CLAIMS

1. Use of

i) a multispecific protein comprising a CD123 binding domain and a CD3 binding domain; and

ii) a second anti-cancer agent

in the manufacture of a medicament for treating a cancer in a subject in need thereof.

2. The use of claim 1, wherein a third anti-cancer agent is to be administered to the subject.

3. The use of claim 1 or 2, wherein the second anti-cancer agent is a chemotherapeutic drug,

and optionally wherein the chemotherapeutic drug is venetoclax, azacitidine, decitabine, daunorubicin, cytarabine, idarubicin, mitoxantrone, or etoposide.

4. The use of claim 1, wherein the second anti-cancer agent is cytarabine,

and wherein the cytarabine is to be administered at a dose of about 1 g/m^2.

5. The use of claim 2, wherein the second anti-cancer agent is mitoxantrone, and the third anti-cancer agent is etoposide, and wherein a fourth anti-cancer agent that is cytarabine is to be administered,

and optionally wherein the mitoxantrone is to be administered at a dose of about 6 mg/m^2/day, the etoposide is to be administered at a dose of about 80 mg/m^2/day, and the cytarabine is to be administered at a dose of about 1 g/m^2/day.

6. The use of claim 1, wherein the second anti-cancer agent is venetoclax,

and wherein the venetoclax is to be administered at a dose of from about 100 to about 400 mg per day.

7. The use of claim 1, wherein the second anti-cancer agent is azacytidine,

and wherein the azacytidine is to be administered at a dose of about 75 mg/m^2/day.

8. The use of claim 2, wherein the second anti-cancer agent is azacitidine and the third anti-cancer agent is venetoclax,

and optionally wherein the azacitidine is to be administered at a dose of about 75 mg/m^2/day and the venetoclax is to be administered at a dose of from about 100 to about 400 mg/day.

9. The use of claim 1, wherein the second anti-cancer agent is decitabine,

and wherein the decitabine is to be administered at a dose of about 20 mg/m^2/day.

10. The use of claim 2, wherein the second anti-cancer agent is decitabine and the third anti-cancer agent is venetoclax,

and optionally wherein the decitabine is to be administered at a dose of about 20 mg/m^2/day and the venetoclax is to be administered at a dose of from about 100 to about 400 mg/day.

11. The use of claim 2, wherein the second anti-cancer agent is daunorubicin, and the third anti-cancer agent is cytarabine,

and optionally wherein the daunorubicin is to be administered at a dose of about 30-90 mg/m^2 and the cytarabine is to be administered at a dose of from about 100 to about 200 mg/m^2.

12. The use of claim 1, wherein the anti-cancer agent is idarubicin,

and wherein the idarubicin is to be administered at a dose of about 12 mg/m^2.

13. The use of claim 2, wherein the second anti-cancer agent is idarubicin and the third anti-cancer agent is cytarabine,

and optionally wherein the idarubicin is to be administered at a dose of about 12 mg/m^2, and cytarabine is to be administered at a dose of from about 100 to about 200 mg/m^2.

14. The use of claim 1, wherein the second anti-cancer agent is azacitidine,

and wherein the azacitidine is to be administered at a dose of about 75 mg/m^2/day.

15. The use of any one of claims 1-14, wherein the multispecific protein comprises: a dimer of two identical polypeptides, wherein each polypeptide comprises, in order from amino-terminus to carboxyl-terminus, or in order from carboxyl-terminus to amino-terminus:

(i) a CD123 binding domain,

(ii) a hinge region,

(iii) an immunoglobulin constant region, and

(iv) a CD3 binding domain.

16. The use of claim 15, wherein the polypeptide comprises, from N-terminus to C-terminus, the CD123 binding domain, the hinge region, the immunoglobulin constant region, and the CD3 binding domain.

17. The use of claim 16, wherein at least one of the CD123 and the CD3 binding domains comprises:

(i) an immunoglobulin heavy chain variable region (VH) comprising HCDR1, HCDR2, and HCDR3; and

(ii) an immunoglobulin light chain variable region (VL) comprising LCDR1, LCDR2, and LCDR3.

18. The use of claim 17,

wherein the CD123 binding domain is a scFv comprising:

a HCDR1 that comprises SEQ ID NO: 10, a HCDR2 that comprises SEQ ID NO: 11, and a HDCR3 that comprises SEQ ID NO: 12; and

a LCDR1 that comprises SEQ ID NO: 13, a LCDR2 that comprises SEQ ID NO: 14, and a LCDR3 that comprises SEQ ID NO: 15;

or

wherein the CD123 binding domain is a scFv comprising:

a VH comprising a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 136, and

a VL comprising a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 134;

or

wherein the CD123 binding domain is a scFv, and wherein the scFv comprises a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 27.

19. The use of claim 17 or 18,

wherein the CD3 binding domain is a scFv comprising:

a HCDR1 that comprises SEQ ID NO: 19, a HCDR2 that comprises SEQ ID NO: 20, and a HDCR3 that comprises SEQ ID NO: 21; and

a LCDR1 that comprises SEQ ID NO: 22, a LCDR2 that comprises SEQ ID NO: 23, and a LCDR3 that comprises SEQ ID NO: 24;

or

wherein the CD3 binding domain is a scFv that comprises:

a VH comprising a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 383 or 387, and

a VL comprising a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 384;

or

wherein the CD3 binding domain is a scFv that comprises a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 27.

20. The use of claim 17, wherein each polypeptide comprises a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 31.

21. The use of any one of claims 1-20, wherein the multispecific protein is to be administered to the subject by IV infusion.

22. The use of any one of claims 1-21, wherein the second anti-cancer agent is to be administered to the subject orally or by IV infusion.

23. The use of any one of claims 1-22, wherein the multispecific protein is to be administered the subject by IV infusion at a dose of 0.3, 1, 3, 6, 9, 12, 18, 20, 24, 30, 36, 48, 50, 60, 75, or 100 μg.

24. The use of any one of claims 1-23, wherein the multispecific protein is to be administered once per week.

25. The use of claim 24, wherein the multispecific protein is to be administered to the subject by IV infusion during a first 28-day cycle; wherein 6 μg of the multispecific protein is to be administered on day 8; 12 μg of the multispecific protein is to be administered on day 15; and 18 μg of the multispecific protein is to be administered on day 22 of the first 28-day cycle,

and optionally wherein the multispecific protein is to be administered to the subject by IV infusion during at least one additional 28-day cycle following the first 28-day cycle, wherein 18 μg of the multispecific protein is to be administered on days 1, 8, 15, and 22 of the at least one additional 28-day cycle.

26. The use of claim 25, wherein cytarabine is to be administered intravenously on days 1-5 of the first 28-day cycle, and days 1-5 of at least one additional 28-day cycle,

and optionally wherein the dose of cytarabine is about 1 g/m^2.

27. The use of claim 25, wherein mitoxantrone, etoposide, and cytarabine are to be administered intravenously on days 1-6 of the first 28-day cycle, and at least one additional 28-day cycle,

and optionally wherein the dose of mitoxantrone is about 6 mg/m^2/day, the dose of etoposide is about 80 mg/m^2/day, and the dose of cytarabine is about 1 g/m^2/day.

28. The use of claim 24, wherein the multispecific protein is to be administered to the subject by IV infusion during a first 28-day cycle; wherein 6 μg of the multispecific protein is to be administered on day 15; and 12 μg of the multispecific protein is to be administered on day 22 of the first 28-day cycle,

optionally wherein the multispecific protein is to be administered to the subject by IV infusion during at least one additional 28-day cycle following the first 28-day cycle, wherein 18 μg of the multispecific protein is to be administered on days 1, 8, 15, and 22 of the at least one additional 28-day cycle,

optionally wherein venetoclax is to be administered orally on days 1-21 of the first 28-day cycle, and days 1-21 of at least one additional 28-day cycle,

optionally wherein the dose of venetoclax is from about 100 to about 400 mg/day,

optionally wherein azacytidine is to be administered intravenously on days 1-7 of the first 28-day cycle, and at least one additional 28-day cycle,

and optionally wherein the dose of azacytidine is about 75 mg/m^2.

29. The use of claim 24, wherein the multispecific protein is to be administered to the subject by IV infusion during a first 28-day cycle, wherein 6 μg of the multispecific protein is to be administered on day 1; 8 μg of the multispecific protein is to be administered on day 12; 18 μg of the multispecific protein is to be administered on day 15; and 18 μg of the multispecific protein is to be administered on day 22 of the first 28-day cycle,

optionally wherein the multispecific protein is to be administered to the subject by IV infusion during at least one additional 28-day cycle following the first 28- day cycle, wherein 18 μg of the multispecific protein is to be administered on days 1, 8, 15, and 22,

optionally wherein cytarabine is to be administered by intravenous infusion on days 1-7 of the first 28-day cycle, and days 1-7 of at least one additional 28-day cycle,

and optionally wherein the dose of cytarabine is from about 100 to about 200 mg/m^2.

30. The use of claim 29, wherein idarubicin is to be administered by intravenous infusion on days 1-3 of the first 28-day cycle, and days 1-3 of at least one additional 28-day cycle,

and optionally wherein the dose of idarubicin is about 12 mg/m^2.

31. The use of claim 24, wherein the multispecific protein is to be administered to the subject by IV infusion during a first 28-day cycle, wherein 6 μg of the multispecific protein is to be administered on day 1; 8 μg of the multispecific protein is to be administered on day 12; 18 μg of the multispecific protein is to be administered on day 15; and 18 μg of the multispecific protein is to be administered on day 22 of the first 28-day cycle,

optionally wherein the multispecific protein is to be administered to the subject by IV infusion during at least one additional 28-day cycle following the first 28-day cycle, wherein 18 μg of the multispecific protein is to be administered on days 1, 8, 15, and 22 of the at least one additional 28-day cycle,

optionally wherein azacytidine is to be administered orally on days 1-14 of the first 28-day cycle, and at least one additional 28-day cycle,

and optionally wherein the dose of azacytidine is about 300 mg/day.

32. The use of any one of claims 1-31,

wherein the cancer is selected from the group consisting of carcinoma, sarcoma, melanoma, kidney cancer, pancreatic cancer, lung cancer, intestinal cancer, prostate cancer, breast cancer, liver cancer, brain cancer, colon cancer, ovarian cancer, hematological cancer, acute myeloid leukemia (AML), myelodysplastic syndrome (MDS), hairy cell leukemia (HCL),

blastic plasmacytoid dendritic cell neoplasm, B-cell acute lymphoblastic leukemia (ALL), or chronic myeloid leukemia (CML).

33. Use of a multispecific protein comprising a CD123 binding domain and a CD3 binding domain in the manufacture of a medicament for treating a cancer in a subject in need thereof.

34. The use of claim 33, wherein the multispecific protein comprises: a dimer of two identical polypeptides, wherein each polypeptide comprises, in order from amino-terminus to carboxyl-terminus, or in order from carboxyl-terminus to amino-terminus:

(i) a CD123 binding domain,

(ii) a hinge region,

(iii) an immunoglobulin constant region, and

(iv) a CD3 binding domain.

35. The use of claim 34,

wherein the polypeptide comprises, from N-terminus to C-terminus, the CD123 binding domain, the hinge region, the immunoglobulin constant region, and the CD3 binding domain;
or

wherein at least one of the CD123 and the CD3 binding domains comprises:

(i) an immunoglobulin heavy chain variable region (VH) comprising HCDR1, HCDR2, and HCDR3; and

(ii) an immunoglobulin light chain variable region (VL) comprising LCDR1, LCDR2, and LCDR3;
or

wherein the CD123 binding domain is a scFv comprising:

a HCDR1 that comprises SEQ ID NO: 10, a HCDR2 that comprises SEQ ID NO: 11, and a HDCR3 that comprises SEQ ID NO: 12; and

a LCDR1 that comprises SEQ ID NO: 13, a LCDR2 that comprises SEQ ID NO: 14, and a LCDR3 that comprises SEQ ID NO: 15;
or

wherein the CD123 binding domain is a scFv comprising:

a VH comprising a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 136, and

a VL comprising a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 134;

or

wherein the CD123 binding domain is a scFv, and wherein the scFv comprises a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 27.

36. The use of claim 34 or 35, wherein the CD3 binding domain is a scFv comprising:

a HCDR1 that comprises SEQ ID NO: 19, a HCDR2 that comprises SEQ ID NO: 20, and a HDCR3 that comprises SEQ ID NO: 21; and

a LCDR1 that comprises SEQ ID NO: 22, a LCDR2 that comprises SEQ ID NO: 23, and a LCDR3 that comprises SEQ ID NO: 24;

or

wherein the CD3 binding domain is a scFv that comprises:

a VH comprising a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 383 or 387, and

a VL comprising a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 384;

or

wherein the CD3 binding domain is a scFv that comprises a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 27.

37. The use of claim 34, wherein each polypeptide comprises a sequence at least 90%, at least 95%, or 100% identical to SEQ ID NO: 31.

38. The use of any one of claims 33-37, wherein the multispecific protein is to be administered to the subject by IV infusion.

39. The use of any one of claims 33-38,

wherein the multispecific protein is to be administered the subject by IV infusion at a dose of 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, or 0.8 μg/kg;

or

wherein the multispecific protein is to be administered the subject by IV infusion at a dose of 0.3, 1, 3, 6, 9, 12, 18, 20, 24, 30, 36, 48, 50, 60, 75, or 100 μg.

40. The use of any one of claims 33-39, wherein the multispecific protein is to be administered once per week.

41. The use of claim 40, wherein the multispecific protein is to be administered to the subject by IV infusion during a first 28-day cycle; wherein 6 μg of the multispecific protein is to be administered on day 8; 12 μg of the multispecific protein is to be administered on day 15; and 18 μg of the multispecific protein is to be administered on day 22 of the first 28-day cycle,

and optionally wherein the multispecific protein is to be administered to the subject by IV infusion during at least one additional 28-day cycle following the first 28-day cycle, wherein 18 μg of the multispecific protein is to be administered on days 1, 8, 15, and 22 of the at least one additional 28-day cycle.

42. The use of claim 40, wherein the multispecific protein is to be administered to the subject by IV infusion during a first 28-day cycle; wherein 6 μg of the multispecific protein is to be administered on day 15; and 12 μg of the multispecific protein is to be administered on day 22 of the first 28-day cycle,

and optionally wherein the multispecific protein is to be administered to the subject by IV infusion during at least one additional 28-day cycle following the first 28-day cycle, wherein 18 μg of the multispecific protein is to be administered on days 1, 8, 15, and 22 of the at least one additional 28-day cycle.

43. The use of claim 40, wherein the multispecific protein is to be administered to the subject by IV infusion during a first 28-day cycle, wherein 6 μg of the multispecific protein is to be administered on day 1; 8 μg of the multispecific protein is to be administered on day 12; 18 μg of the multispecific protein is to be administered on day 15; and 18 μg of the multispecific protein is to be administered on day 22 of the first 28-day cycle,

and optionally wherein the multispecific protein is to be administered to the subject by IV infusion during at least one additional 28-day cycle following the first 28-day cycle, wherein 18 μg of the multispecific protein is to be administered on days 1, 8, 15, and 22.

44. The use of claim 40, wherein the multispecific protein is to be administered to the subject by IV infusion during a first 28-day cycle, wherein 6 μg of the multispecific protein is to be administered on day 1; 8 μg of the multispecific protein is to be administered on day 12; 18 μg

of the multispecific protein is to be administered on day 15; and 18 μg of the multispecific protein is to be administered on day 22 of the first 28-day cycle,

and optionally wherein the multispecific protein is to be administered to the subject by IV infusion during at least one additional 28-day cycle following the first 28-day cycle, wherein 18 μg of the multispecific protein is to be administered on days 1, 8, 15, and 22 of the at least one additional 28-day cycle.

45. The use of any one of claims 33-44,

wherein the cancer selected from the group consisting of carcinoma, sarcoma, melanoma, kidney cancer, pancreatic cancer, lung cancer, intestinal cancer, prostate cancer, breast cancer, liver cancer, brain cancer, colon cancer, ovarian cancer, hematological cancer,

acute myeloid leukemia (AML), myelodysplastic syndrome (MDS), hairy cell leukemia (HCL), blastic plasmacytoid dendritic cell neoplasm, B-cell acute lymphoblastic leukemia (ALL), or chronic myeloid leukemia (CML).