CLAIMS

What is claimed is:

1. A method of adjusting the pH of a cell culture medium, comprising:

obtaining, for the cell culture medium, a functional relationship between a concentration of dissolved carbon dioxide in the cell culture medium and a mole fraction of gaseous carbon dioxide applied to the cell culture medium, and a concentration of net medium acids in the cell culture medium;

adding carbonate salt or bicarbonate salt to the cell culture medium to obtain a desired carbonate salt or bicarbonate salt concentration in the cell culture medium; and

determining, using a charge balance model, an amount of strong acid or strong base to be added to the cell culture medium to adjust the pH of the cell culture medium to a desired pH, wherein the charge balance model is based on at least the functional relationship between the concentration of dissolved carbon dioxide in the cell culture medium and the mole fraction of gaseous carbon dioxide applied to the cell culture medium, the concentration of net medium acids in the cell culture medium, the desired carbonate salt or bicarbonate salt concentration in the cell culture medium, and the desired pH.

2. The method of claim 1, further comprising adding the determined amount of strong acid or strong base to the cell culture medium, thereby making a pH-adjusted cell culture medium.

3. The method of claim 1 or 2, wherein the carbonate salt or the bicarbonate salt is sodium carbonate or sodium bicarbonate.

4. The method of any one of claims 1-3, further comprising supplementing the cell culture medium with one or more ionic compounds, wherein the charge balance model is further based on the concentration of the one or more ionic compounds.

5. The method of claim 4, wherein the one or more ionic compounds comprises one or more amino acids or ammonium chloride.

6. The method of claim 5, wherein the one or more amino acids comprises glutamine, asparagine, or glutamic acid.

7. The method of any one of claims 1-6, wherein the strong base is sodium hydroxide.

8. The method of any one of claims 1-7, wherein the strong acid is hydrochloric acid.

9. The method of any one of claims 1-8, wherein the charge balance model is defined by:

$$[Na^+] - [HCO_3^-] - 2 * [CO_3^{2-}] + [H^+] - [OH^-] - [NMA^-] - [A^-] + [B^+] = 0$$

wherein:

[Na^+] is a concentration of sodium ions added as sodium hydroxide, sodium bicarbonate, or sodium carbonate to the cell culture medium;

[H^+] is a concentration of protons in the cell culture medium needed to obtain the desired pH;

[OH^-] is a concentration of hydroxide anions in the cell culture medium;

[NMA^-] is the concentration of net medium acid ions in the cell culture medium;

[A^-] is a concentration of negatively charged ions added to the cell culture medium, multiplied by the absolute value of their charge, excluding any OH^- or negatively charged ions included in [NMA-]; and

[B^+] is a concentration of positively charged ions added to the cell culture medium, multiplied by the absolute value of their charge, excluding any H^+, sodium ions included in [Na^+], or positively charged ions included in [NMA-].

10. The method of claim 9, wherein:

$$[HCO_3^-] = \frac{K_0 * K_1 * P * (yCO_2)^m}{100 * K_H * [H^+]}$$

$$[CO_3^{2-}] = \frac{K_0 * K_1 * K_2 * P * (yCO_2)^m}{100 * K_H * [H^+]^2}$$

wherein:

K_0, K_1, and K_2 are dissociation constants for bicarbonate and carbonate anions;

P is a gas pressure applied to the cell culture medium;

yCO_2 is a molar percentage of CO_2 gas phase applied to the cell culture medium; and

m and K_H are each empirically determined parameters for the cell culture medium.

11. The method of any one of claims 1-10, wherein the concentration of net medium acids in the cell culture medium is modeled in the charge balance model as a function of pH of the cell culture medium.

12. The method of claim 11, wherein the concentration of net medium acids in the cell culture medium is modeled as:

$$[NMA^-] = [C_{0p} + C_{1p} * (pH - 7)]$$

wherein:

$[NMA^-]$ is the concentration of net medium acid ions in the cell culture medium; and

C_{0p} and C_{1p} are each empirically determined constants for the cell culture medium.

13. The method of any one of claims 1-10, wherein the concentration of net medium acids in the cell culture medium is modeled in the charge balance model as a function of temperature.

14. The method of any one of claims 1-10, wherein the concentration of net medium acids in the cell culture medium is modeled in the charge balance model as a function of pH and temperature.

15. A method of culturing cells, comprising:

adjusting the pH of a cell culture medium according to the method of any one of claims 1-14; and

culturing cells in the pH-adjusted cell culture medium.

16. The method of claim 15, wherein the cells are mammalian cells.

17. The method of claim 15 or 16, wherein cells are Chinese hamster ovary (CHO) cells.

18. A system, comprising:

one or more processors; and

a memory communicatively coupled to the one or more processors and configured to store instructions that, when executed by the one or more processors, cause the system to:

receive, at the one or more processors, for a cell culture medium, one or more parameters indicating a functional relationship between a concentration of dissolved carbon dioxide in the cell culture medium and a mole fraction of gaseous carbon dioxide applied to the cell culture medium, and a net medium acids parameter indicating a concentration of net medium acids in the cell culture medium;

receive, at the one or more processors, a carbonate salt or bicarbonate salt parameter indicating a desired carbonate salt or bicarbonate salt concentration in the cell culture medium;

receive, at the one or more processors, a pH parameter indicating a desired pH of the cell culture medium; and

determine, using a charge balance model, an amount of strong acid or strong base to be added to the cell culture medium to adjust the pH of the cell culture medium to the desired pH, wherein the charge balance model is based on at least the functional relationship between the concentration of dissolved carbon dioxide in the cell culture medium and the mole fraction of gaseous carbon dioxide applied to the cell culture medium, the concentration of net medium acids in the cell culture medium, the desired carbonate salt or bicarbonate salt concentration in the cell culture medium, and the desired pH.

19. A method for producing a polypeptide in a host cell expressing the polypeptide, comprising culturing the host cell in a cell culture medium by preparing a cell culture medium with sodium bicarbonate to tightly control the pH of the medium, comprising:

determining the excipients and relative amounts to be added to a cell culture medium to define a recipe,

preparing a solution using the recipe and determining the pH of the solution to define a first data set;

placing the solution in a CO_2 gassed and agitated bioreactor and allowing it to equilibrate to determine the resulting pH and pCO2 values to define a second data set;

placing the solution in an incubator at a defined temperature and molar percent CO_2 and determining the pH and pCO_2 measurements to define a third data set;

using the first, second, and third data sets and a pH model according to:

$$\mathrm{pH} - \mathrm{pK_a} - \log[\mathrm{B}] = \log\left(\frac{1}{\mathrm{s}}\right) - \mathrm{m} * \log\left(\mathrm{y_{CO_2}}\right)$$

to solve for the parameter values of m, s, and net medium acids simultaneously by minimizing:

$$\sum(\mathrm{k} * [\mathrm{C^k}]) = 0;$$

defining a target pH for the cell culture medium and adding an appropriate concentration of base to the cell culture medium as determined from the pH model to achieve the pH equivalence wherein the cell culture medium pH is tightly controlled; and

producing the polypeptide.

20. The method of claim 19, wherein the excipients are selected from the group consisting of glutamine, glutamate, asparagine, ammonium chloride, sodium chloride, and sodium hydroxide.