

CLAIMS

What is claimed is:

1. An isolated and purified active substance comprising E3LΔ83N-TK⁻-hFlt3L in replicative or inactivated form suitable for use as an immunotherapeutic agent against a malignant solid tumor.
2. A method for treating a subject afflicted with one or more solid malignant tumors, the method comprising delivering to cells of the tumor replication competent or inactivated E3LΔ83N-TK⁻-hFlt3L virus and thereby treating the tumor.
3. The method of claim 2 wherein the amount is effective to accomplish one or more of the following:
 - induce the immune system of the subject to mount an immune response against the tumor;
 - reduce the size of the tumor;
 - eradicate the tumor;
 - inhibit growth of the tumor;
 - inhibit metastasis of the tumor; and
 - reduce or eradicate metastatic tumor.
4. The method of claim 3 wherein the tumor includes tumor located at the site of delivery, or tumor located both at said site and elsewhere in the body of the subject.
5. The method of any one of claims 2 – 4 wherein the immune response comprises one or more of the following:
 - increase in cytotoxic CD8⁺ T cells within the tumor and/or in tumor-draining lymph nodes;
 - induction of maturation of dendritic cells infiltrating said tumor through induction of type I IFN;
 - induction of activated CD4⁺ effector T cells in the subject recognizing tumor cells within the tumor or systemically

- increase of CD103+ dendritic cells in noninjected tumors of the subject.
6. The method of claim 2 – 5 wherein the tumor is primary or metastatic melanoma or breast carcinoma or colon carcinoma.
7. A method for treating a solid malignant tumor in a subject comprising delivering to tumor cells of the subject an amount of replication competent or inactivated E3LΔ83N-TK⁻-hFlt3L virus effective to induce the immune system of the subject to mount an immune response against the tumor.
8. The method of claim 7 wherein the immune response is systemic.
9. The method of claim 7 wherein the immune response effects or contributes to one or more of the following: reduction of the size of the tumor, eradication of the tumor, inhibition of tumor or metastatic growth.
10. The method of claim 7 or 8 wherein the virus is effective to accomplish one or more of the following:
- induce the immune system of the subject to mount an immune response against the tumor;
 - reduce the size of the tumor;
 - eradicate the tumor;
 - inhibit growth of the tumor;
 - inhibit metastasis of the tumor; and
 - reduce or eradicate metastatic tumor.
11. A method for treating a malignant tumor in a subject, the method comprising delivering to tumor cells of the subject replication competent or inactivated E3LΔ83N-TK⁻-hFlt3L virus in an amount effective to induce the immune system of the subject to mount an immune response against the tumor and conjointly administering or having administered to the subject a second amount of an immune checkpoint blocking agent effective to block

immune suppressive mechanisms within the tumor elicited by tumor cells, stromal cells, or tumor infiltrating immune cells.

12. A method for treating a malignant tumor in a subject, the method comprising delivering or having delivered to tumor cells of the subject replication competent or inactivated E3LΔ83N-TK⁻-hFlt3L virus in an amount effective to induce the immune system of the subject to mount an immune response against the tumor and conjointly administering or having administered to the subject a second amount of an immune checkpoint blocking agent effective to block immune suppressive mechanisms within the tumor elicited by tumor cells, stromal cells, or tumor infiltrating immune cells.

13. The method of claim 10 or 11 wherein the conjoint administration is effective to accomplish one or more of the following:

- induce the immune system of the subject to mount an immune response against the tumor;
- reduce the size of the tumor;
- eradicate the tumor;
- inhibit growth of the tumor;
- inhibit metastasis of the tumor; and
- reduce or eradicate metastatic tumor.

14. The method of claim 11 or 12 wherein the tumor is primary or metastatic malignant melanoma or breast carcinoma or colon carcinoma.

15. The method of any one of claims 2-13 wherein the virus is heat-inactivated.

16. A composition comprising an effective amount for treating a patient afflicted with a solid malignant tumor an active ingredient comprising E3LΔ83N-TK⁻-hFlt3, in replicative or inactivated form, or both, and a pharmaceutically acceptable excipient.

17. The composition of claim 16 wherein the amount is effective to accomplish one or more of the following: reduce the size of the tumor, eradicate the tumor, inhibit

growth of the tumor, or inhibit metastasis or metastatic growth of the tumor and thereby treat the tumor.

18. The composition of claim 16 wherein the amount is effective to elicit in the treated subject an immune response against the tumor and any metastases thereof, upon local delivery to tumor cells of the subject.
19. The composition of claim 17 wherein the immune response includes one or more of the following:
 - oncolysis of tumor cells and release of tumor antigen;
 - an increase in cytotoxic CD8+ T cells within the tumor and/or in tumor-draining lymph nodes;
 - induction of maturation of dendritic cells infiltrating said tumor or circulating in remote locations within the patient's body through induction of type I IFN;
 - induction of effector CD4+ T cells in the subject recognizing tumor cells within the tumor and/or in tumor draining lymph nodes;
 - increase of CD103+ dendritic cells in noninjected tumors of the subject.
20. An isolated purified active substance selected from the group consisting of E3LΔ83N-TK⁻, E3LΔ83N-TK⁻-GM-CSF, and E3LΔ83N-TK⁻-Flt3L, in replicative or inactivated form, suitable for use as an immunotherapeutic agent against a malignant solid tumor.
21. A composition comprising an effective amount for treating a patient afflicted with a solid malignant tumor of an active ingredient comprising one or more of E3LΔ83N-TK⁻, E3LΔ83N-TK⁻-GM-CSF, and E3LΔ83N-TK⁻-hFlt3L viruses and viral constructs, each optionally in replicative or inactivated form, and a pharmaceutically acceptable excipient.
22. The composition of claim 21 containing two or more of said viruses and viral constructs.

23. The composition of claim 21 or 22 wherein the amount is effective to accomplish one or more of the following: reduce the size of the tumor, eradicate the tumor, inhibit growth of the tumor, or inhibit metastasis or metastatic growth of the tumor and thereby treat the tumor.

24. The composition of claim 21 or 22 wherein the amount is effective to elicit in the treated subject an immune response against the tumor and other tumors in the treated subject's body, upon local delivery to tumor cells of the subject.

25. The composition of claim 24 wherein the immune response may include one or more of the following:

- oncolysis of tumor cells and release of tumor antigen;
- an increase in cytotoxic CD8+ T cells within the tumor and/or in tumor-draining lymph nodes;
- induction of maturation of dendritic cells infiltrating said tumor or circulating in remote locations within the patient's body through induction of type I IFN;
- induction of effector CD4+ T cells in the subject recognizing tumor cells within the tumor and/or in tumor draining lymph nodes
- increase of CD103+ dendritic cells in noninjected tumors of the subject.

26. A method for treating a solid malignant tumor in a subject comprising delivering to tumor cells of the subject an amount of one or more of E3LΔ83N-TK-, E3LΔ83N-TK--GM-CSF, and E3LΔ83N-TK--hFlt3L viruses and viral constructs, each optionally in replicative or inactivated form, effective to induce the immune system of the subject to mount an immune response against the tumor.

27. A method for treating a solid malignant tumor in a subject comprising delivering to tumor cells of the subject an amount of one or more of E3LΔ83N-TK⁻, E3LΔ83N-TK⁻-GM-CSF, and E3LΔ83N-TK⁻-hFlt3L viruses and viral constructs, including replicative and inactivated versions of each of the foregoing, effective to accomplish one or more of the following (regardless of order):

- induce the immune system of the subject to mount an immune response against the tumor;

- reduce the size of the tumor;
- eradicate the tumor;
- inhibit growth of the tumor;
- inhibit metastasis of the tumor; and
- reduce or eradicate metastatic tumor.

28. The method of claim 26 wherein the immune response may include one or more of the following immunological effects

- oncolysis of tumor cells and release of tumor antigen;
- an increase in cytotoxic CD8⁺ T cells within the tumor and/or in tumor-draining lymph nodes;
- induction of maturation of dendritic cells infiltrating said tumor or circulating in remote locations within the patient's body through induction of type I IFN;
- induction of effector T cells in the subject recognizing tumor cells within the tumor and/or in tumor draining lymph nodes
- increase of CD103⁺ dendritic cells in noninjected tumors of the subject.

29. A method for treating a malignant tumor in a subject, the method comprising delivering to tumor cells of the subject replication competent or inactivated E3LΔ83N-TK⁻, E3LΔ83N-TK⁻-GM-CSF, and E3LΔ83N-TK⁻-hFlt3L viruses or viral constructs, each optionally in replicative or inactivated form, in an amount effective to induce the immune system of the subject to mount an immune response against the tumor and conjointly administering to the subject a second amount of an immune checkpoint blocking agent effective to block immune suppressive mechanisms within the tumor elicited by tumor cells, stromal cells, or tumor infiltrating immune cells.

30. The method of claim 29 wherein the conjoint administration is effective to accomplish one or more of the following:

- induce the immune system of the subject to mount an immune response against the tumor;
- reduce the size of the tumor;
- eradicate the tumor;

- inhibit growth of the tumor;
- inhibit metastasis of the tumor; and
- reduce or eradicate metastatic tumor.

31. A method for treating a malignant tumor in a subject, wherein the subject has been previously treated or dosed with replication competent or inactivated E3LΔ83N-TK⁻, E3LΔ83N-TK⁻-GM-CSF, and E3LΔ83N-TK⁻-hFlt3L viruses or viral constructs, each optionally in replicative or inactivated form, in an amount effective to induce the immune system of the subject to mount an immune response against the tumor

the method comprising delivering to the subject tumor cells of the subject an amount of an immune checkpoint blocking agent effective to block immune suppressive mechanisms within the tumor elicited by tumor cells, stromal cells, or tumor infiltrating immune cells.

32. A method for treating a malignant tumor in a subject, wherein the subject has been previously treated or dosed with an amount of an immune checkpoint blocking agent effective to block immune suppressive mechanisms within the tumor elicited by tumor cells, stromal cells, or tumor infiltrating immune cells

the method comprising delivering to the subject tumor cells of the subject replication competent or inactivated E3LΔ83N-TK⁻, E3LΔ83N-TK⁻-GM-CSF, and E3LΔ83N-TK⁻-hFlt3L viruses or viral constructs, each optionally in replicative or inactivated form, in an amount effective to induce the immune system of the subject to mount an immune response against the tumor.

33. The method of claim 29, 31, or 32, wherein the immune checkpoint blocking agent comprises CTLA-4, CD80, CD86, PD-1, PDL1, PDL2, LAG3, B7-H3, B7-H4, TIM3, ICOS, II DLBCL inhibitors, BTLA, or any combination thereof.

34. The method of claim 33, wherein the immune checkpoint blocking agent comprises ipilimumab, nivolumab, pembrolizumab, pidilizumab, AMP-224, MPDL3280A, BMS-936559, MEDI4736, MSB 00107180, or any combination thereof.