Application of Monoclonal Antibodies against Naturally Occurring Bioactive Ingredients

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Abstract

Monoclonal antibodies (Mabs) are widely used in a variety of fields, including protein identification, life sciences, medicine, and natural product chemistry. This review focuses on Mabs against naturally occurring active compounds. The preparation of Mabs against various active compounds began in the 1980s, and now there are fewer than 50 types. Eastern blotting, which was developed as an antibody staining method for low-molecular-weight compounds, is useful for its ability to visually represent specific components. In this method, a mixture of lower-molecular-weight compounds, particularly glycosides, are separated by thin-layer chromatography (TLC). The compounds are then transferred to a membrane by heating, followed by treatment with potassium periodate (KIO_4) to open the sugar moiety of the glycoside on the membrane to form an aldehyde group. Proteins are then added to form Schiff base bonds to enable adsorption on the membrane. A Mab is bound to the glycoside moiety on the membrane and reacts with a secondary antibody to produce color. Double Eastern blotting, which enables the simultaneous coloration of two glycosides, can be used to evaluate quality and estimate pharmacological effects. An example of staining by Eastern blotting and a component search based on the results will also be presented. A Mab-associated affinity column is a method for isolating antigen molecules in a single step. However, the usefulness of the wash fractions that are not bound to the affinity column is unknown. Therefore, we designated the wash fraction the “knockout extract”. Comparing the nitric oxide (NO) production of a glycyrrhizin (GL)-knockout extract of licorice with a licorice extract revealed that the licorice extract is stronger. Therefore, the addition of GL to the GL-knockout extract of licorice increased NO production. This indicates that

GL has synergic activity with the knockout extract. The GL-knockout extract of licorice inhibited high-glucose-induced epithelial–mesenchymal transition in NRK-52E cells, primarily by suppressing the Notch2 pathway. The real active constituent in licorice may be constituents other than GL, which is the causative agent of pseudohyperaldosteronism. This suggests that a GL-knockout extract of licorice may be useful for the treatment of diabetic nephritis.

Keywords:

monoclonal antibody; eastern blotting; knockout extractives; pharmacological activity; Kampo medicine