



Segment-specific targeting via RNA interference mediates down-regulation of OPN expression in hepatocellular carcinoma cells

F. Lin^{1*}, C.M. Huang^{2*}, J. Cao¹, Z.H. Pei¹, W.L. Gu¹, S.F. Fan¹, K.P. Li¹ and C.M. Lin¹

¹Department of General Surgery, Guangzhou First People's Hospital Affiliated to Guangzhou Medical University, Guangzhou, Guangdong, China

²Department of General Surgery, People's Hospital of Guangdong Province, Guangzhou, Guangdong, China

*These authors contributed equally to this study.

Corresponding author: J. Cao

E-mail: caojiecn@yeah.net

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ABSTRACT. Osteopontin (OPN) plays an important role in the metastasis and recurrence of tumors after resection of hepatocellular carcinoma (HCC). In this study, the down-regulation effect on OPN expression in HCC cells of RNA interference (RNAi) molecules designed to target different segments of OPN was investigated to identify a more effective site for OPN knockdown. Specific small interfering RNAs (siRNAs A, B, and C) of *OPN* were synthesized and transfected into an HCC cell line (HEP-G2; representing the OPNi-A, OPNi-B, and OPNi-C groups). Fluorescent quantitative polymerase chain reaction and immunohistochemical methods were used to detect the mRNA and protein expression of OPN before and after RNAi. Results showed that after transfection, the fluorescence intensity of the OPNi-A group was greater than those of the OPNi-B and OPNi-C groups. After 48 h of transfection, the Δ CT values of *OPN* mRNA expression in the OPNi-

A-C groups increased from 8.31 ± 1.58 , 8.78 ± 1.49 , and 8.25 ± 1.51 to 12.14 ± 1.43 , 10.22 ± 1.97 , and 10.48 ± 1.88 , respectively ($P < 0.05$), and the OPN protein levels (immunohistochemistry scores) decreased from 6.44 ± 1.67 , 5.43 ± 2.05 , and 5.45 ± 2.52 to 2.84 ± 1.52 , 4.43 ± 1.65 , and 3.95 ± 1.43 points, respectively. These results indicated that RNAi based on different segments of the *OPN* gene had different down-regulatory effects on OPN expression. Synthesis of targeted siRNA aimed at specific *OPN* segments might have important significance for dealing with the invasiveness and metastasis of HCC cells.

Key words: Osteopontin; Hepatocellular carcinoma; RNA interference; Down-regulation effect