American trypanosomiasis also known as Chagas disease (CD) is a neglected disease caused by the intracellular protozoan Trypanosoma cruzi. Sesquiterpenes lactones (STL) like cynaropicrin, are terpenoid compounds characteristic of the Asteraceae family, exhibiting a wide variety of chemical structures with pharmacological effects in a large number of biological test systems including anti-inflammatory, anti-tumour and antimicrobial effects. As part of multidisciplinary study to identify novel anti-T.cruzi candidates, bloodstream trypomastigotes (BTs – Y strain) were incubated at 37°C for 24 h in the presence of increasing doses (0-6.65 µg/mL) of cynaropicrin. Our data showed that this SL exhibited an EC50 value of 1 ± 0.2 µg/mL, being lower than the reference drug, benznidazole (EC50 3 µg/mL). Aiming to analyze the cellular targets of this compound, transmission electron microscopy analysis of BTs treated or not for 2 h at 37°C with the corresponding EC50/24h was performed. The samples were fixed for 60 min at 4 °C with 2.5% glutaraldehyde 2.5 mM CaCl2 in 0.1 M cacodylate buffer, pH 7.2 and post-fixed for 1 h at 4 °C with 1% OsO4, 0.8% potassium ferricyanide 2.5 mM CaCl2 using the same buffer. Next, treated and untreated parasites were routinely processed for TEM and examined using a Zeiss EM10C electron microscope (Oberkochen, Germany). Ultrastructural analysis demonstrated that while untreated parasites exhibited normal morphology like mitochondrion, kinetoplast and flagellum (Fig. 1B), cynaropicrin treated BTs showed intense intracellular vacuolization, occurrence of large multivesicular profiles and membrane projections (Fig. 1C), which can be suggestive of autophagy, a type II programmed cell death (PCD) mechanism. Biochemical studies are underway in order to better characterize the molecular events associated to the triggering of T.cruzi cell death by SLs agents.

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Fig. 1: Chemical structure of cynaropicrin (A). Transmission electron micrographs of cynaropicrin effect on bloodstream trypanosomes: Untreated parasites display typical morphology (B), while treated parasites (C) show vacuolization (*) and plasma membrane projection (arrow).