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Dehydrodieugenol isolated from *Nectandra leucantha* (Lauraceae) induces plasma membrane permeabilization of *Trypanosoma cruzi*

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Abstract: Parasitic disease are responsible for considerable morbidity and mortality worldwide, especially in underdeveloped and developing countries. Among the types of diseases that still attack Latin America population, are those caused by protozoa, such as, Chagas disease. It's estimated that about 6 to 7 million people are infected worldwide, mostly in Latin America [1]. Nifurtimox and benznidazole are the two currently drugs used in the treatment of Chagas disease, presenting several side effects such as, hepatic intolerance, hypersensitivity reactions, neuropsychiatric symptoms and others [2]. Aiming the discovery of new prototypes to the treatment of Chagas disease in the Brazilian flora, a previous work reported the isolation of three new neolignans from twigs of *Nectandra leucantha* with significant antileishmanial and immunomodulatory activity against *Leishmania donovani* [3]. In this work, the hexane extract from leaves of *N. leucantha* displayed promising activity against amastigotes and trypomastigotes of *Trypanosoma cruzi* and was subjected to a bioactivity guided fractionation using HSCCC and Sephadex LH-20 to afford one active neolignan, identified as dehydrodieugenol. Amastigotes and trypomastigotes of *T. cruzi* were incubated with dehydrodieugenol displaying IC_{50} values 15.05 μM (13.27 to 17.07 μM) and 11.47 μM (9.28 to 14.18 μM) respectively, while the standard drug, benznidazole, displayed IC_{50} values 440.7 μM (272.4 to 478.4 μM) and 319.7 μM (283.8 to 360.1 μM). Citotoxicity against NCTC cells of dehydrodieugenol and benznidazole were also evaluated with IC_{50} values 58.15 μM and 469.9 μM respectively. Therefore, selectivity index of isolated compound and standard drug were calculated for 3.9 and 1.1 respectively. The death mechanisms of dehydrodieugenol was investigated by using the dye SYTOXGreen® (plasma membrane permeability), probe rhodamine 123 (mitochondrial membrane potential) and H_2DCf-DA (reactive oxygen species induction). No changes in the production of ROS and in the mitochondrial membrane potential were observed, while the exposure of trypomastigotes of *T. cruzi* to dehydrodieugenol led to a gradual increase in SYTOXGreen® influx, demonstrating a time-dependent alteration in permeability of the plasma membrane, This result suggested that this compound alter the fluidity and integrity of the plasma membrane of *T. cruzi* parasites. This effect could be related to the affinity for ergosterol, which is the primary component of the *T. cruzi* membrane and is functionally linked to maintenance of structural integrity and protection from biotic stress [4]. In view that compounds in clinical use, for parasitical diseases, induces pore formation in the plasma of parasites, due to the high affinity for ergosterol [5], the identification of new biotargets involved in the synthesis of ergosterol, could be essential for the development of more efficient therapeutic alternatives. (FAPESP, CAPES and CNPq)

References:

[1] World Health Organization. 2015. Chagas disease (American trypanosomiasis - Fact sheet N°340). Retrieved from: http://www.who.int/mediacentre/factsheets/fs340/en/
[2] Rojo, G. *et al*, 2014. Toxic and therapeutic effects of nifurtimox and benznidazole on *Trypanosoma cruzi* ex vivo infection of human placental chorionic villi explants. Acta Tropica, 132: 112-118.
[3] Costa-Silva, T.A. *et al*, 2015. Immunomodulatory and Antileishmanial Activity of Phenylpropanoid Dimers Isolated from *Nectandra leucantha*. Journal of Natural Products, 78: 653–657.
[4] Souza, W. and Rodrigues, J.C.F. 2009. Sterol Biosynthesis Pathway as Target for Anti-trypanosomatid Drugs. Interdisciplinary Perspectives on Infectious Diseases, 2009: 1-19.
[5] Saha, A.K., *et al.* 1986. Mechanism of action of amphotericin B on *Leishmania donovani* promastigotes. Molecular Biochemical Parasitology, 19: 195-200.