論文 / 著書情報
Article / Book Information

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（博士課程）
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論 文 要 旨

THESIS SUMMARY

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要旨（英文 800 語程度）
Thesis Summary（approx.800 English Words ）

Tumor engineering emphasizes the importance of tumor tissue models in cancer disease research. Tissue engineering techniques have been used to recreate the tumor microenvironment and build tumor tissue models. Self-assembling peptides can form various nanostructures to provide biomaterials for 3D cell culture and drug delivery. The hydrogel is fabricated from self-assembled peptide nanofibers that can mimic the fibrous structures of natural ECM in water. Biocompatible hydrogels can be used for tumor engineering. Cancer cells cultured in biomimetic hydrogels can retain features of tumors formed in vivo. Therefore, tumor tissue models can be constructed in vitro using self-assembling peptide hydrogels.

In the previous study, $(FFiK)_2$ was designed as a self-assembling peptide that can assemble into networked nanofibers in water and form a hydrogel. However, $(FFiK)_2$ showed low cell adhesion and proliferation activity. In this study, $(FFiK)_2$ was functionalized by conjugation with various bioactive sequences derived from natural extracellular proteins to install cell adhesion ability and cell proliferation activity into $(FFiK)_2$. Functionalized $(FFiK)_2$ derivatives were used for 3D culture of breast cancer MCF-7 cells to construct breast tumor models. The constructed tumor model is useful for elucidating the behavior of cancer cells and for testing anticancer drugs.

Four bioactive sequences were selected in this study; RGDS, PHSRN derived from fibronectin and AG73, C16 derived from laminin. The peptide secondary structure was validated by circular dichroism and ATR-IR spectra measurements. All the $(FFiK)_2$ derivatives retained the typical β-sheet structure. Therefore, the introduction of bioactive sequences did not disturb the self-assembling ability of $(FFiK)_2$ derivatives. TEM observation revealed that $(FFiK)_2$ derivatives successfully formed networked peptide nanofibers in water by co-assembly with the parent $(FFiK)_2$. MCF-7 cells were cultured in a $(FFiK)_2$ and $(FFiK)_2$ derivative hydrogels. The cell adhesion test revealed that the introduction of bioactive sequences significantly promoted the adhesion of MCF-7 cells to functionalized hydrogels. MCF-7 cells were successfully encapsulated in $(FFiK)_2$ hydrogels and proliferated without significant cytotoxicity. MCF-7 cells proliferated with spheroid formation in hydrogels displaying RGDS, PHSRN, or C16 sequences, which mimics tumor spheroid formation in vivo. On the other hand, MCF-7 cells migrated into the hydrogel displaying the AG73 sequence. The AG73 sequence was able to bind to syndecan in breast cancer cells and promote tumor cell adhesion and invasion. Consequently, the spheroid model and metastatic model of MCF-7 were successfully developed using functionalized hydrogels of $(FFiK)_2$ derivatives with bioactive sequences. Two different breast cancer cell models can be used for testing of anticancer drugs.

Doxorubicin (DOX) was used as a typical anticancer drug to evaluate its cell-killing activity. 3D cultured MCF-7 cells demonstrated higher DOX resistance than those cultured on a 2D microplate. This suggests that the culture dimension is important for drug resistance of MCF-7 cells. The spheroid model exhibited higher DOX resistance than the migration model. The expression level of P-glycoprotein (P-gp), a drug efflux pump, was up-regulated in spheroid models of $(FFiK)_2$ and $(FFiK)_2$-RGDS hydrogels. These results suggest that spheroid formation is important for high drug resistance. 3D culture of MCF-7 cells in functionalized peptide hydrogels can produce both the spheroid model resistant to DOX and the metastatic model sensitive to DOX. These findings are useful for elucidating the behavior of MCF-7 cells and for screening anticancer drugs using tumor tissue models.

In conclusion, $(FFiK)_2$ derivatives successfully improved the bioactivities of $(FFiK)_2$ through functionalization with bioactive sequences derived from ECM proteins. The co-assembly method produced hydrogels displaying bioactive sequences and promoted cell proliferation or migration of MCF-7 cells. Cell behaviors such as spheroid formation or migration could be regulated using proper bioactive sequences. The spheroid tumor model that is resistant to drugs and the metastatic model that is sensitive to drugs developed in this study are helpful for developing tailored tissue models that will be important for tumor tissue engineering in the future.

備考 ： 論文要旨は、和文 2000 字と英文 300 語を 1 部ずつ提出するか、もしくは英文 800 語を 1 部提出してください。
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