hospital visitors and healthcare workers to preventing *C. difficile* infection. The study resulted in the following publication (Costa et al., 2014; Quesada-Gómez et al., 2015; Costa et al., 2016; Costa et al, 2017). I served as the corresponding author on three of these articles.

Costa CL, Quesada-Gómez C, de Carvalho CB, González RH, Gifoni MA, Ribeiro RA, **de Castro Brito GA.** Community-acquired diarrhea associated with Clostridium difficile in an HIV-positive cancer patient: first case report in Latin America. Int J Infect Dis. 2014 Sep; 26:138-9. doi: 10.1016/j.ijid.2014.06.010.

Quesada-Gómez C, López-Ureña D, Acuña-Amador L, Villalobos-Zúñiga M, Du T, Freire R, Guzmán-Verri C, del Mar Gamboa-Coronado M, Lawley TD, Moreno E, Mulvey MR, **de Castro Brito GA**, Rodríguez-Cavallini E, Rodríguez C, Chaves-Olarte E. Emergence of an outbreak-associated Clostridium difficile variant with increased virulence. J Clin Microbiol. 2015 Apr;53(4):1216-26. doi: 10.1128/JCM.03058-14.

Costa CL, López-Ureña D, de Oliveira Assis T, Ribeiro RA, Silva RO, Rupnik M, Wilcox MH, de Carvalho AF, do Carmo AO, Dias AA, de Carvalho CB, Chaves-Olarte E, Rodríguez C, Quesada-Gómez C, **de Castro Brito GA**. A MLST Clade 2 Clostridium difficile strain with a variant TcdB induces severe inflammatory and oxidative response associated with mucosal disruption. Anaerobe. 2016 Aug; 40:76-84. doi:10.1016/j.anaerobe.2016.06.005.

Costa CL, Mano de Carvalho CB, González RH, Gifoni MAC, Ribeiro RA, Quesada-Gómez C, **Brito GAC**. Molecular epidemiology of Clostridium difficile infection in a Brazilian cancer hospital. Anaerobe. 2017 Dec;48:232-236. doi:10.1016/j.anaerobe.2017.

Complete List of Published Work in MyBibliography:
https://www.ncbi.nlm.nih.gov/myncbi/1JW5h50UyCaAs/bibliography/public/

D. Research Support

Ongoing Research Support

PRONEX/FUNCAP/CNPq Edital 02/2015 (Brito) 01/01/16-01/01/22
Center of Excellence in *Clostridium Difficile* from Research in State of Ceará (NEPEC-CE)
This project proposes to continue a comprehensive study of the pathogenesis and epidemiology of the disease induced by *Clostridioides difficile*, as well as the characterization of strains isolated in Ceará state hospitals with a look back to the diagnosis, prevention and treatment. We will address determinants of environment relationship, host and microbial agent, the following parameters: (1) infection; (2) pathogenesis in animal models and in vitro (3) Biological and molecular characterization of *C. difficile* strains isolated and (5) effects the enteric nervous system and gut motility. Role: PI

PROCAD-CAPES Edital 071/2013 (Moura, Neto, Brito - MPI) 10/01/14-01/12/21
Enteric glia in metabolic and inflammatory disease
This is a collaborative study between three Brazilian Universities: University Federal of Rio de Janeiro, University of Ceará and University of Alagoas, which the goal is to investigate the role of enteric glia in metabolic disease and intestinal inflammatory disease such as *C. difficile* induced enteritis in mouse model and intestinal mucositis induced by antineoplastic drugs. Role: co-PI

Completed Research Support

Edital MCT/CNPq N º 14/2013–Universal (Brito) 01/01/14-12/31/17
Clostridium difficile Toxin A on WNT/beta-catenin and TGF-beta/SMAD pathway
The goal of this study was to investigate the role of the two interconnected pathways, TGF-β/SMAD and Wnt/β-catenina, in *C. difficile* TcdA induced damage in intestinal epithelial cell line and in mouse ileal loop model. Role: PI

PPSUS-CNPq/-03/2012 (Brito) 12/21/11-06/30/15
Science and Technology Ministry/Health Science Ministry
Clostridium difficile strains in patients with diarrhea in Hospital Haroldo Juaçaba of Cancer Institute of Ceará
The goal of this study is to make the diagnosis, isolate, investigate the genotype of the strains found in in cancer patients with diarrhea in HHJ of Cancer Institute of our State-Brazil and compare the virulence of these strains in mouse model of ileal loop. This was a cooperative grant with Carlos Quezada Gómez and Esteban Chaves Olarte from University of Costa Rica. Role: PI

International Collaboration- Edital 05/2013 (Brito) 08/01/13-08/31/15
CAPES/FUNCAP
Clostridium Difficile TcdA and TcdB from different genotypes: cytotoxic and virulent
The goal of this study was to reinforce the existing interaction between UFC/Brazil and University of Costa Rica research group in *C. difficile* investigation.

CNPq/MCT- (Lima) 01/01/09-12/31/14
Instituto de Biomedicina do Semi-árido Brasileiro (IBISAB)
The *IBISAB* intends to function as leader in basic science, pre-clinical (*in vitro* and *in vivo*) and clinical research of markers and bioproducts, including genetic, environmental and etiological determinants of high prevalence endemic diseases in the Semi-Arid and Northeastern Brazil regions.
Role: Member of Management Committee