continues to be a major challenge for tumor immunologists. Thus, there is a high need for an effective and well-tolerated immunotherapy for the treatment of cancer.

SUMMARY

The present disclosure provides a novel combinatorial immunotherapeutic approach for the treatment of cancer.

In a first aspect, the present disclosure provides for a kit-of-parts comprising a. a polyplex comprising a double stranded RNA (dsRNA) and a polymeric conjugate, wherein said dsRNA is polyinosinic-polycytidylic acid double stranded RNA (polyIC), wherein said polymeric conjugate comprises a polyethyleneimine (PEI), one or more polyethylene glycol (PEG) moieties and one or more targeting moieties, wherein said PEI is covalently bound to one or more PEG moieties, and each of said one or more PEG moieties is linked to one of said one or more targeting moieties, and wherein each of said one or more targeting moieties is capable of binding to a cancer antigen; and b. at least one antibody, wherein said at least one antibody is capable of modulating an immune checkpoint protein, and wherein said at least one antibody capable of modulating an immune checkpoint protein is (i) at least one antibody capable of agonizing a co-stimulatory immune checkpoint protein, or (ii) at least one antibody capable of antagonizing a co-inhibitory immune checkpoint protein; or (iii) a mixture of at least one antibody of (i) and at least one antibody of (ii); and wherein said immune checkpoint protein is selected from the group consisting of 4-1BB, 4-1BB ligand, PD-1, PD-L1, PD-L2 and CTLA-4.

In a further aspect, the present disclosure provides for a composition comprising a. a polyplex comprising a double stranded RNA (dsRNA) and a polymeric conjugate, wherein said dsRNA is polyinosinic-polycytidylic acid double stranded RNA (polyIC), wherein said polymeric conjugate comprises a polyethyleneimine (PEI), one or more polyethylene glycol (PEG) moieties and one or more targeting moieties, wherein said PEI is covalently bound to one or more PEG moieties, and each of said one or more PEG moieties is linked to one of said one or more targeting moieties, and wherein each of said one or more targeting moieties is capable of binding to a cancer antigen; and b. at least one antibody, wherein said at least one antibody is capable of modulating an immune checkpoint protein, and wherein said at least one antibody capable of modulating an immune checkpoint protein is (i) at least one antibody capable of agonizing a co-stimulatory immune checkpoint protein, or (ii) at least one antibody capable of antagonizing a co-inhibitory immune checkpoint protein; or (iii) a mixture of at

least one antibody of (i) and at least one antibody of (ii); and wherein said immune checkpoint protein is selected from the group consisting of 4-1BB, 4-1BB ligand, PD-1, PD-L1, PD-L2 and CTLA-4.

In a first ~~aspect~~example, the present disclosure provides for a kit-of-parts comprising

a. a polyplex comprising a double stranded RNA (dsRNA) and a polymeric conjugate, wherein said polymeric conjugate comprises a polyethyleneimine (PEI), one or more polyethylene glycol (PEG) moieties and one or more targeting moieties, wherein said PEI is covalently bound to one or more PEG moieties, and each of said one or more PEG moieties is linked to one of said one or more targeting moieties, and wherein each of said one or more targeting moieties is capable of binding to a cancer antigen; and

b. at least one antibody, wherein said at least one antibody is capable of modulating an immune checkpoint protein.

In a further example~~aspect~~, the present disclosure provides for a composition comprising

a. a polyplex comprising a double stranded RNA (dsRNA) and a polymeric conjugate, wherein said polymeric conjugate comprises a polyethyleneimine (PEI), one or more polyethylene glycol (PEG) moieties and one or more targeting moieties, wherein said PEI is covalently bound to one or more PEG moieties, and each of said one or more PEG moieties is linked to one of said one or more targeting moieties, and wherein each of said one or more targeting moieties is capable of binding to a cancer antigen; and

b. at least one antibody, wherein said at least one antibody is capable of modulating an immune checkpoint protein.

In a further example, the present disclosure provides for the composition or the kit-of-parts according to the disclosure for use in the treatment of cancer in a mammal. In a further aspect, the present disclosure provides for the composition or the kit-of-parts according to the disclosure for use in therapy. In a further aspect, the present disclosure provides for the use of the composition or the kit-of-parts according to the disclosure in the manufacture of a medicament for treating cancer in a mammal~~, preferably a human~~.

The inventors surprisingly found that combination treatment with the polyplex comprising double stranded RNA (dsRNA) and the polymeric conjugate and one or more anti-checkpoint antibodies according to the disclosure enforces activity of the immune system

CLAIMS

1. A kit-of-parts comprising

a. a polyplex comprising a double stranded RNA (dsRNA) and a polymeric conjugate,

wherein said dsRNA is polyinosinic-polycytidylic acid double stranded RNA (polyIC),

wherein said polymeric conjugate comprises a polyethyleneimine (PEI), one or more polyethylene glycol (PEG) moieties and one or more targeting moieties,

wherein said PEI is covalently bound to one or more PEG moieties, and each of said one or more PEG moieties is linked to one of said one or more targeting moieties, and

wherein each of said one or more targeting moieties is capable of binding to a cancer antigen; and

b. at least one antibody, wherein said at least one antibody is capable of modulating an immune checkpoint protein, and wherein said at least one antibody capable of modulating an immune checkpoint protein is

(i) at least one antibody capable of agonizing a co-stimulatory immune checkpoint protein, or

(ii) at least one antibody capable of antagonizing a co-inhibitory immune checkpoint protein; or

(iii) a mixture of at least one antibody of (i) and at least one antibody of (ii); and

wherein said immune checkpoint protein is selected from the group consisting of 4-1BB, 4-1BB ligand, PD-1, PD-L1, PD-L2 and CTLA-4.

2. A composition comprising

a. a polyplex comprising a double stranded RNA (dsRNA) and a polymeric conjugate,

wherein said dsRNA is polyinosinic-polycytidylic acid double stranded RNA (polyIC),

wherein said polymeric conjugate comprises a polyethyleneimine (PEI), one or more polyethylene glycol (PEG) moieties and one or more targeting moieties,

wherein said PEI is covalently bound to one or more PEG moieties, and each

of said one or more PEG moieties is linked to one of said one or more targeting moieties, and

wherein each of said one or more targeting moieties is capable of binding to a cancer antigen; and

b. at least one antibody, wherein said at least one antibody is capable of modulating an immune checkpoint protein, and wherein said at least one antibody capable of modulating an immune checkpoint protein is

(i) at least one antibody capable of agonizing a co-stimulatory immune checkpoint protein, or

(ii) at least one antibody capable of antagonizing a co-inhibitory immune checkpoint protein; or

(iii) a mixture of at least one antibody of (i) and at least one antibody of (ii); and

wherein said immune checkpoint protein is selected from the group consisting of 4-1BB, 4-1BB ligand, PD-1, PD-L1, PD-L2 and CTLA-4.

~~3. The composition of claim 1 or the kit of parts of claim 2, wherein said at least one antibody capable of modulating an immune checkpoint protein is~~

~~(i) at least one antibody capable of agonizing a co-stimulatory immune checkpoint protein, wherein said co-stimulatory immune checkpoint protein is selected from the group consisting of 4-1BB, 4-1BB ligand (4-1BBL), CD40, CD40 ligand (CD40L), OX40, OX-40 ligand (OX-40L), GITR, GITR ligand (GITRL), ICOS and ICOS ligand (ICOSL); or~~

~~(ii) at least one antibody capable of antagonizing a co-inhibitory immune checkpoint protein, wherein said co-inhibitory immune checkpoint protein is selected from the group consisting of PD-1, PD-L1, PD-L2, CTLA-4, B7-H3, B7-H4, VISTA, LAG-3, Galectin-9, TIM-3, and TIGIT; or~~

~~(iii) a mixture of at least one antibody of (i) and at least one antibody of (ii).~~

~~4~~3. The composition or the kit-of-parts according to any one of the preceding claims, wherein ~~said immune checkpoint protein is selected from the group consisting of 4-1BB, 4-1BB ligand, PD-1, PD-L1, PD-L2 and CTLA-4, more preferably~~ said immune checkpoint protein is selected from the group consisting of 4-1BB, 4-1BB ligand, PD-1, PD-L1, and PD-L2.

~~5~~4. The composition or the kit-of-parts according to any one of the preceding claims, wherein said immune checkpoint protein is 4-1BB or PD-1.

~~6~~5. The composition or the kit-of-parts of any one of the preceding claims, wherein said at least one antibody capable of modulating an immune checkpoint protein is a mixture of

(i) an antibody capable of modulating immune checkpoint protein CD27 and an antibody capable of modulating an immune checkpoint protein selected from the group consisting of PD-1, PD-L1, and PD-L2;

(ii) an antibody capable of modulating immune checkpoint protein CD40 and at least one antibody capable of modulating an immune checkpoint protein selected from the group consisting of PD-1, PD-L1, PD-L2, and CTLA-4;

(iii) an antibody capable of modulating immune checkpoint protein GITR and at least one antibody capable of modulating an immune checkpoint protein selected from the group consisting of PD-1, PD-L1, PD-L2, and CTLA-4;

(iv) an antibody capable of modulating immune checkpoint protein OX40 and at least one antibody capable of modulating an immune checkpoint protein selected from the group consisting of PD-1, PD-L1, PD-L2, 4-1BB, and CTLA-4;

(v) an antibody capable of modulating immune checkpoint protein 4-1BB and at least one antibody capable of modulating an immune checkpoint protein selected from the group consisting of PD-1, PD-L1, PD-L2, OX40, LAG-3 and CTLA-4; or

(vi) an antibody capable of modulating immune checkpoint protein ICOS and at least one antibody capable of modulating an immune checkpoint protein selected from the group consisting of PD-1, PD-L1, and PD-L2.

~~7~~6. The composition or the kit-of-parts according to any one of the preceding claims, wherein said PEI is covalently bound to one, two or three PEG moieties, preferably to one or three PEG moieties.

~~8~~7. The composition or the kit-of-parts according to any one of the preceding claims, wherein said polyethyleneimine (PEI) is linear polyethyleneimine (LPEI).

~~9. The composition or the kit-of-parts according to any one of the preceding claims, wherein said dsRNA is polyinosinic-polycytidylic acid double-stranded RNA~~

~~(polyIC).~~

~~10~~8. The composition or the kit-of-parts according to any one of the preceding claims, wherein said cancer antigen is EGFR, HER2 or PSMA.

9. The composition or the kit-of-parts according to any one of the preceding claims, wherein~~, preferably~~ said cancer antigen is EGFR.

~~11~~10. The composition or the kit-of-parts according to any one of the preceding claims, wherein said one or more targeting moieties are selected from the group consisting of EGF, a HER2 affibody, a HER2 antibody, and a DUPA moiety ($HOOC(CH_2)_2$-CH(COOH)-NH-CO-NH-CH(COOH)-$(CH_2)_2$-CO-).

~~12~~11. The composition or the kit-of-parts according to any one of the preceding claims, wherein said one or more targeting moieties is EGF.

12. The composition or the kit-of-parts according to any one of the preceding claims, wherein said one or more targeting moieties is~~, preferably~~ human EGF.

13. The composition or the kit-of-parts according to any one of the preceding claims for use in therapy.

14. Use of the composition or the kit-of-parts according to any one of the preceding claims in the manufacture of a medicament for treating cancer in a mammal.

15. The use of claim 14, wherein said mammal is~~, preferably~~ a human.

~~15~~16. The use of claim 14 or claim 15, wherein said cancer is selected from the group consisting of melanoma, non-small-cell lung cancer, breast cancer, ovarian cancer, cervical cancer, endometrial cancer, vulva cancer, urothelial cancer, bladder cancer, renal cancer, esophageal cancer, gastric cancer, pancreatic cancer, colorectal cancer, glial tumor, head and neck cancer, prostate cancer, penile cancer, testicular-embryonal cancer, neuroendocrine tumor, and hepatocellular cancer.

~~16~~17. The use of any one of the claims 14 ~~or 15~~to 16, wherein the polyplex is to be administered separately from the at least one antibody capable of modulating an immune checkpoint protein to said mammal.

18. The use of claim 17, wherein said mammal is a~~, preferably~~ human.