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IMPLANTATION-ASSOCIATED PROTEINASE ACTIVITIES IN A MARSUPIAL (MACROPUS EUGENII). H.-W. Denker* and C.H. Tyndale-Biscoe, Abteilung Anatomie der RWTH, Aachen (West Germany), and CSIRO Division of Wildlife Research, Canberra (Australia).

In all marsupials, the early embryo is surrounded by 3 coatings: zona pellucida, mucolemma and a keratinous shell membrane (Hughes, J. Reprod. Fert. 39, 173, 1974). In the tammar wallaby (Macropus eugenii) the former two disappear at about day 10, and the shell membrane persists until about day 18 of the 27 day gestation period at which time the trophoblast becomes intimately apposed to the uterine epithelium. Using a sensitive gelatin substrate film technique optimized previously for the detection of trophoblast-dependent proteinase (blastolemmase) in the rabbit (Denker, Histochemistry 38, 331 and 39, 193, 1974; Adv. Anat. Embryol. Cell Biol. 53 Part 5, 1977), a study of implantation-associated proteinase activity was performed in tammar embryos and uteri collected on days 18, 19 and 20 of pregnancy.

Two different gelatinolytic proteinase activities were found: 1) An SH-dependent endopeptidase of the cathepsin type with acid pH optimum, localized predominantly in trophoblast and endoderm of the avascular yolk sac region, and in glandular epithelium and some stroma cells of the endometrium. 2) A particularly interesting proteinase activity was found on day 19 in the extracellular space between trophoblast and uterine epithelium in the avascular yolk sac region where the shell membrane was in the process of undergoing disintegration. This enzyme was not detected in any other location nor at any of the other stages investigated. It shows an alkaline pH optimum like rabbit blastolemmase which is likewise most active in disintegrating blastocyst coverings. However, according to the results of inhibitor experiments, the enzymes are not homologous. The rabbit enzyme is a serine endopeptidase whereas the marsupial one reveals properties of an SH proteinase: It is inhibited by iodoacetamide and antipain, not by aprotinin, alpha-1-antitrypsin and soybean trypsin inhibitor; it does not require divalent cations. These preliminary data are indicative of an independent evolution, in the marsupial, of an enzyme which appears to be functionally analogous but structurally not homologous to rabbit blastolemmase. (Supported in part by Deutsche Forschungsgemeinschaft grant No. De 181/9-4).

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