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Bacillus intermedius ribonuclease (BINASE) induces apoptosis in human ovarian cancer cells



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ABSTRACT

The cytotoxic effects of *Bacillus intermedius* RNase (binase) towards ovarian cancer cells (SKOV3 and OVCAR5) were studied in comparison to normal ovarian epithelial cells (HOSE1 and HOSE2). Binase decreased viability and induced the selective apoptosis of ovarian cancer cells. The apoptosis rate was 50% in SKOV3 and 48% in OVCAR5 cells after 24 h of binase treatment (50 µg/ml). Binase-induced apoptosis in these cell lines was accompanied by caspase-3 activation and poly(ADP-ribose) polymerase fragmentation. Normal ovarian epithelial cells were not affected by binase, except for a slight decrease of HOSE2 cell viability and the appearance of traces of activated caspase-3, but not the poly(ADP-ribose) polymerase 85-kDA fragment. Binase did not induce alteration of EZH2 (enhancer of zeste-homolog-2) protein expression neither, in tumor nor in normal cells. In conclusion, selective binase-induced cell death and apoptosis via poly(ADP-ribose) polymerase fragmentation may serve as a new treatment option against ovarian cancer progression.

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1. Introduction

Ovarian cancer affects approximately 1 in 70 women during their lifetime and is regarded as the most lethal gynecologic malignancy (Krylova et al., 2006). It takes 7th place in the list of age-standardized cancer mortality rates for female population in the world (Ferlay et al., 2013) and 5th place in the USA (Howlader et al., 2012). More than 90% of ovarian cancers are classified of epithelial origin. Various

targeted therapeutics – including monoclonal antibodies against growth factor receptors, inhibitors of tyrosine kinase, poly(ADP-ribose) polymerase (PARP) and angiokinas – have been explored for ovarian cancer therapy (Campos and Ghosh, 2010). A novel principle in anticancer therapy is based on destroying tumor cell RNA by exogenous cytotoxic ribonucleases (RNases) (Ardelt et al., 2003; Cabrera-Fuentes et al., 2012; Fischer et al., 2014, 2013; Leland and Raines, 2001; Libonati et al., 2008). Some cytotoxic RNases of bacterial nature, such as *Bacillus intermedius* – binase (Cabrera-Fuentes et al., 2013; Makarov and Ilinskaya, 2003; Makarov et al., 2008), *Bacillus amyloliquefaciens* – barnase (Edelweiss et al., 2008; Ulyanova et al., 2011) and *Streptomyces aureofaciens* RNase Sa3 (Sevcik et al., 2002), are not inhibited by mammalian ribonuclease inhibitor and may selectively induce apoptosis of different tumor cell lines.

Binase is a well-characterized, highly cationic RNase that hydrolyzes RNA molecules, predominantly after purine (guanine) residues, with formation of 3'-mono- and 3'-

Abbreviations: Binase, *Bacillus intermedius* RNase; EOC, Epithelial ovarian cancer; EZH2, Enhancer of zeste homolog 2 protein; PARP, Poly(ADP-ribose) polymerase; RNase, Ribonuclease.

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