Acrosin system of dog spermatozoa and reproductive tract secretions

R. Strzeżek, M. Koziorowska-Gilun, K. Filipowicz

Department of Animal Biochemistry and Biotechnology, Faculty of Animal Bioengineering, University of Warmia and Mazury in Olsztyn, 10-718 Olsztyn, Oczapowskiego 5, Poland

Abstract

The aim of this study was to determine the activity of proacrosin and acrosin in spermatozoa originating from the sperm-rich fractions (SRF) and whole ejaculates (WE) of dog semen. In addition, experiments were conducted to determine the activity of antitrypsin inhibitors in the fluids of different ejaculate fractions and whole seminal plasma. Ejaculates were collected from five dogs of mixed breed and one Beagle dog (aged from 2 to 9 years).

In the SRF, it was confirmed that the activity of the free acrosin form was predominant (acrosin / proacrosin; 2.38 ± 0.22 / 1.05 ± 0.08 mIU / 10^6 spermatozoa). On the other hand, spermatozoa originating from the WE exhibited significantly higher (p<0.05) proacrosin activity (proacrosin / acrosin; 2.19 ± 0.19 / 1.30 ± 0.11 mIU / 10^6 spermatozoa). Furthermore, acrosin inhibitor activity was lower in the fluids of the pre-sperm fraction (0.09 ± 0.006 IU / cm³), whereas it was higher in the fluids of the post-sperm fraction (0.11 ± 0.007 IU / cm³). Using PAGE analysis, the antitrypsin activity of the enzyme was represented by the presence of one electrophoretic band in the fluids of the pre-sperm and post-sperm fractions and whole seminal plasma. Furthermore, two electrophoretic bands were detected in the fluids of the SRF. The findings of this study indicate that specific proteinase inhibitors present in the individual ejaculate fractions of dog semen may act by stabilizing the sperm acrosin system.

Key words: canine, dog, semen, acrosin, proacrosin, antitrypsin inhibitors

Introduction

Acrosin (EC 3.4.21.10) is a trypsin-like endoprotease present in the acrosome of all mammalian spermatozoa (Urch et al 1991). The biological function of acrosin is to catalyze the hydrolytic reaction of splitting the sialprotein zona pellucida (Mann and Lutwak-Mann 1981). The presence of acrosin was detected in spermatozoal extracts of many animal species and humans (Anderson et al. 1981). This enzyme in ejaculated spermatozoa is found in the form of inactive zymogen (proenzyme), the so-called proacrosin, stabilized by natural specific inhibitors (Torska 1989). Acrosin inhibitors found in the seminal plasma are adsorbed on the outer surface of the acrosomal plasma membrane (Goodpasture et al. 1980). It was shown that inhibitors reverse the catalytic action of acrosin, released in case of premature activation of proacrosin, thus protecting acrosome proteins against intracellular degradation (Torska 1989). They form inactive bonds with acrosin and thus protect semen proteins against proteolytic effect of the enzyme liberated from damaged or dead spermatozoa (Fritz et al. 1975).