Expression of Hsp47 in Oral Squamous Cell Carcinoma Cell Line, HSC-3 Noriko Okuyama, Junko Yusa and Hiroshi Ito

Heat shock protein 47 (Hsp47) participates in the synthesis of various types of collagen as a collagen-specific molecular chaperone. Hsp47 is expressed not only in the collagen-producing cells but also in normal and neoplastic keratinocytes, but the role of Hsp47 in keratinocytes is not clear. To resolve this problem, we examined the expression pattern of Hsp47 in human oral squamous cell carcinoma cell line, HSC-3, by Western blot and RT-PCR analyses at three culture stages, i.e., proliferating stage, confluent stage, and post-confluent stage. The mRNA levels of type IV collagen and laminin 5, major components of basement membrane, were analyzed at the same culture stages. To examine the intracellular spatial relationship of Hsp47 and type IV collagen expression, a double-immunofluorescence was performed at the proliferating stage. We also examined the expression pattern of other types of collagen by RT-PCR.

The Hsp47 expression increased in terms of protein and mRNA levels with the proliferation of cells and decreased after the confluence. The mRNA of type IV collagen showed almost the same expression pattern as that of Hsp47, implying a direct relationship between the two types of molecules. Unlike Hsp47 and type IV collagen, the mRNA level of laminin 5 at the proliferating stage was as high as at the confluent stage and decreased at the post-confluent stage. The decrease of these molecules may indicate the decreased production of basement membrane. The intensity of immunofluorescence staining of type IV collagen was detected in filopodia of the cells located at the periphery of the colonies, suggesting the production of basement membrane for cell migration. The co-expression of Hsp47 and type IV collagen visualized by the double-immunofluorescence was found in the perinuclear region, implying the bind of both molecules in endoplasmic reticulum.

The present results provide more definite evidence for the hypothesis that the expression of Hsp47 in the keratinocytes participates in the synthesis of type IV collagen which is used to produce basement membrane which enables cell migration. The decreased expression of Hsp47 and type IV collagen in HSC-3 cells at the post-confluent stage may facilitate the motility of neoplastic cells through the attenuation of cell-matrix contact.

Key words: Hsp47, type IV collagen, basement membrane, migration