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Supplemental Material to:

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Ribonuclease binase inhibits primary tumor growth and metastases via apoptosis induction in tumor cells

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Supplementary material

Determination of binase activity in blood serum. Cleavage of poly(I) (Sigma Aldrich) in blood serum samples was measured as described elsewhere (1). An aliquot of blood serum sample (2-8 mkl) was added to 2 ml of PBS buffer (pH 7.4), containing 0.1 mM poly(I). RNase activity was expressed in relative units. which show the increase of absorption at 248 nm in 1 min divided by the amount of blood serum (Fig. 1S). Binase concentration in samples was determined by comparing the RNase activity in the blood serum samples with the activity of binase in standard solution (Table 1S).

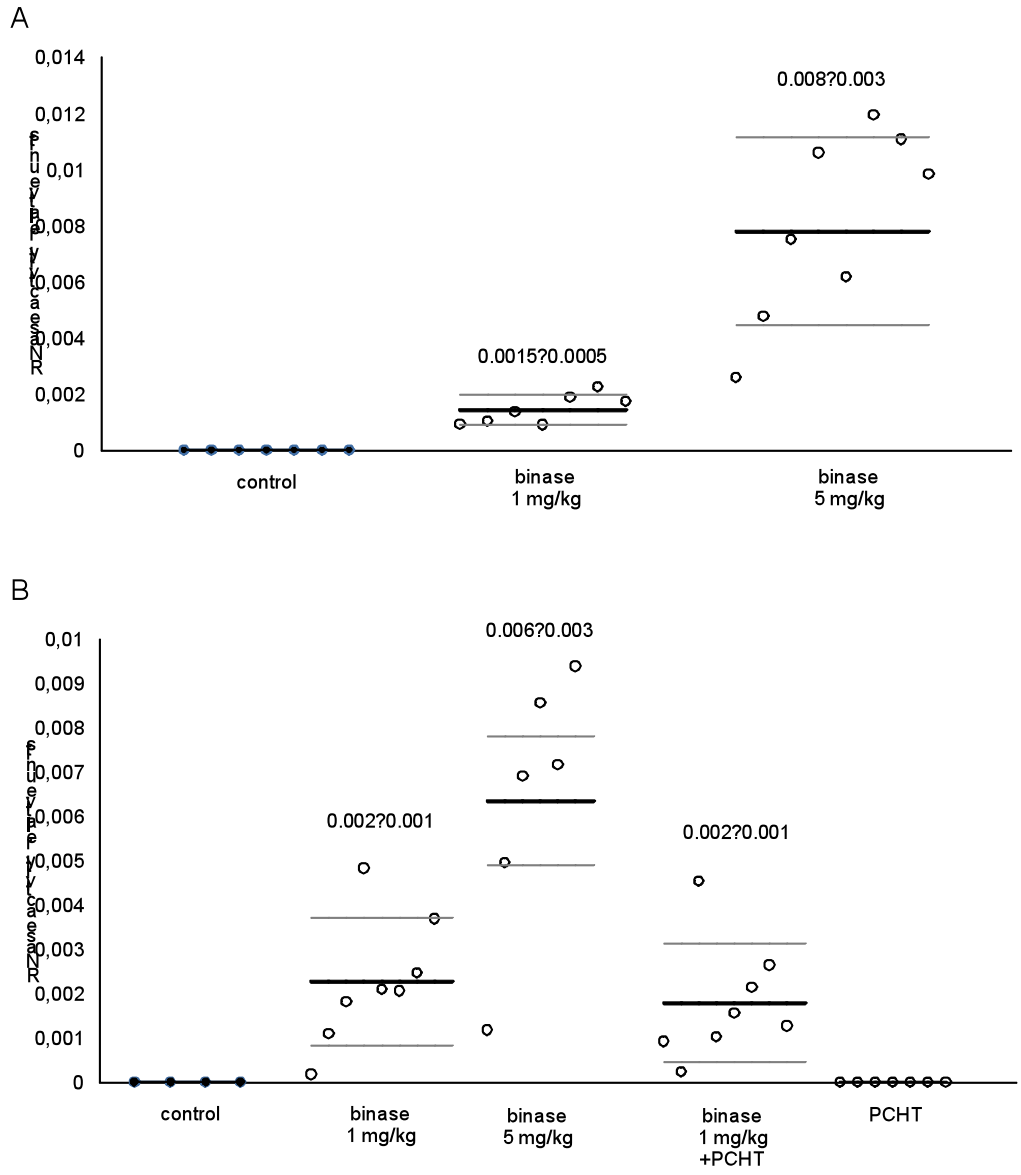

Figure 1S. Relative RNase activity in blood serum after 15 min post injection. (A) Mice with metastatic model of B16 and (B) RLS_{40}-bearing mice. For each group of animals the value of MEAN±SE is indicated.

Table 1S. Binase concentration in blood serum.

Sample	Binase concentration x10^8 M								
	RLS_{40}-bearing mice								Mean
Binase 1 mg/kg	0.80	4.87	8.12	21.59	9.32	9.20	11.01	16.47	10.17
Binase 5 mg/kg	5.25	22.13	30.86	38.17	31.95	41.88			28.37
Binase 1 mg/kg + PCHT	4.12	1.08	20.27	4.58	6.98	9.57	11.80	5.66	8.00
	Mice with metastatic model of B16								
Binase 1 mg/kg	4.23	4.70	6.13	4.06	8.50	10.15	7.82		6.51
Binase 1 mg/kg	11.60	21.33	33.50	47.30	27.56	53.38	49.49	43.98	36.02

References

1. Yakovlev GI, Moiseyev GP, Struminskaya NK, Borzykh OA, Kipenskaya LV, Znamenskaya LV et al. Mutational analysis of the active site of RNase of *Bacillus intermedius* (BINASE). FEBS Lett 1994; 354:305-6.