

CH-87-18 INDUCED-ANTIBACTERIAL FACTORS IN THE HEMOLYMPH OF Rhodnius
prolixus

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The humoral antibacterial defense reactions in insects have been subject of increasing interest over the last years, due in large measure to a series of important and stimulating findings with the diapausing pupae of the lepidopteran Hyalophora cecropia (H. G. Boman & D. Hultmark, 1981, Trends Biochem. Sci., 6: 306-309; D. Hultmark et al., 1983, The EMBO J., 2: 571-576). However, relatively few investigations on hematophagous hemipterans have been carried out, in spite of their great significance as vectors for Chagas' disease. Our laboratory has therefore investigated the humoral reaction of the bloodsucking bug, Rhodnius prolixus, against bacteria infection. Injection of live Streptococcus mutans into adults of R. prolixus results in the appearance in the hemolymph of small proteins showing antibacterial activity. One of these substances is a peptide, heat-stable, with molecular weight of 7,000 Daltons which acts only on bacteria in growth phase (P. Azambuja et al., 1985, An. Acad. brasil. Ciênc., 57:133-134; P. Azambuja et al., 1986, J. Insect Physiol., 32: 807-812). This activity is related to (i) the disappearance of the S. mutans from the hemolymph and (ii) the protection observed in vaccinated insects challenged with a lethal dose of S. mutans. The induced-peptide is able to liberate of labelled-DNA in the supernatant of the culture medium of S. mutans previously labelled with H^3 -thymidine (P. Azambuja et al., 1986), and it seems to belong to a family of basic peptides with antibacterial activity due to its characteristics to be very similar with cecropin (D. Hultmark et al., 1980, Eur. J. Biochem., 106: 7-16).

On the other hand, using Micrococcus lysodeikticus cell walls as a substrate, a second inducible antibacterial factor was characterized as lysozyme (P. Azambuja & E. S. Garcia, 1987, Brazilian J. Med. Biol. Res., in press). The enzyme has a rate of lytic activity strongly negatively dependent on ionic strength (Na^+ and K^+) and it presents a thermostability at acidic pH. It also binds to SP-Sephadex at pH 5.5, and has an apparent molecular weight of 15,000 Daltons. All these properties make Rhodnius lysozyme

strikingly similar to other insect and vertebrate lysozymes. It seems that the site of synthesis of the induced antibacterial factors is the fat body since in vitro incubation of this tissue from insects inoculated with bacteria releases antibacterial substances into culture medium (P. Azambuja & E. S. Garcia, 1987). The question of the possible importance of the hemocytes for lysozyme production is now under investigations.

Finally, injection of Enterobacter cloacae induces an antibacterial factor which is unable to lysis itself but kill Escherichia coli. Notwithstanding, these antibacterial factors, recent data from our laboratory indicate that other factors may be produced by the insects after bacteria inoculation showing a certain degree of specificity in the building up the humoral immunity system in R. prolixus

These studies were financed by the CNPq (PIDE Nº 400627/85) and FINEP (Nº 43.86.0197.00).