Citation List

[Patent Documents]

[Patent Document 1] US20100331527

[Patent Document 2] US20130018174

Summary

[Problems to be Solved by the Invention]

An objective of the present invention is to provide methods for efficiently purifying an Fc region-containing polypeptide, in particular a bispecific antibody, using a Protein A resin column.

[Means for Solving the Problems]

As a result of dedicated research to solve the above-mentioned problems, the present inventors discovered that, by preparing an Fc region whose first polypeptide chain and second polypeptide chain have binding activities to a Protein A resin that are different from each other, the dynamic binding capacity of the antibody is increased, and the antibody purification efficiency is increased, and completed the present disclosure.

In one aspect, the present disclosure refers to a method for producing and purifying a heterologous Fc region-containing polypeptide, which comprises the steps of: (a) preparing a first polypeptide chain and a second polypeptide chain of an Fc region having binding activities to a Protein A resin that are different from each other; (b) comparing the dynamic binding capacity of a heterologous Fc region-containing polypeptide comprising the first polypeptide chain and the second polypeptide chain prepared in step (a) for the Protein A resin in Protein A column chromatography with the dynamic binding capacity of a homologous Fc region-containing polypeptide comprising two polypeptide chains having substantially the same binding activity to said resin for the Protein A resin in Protein A column chromatography to confirm the elevated dynamic binding capacity of the former Fc region-containing polypeptide compared with that of the latter Fc region-containing polypeptide; (c) contacting a sample comprising the heterologous Fc region-containing polypeptide with said resin; and (d) collecting the heterologous Fc region-containing polypeptide bound to the resin.

Specifically, the present disclosure provides the following:

[1] a method for increasing the dynamic binding capacity of an Fc region-containing polypeptide for a Protein A resin in Protein A column chromatography;

CLAIMS

1. A method for producing and purifying a heterologous Fc region-containing polypeptide, which comprises the steps of:

(a) preparing a first polypeptide chain and a second polypeptide chain of an Fc region having binding activities to a Protein A resin that are different from each other;

(b) comparing the dynamic binding capacity of a heterologous Fc region-containing polypeptide comprising the first polypeptide chain and the second polypeptide chain prepared in step (a) for the Protein A resin in Protein A column chromatography with the dynamic binding capacity of a homologous Fc region-containing polypeptide comprising two polypeptide chains having substantially the same binding activity to said resin for the Protein A resin in Protein A column chromatography to confirm the elevated dynamic binding capacity of the former Fc region-containing polypeptide compared with that of the latter Fc region-containing polypeptide;

(c) contacting a sample comprising the heterologous Fc region-containing polypeptide with said resin; and

(d) collecting the heterologous Fc region-containing polypeptide bound to the resin.

2. The method of claim 1, wherein said step (a) is preparing a first polypeptide chain that binds to the resin as the first polypeptide chain of the Fc region, and preparing a second polypeptide chain that does not bind to the resin or shows weaker binding to the resin compared to the first polypeptide chain, as the second polypeptide chain of the Fc region.

3. The method of claim 1 or 2, wherein said step (a) is modifying the Fc region of the Fc region-containing polypeptide for purification, so that the first polypeptide chain of the Fc region binds to the resin, and the second polypeptide chain of the Fc region does not bind to the resin or shows weaker binding to the resin compared to the first polypeptide chain.