of soluble PD-L1, is therefore a novel potential target for therapy to accompany immune checkpoint blockade.

PD-1 (also known as programmed cell death protein 1 and CD279) is a checkpoint protein on T cells, which acts as a type of “off switch” to assist in preventing the T cells from attacking normal healthy cells in the body. It does this when it binds to PD-L1 (also known as programmed death-ligand 1 and CD274)], a protein on some normal and cancer cells. When PD-1 binds to PD-L1 on a cell, it induces a signal that instructs the T cell to leave the other cell alone. Some cancer cells produce large amounts of PD-L1, which helps them avoid identification by the adaptive immune system and thus evade attack from the host immune system.

Immune checkpoint proteins present on immune cells and/or cancer cells [e.g. CTLA4 (also known as cytotoxic T-lymphocyte-associated protein 4 and CD152), LAG3 (also known as lymphocyte-activation gene 3 and CD223), PD-1 and PD-L1, TIGIT, TIM-3, and BTLA] are molecular targets that have been found to play an important role in regulating anti-tumour immune responses. Inhibitors of these immune checkpoint proteins (e.g. CTLA4, LAG3, TIGIT, TIM-3, BTLA, PD-1 and/or PD-L1 inhibitors) promote an anti-tumour immune response that can be utilised to effectively treat certain forms of cancer.

OX40L, also known as TNFSF4 and CD252, is expressed on antigen presenting cells but has been described in many other cells including melanoma cells. It binds to OX40, a member of the TNF Receptor superfamily, on T lymphoctes to promote activation of effector T cells. There are several clinical studies underway investigating the possible role of OX40L as a way of enhancing anti-neoplastic cell T lymphocyte responses, including in combination with drugs that block the PD-L1/PD-1 axis.

Ligation of OX40L with OX40 in T-lymphocytes promotes maintenance and generation of memory CD8+ T cells. Furthermore, it has been shown that induced expression of OX40L in tumours led induction of antitumour immunity (Andarini, Sita, et al. "Adenovirus vector-mediated in vivo gene transfer of OX40 ligand to tumor cells enhances antitumor immunity of tumor-bearing hosts." *Cancer Research* 64(9):3281-3287, 2004).

SUMMARY

Data is presented in the example section herein that shows that the NUC-7738 causes a reduction in mRNA and protein of extra-cellular forms of PD-L1, in particular soluble and exosomal PD-L1. PD-L1 produced by cancer cells is known to be important in aiding the

cancer cell to avoid host immune attack. Soluble PD-L1 and exosomal PD-L1 (types of extra cellular PD-L1) can act as a decoy protein blocking activation of T-cells and reducing their capacity to kill tumour cells. Accordingly, NUC-7738 can be used to reduce the production of this decoy protein rendering the cancer cell more susceptible to attack via the host's immune system. Data is presented in the example section that shows that NUC-7738 also causes an increase in the amount of OX40L produced by cancer cells. By reducing the amount of extra cellular PD-L1 (e.g soluble PD-L1 and/or exosomal PD-L1), and/or increasing the amount of OX40L produced by the cancer cells, NUC-7738 is exhibiting the properties of an immune sensitiser and so can be used alone or to potentiate the therapeutic effect of oncology agents or therapies, which can include immune checkpoint inhibitors (e.g. CTLA4, LAG3, PD-1 or PD-L1 inhibitors), as well as cancer vaccines and adoptive cell therapies such as chimeric antigen receptor T cells (CAR-T), tumor-infiltrating lymphocyte (TIL) and natural killer (NK) cell therapies. NUC-7738 may also be suitable for treating cancer patients with high levels of soluble PD-L1 and/or exosomal PD-L1 in the plasma and/or with tumours expressing low levels of OX40L and for use in treating cancers that have developed resistance to certain immune checkpoint inhibitors. Furthermore, by reducing the amount of the decoy protein PD-L1, it may allow the use of lower doses of an immune checkpoint inhibitor, and so not only reduce the toxic side effects of such agent but allow patients that have had to cease treatment with an immune checkpoint inhibitor, for example due to toxicity, to reinstate treatment with the or an immune checkpoint inhibitor but with a lower less toxic dose. NUC-7738 is therefore suitable for use in combination with an immune checkpoint inhibitor in the treatment of a cancer patient that has had their treatment with an immune checkpoint inhibitor stopped, perhaps for toxicity reasons.

Thus, the present disclosure relates, in part, to methods for treating a proliferative disease such as cancer. Suitably the method comprises administering a therapeutically effective amount of a composition to a subject in need of such composition, wherein said composition comprises NUC-7738. Suitably, the therapeutically effective amount is an amount capable of reducing the production of extra-cellular PD-L1 (such as soluble PD-L1 and/or exosomal PD-L1) by the cancer cells and/or increasing the production of OX40-L by the cancer cells. In particular examples, the NUC-7738 is capable of reducing the levels of soluble PD-L1 produced by cancer cells. In particular examples, the NUC-7738 is capable of reducing the levels of exosomal PD-L1 produced by cancer cells. In particular examples, the NUC-7738 is capable of increasing the production of OX40-L by the cancer cells. Suitably, the NUC-7738 enhances the host's adaptive immune system. In particular examples, the NUC-7738 is acting as an immune sensitiser. In particular examples, the NUC-7738 can be used to potentiate the effects of immunotherapy. Suitably the immunotherapy is an

immune checkpoint inhibitor or a cancer vaccine or an adoptive cell therapy (also known as cellular immunotherapy) such as CAR-T cell therapy. In particular examples, the NUC-7738 is used in combination with an immune checkpoint inhibitor. In particular examples the NUC-7738 is used in combination with an immune checkpoint inhibitor selected from pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. In particular examples, the NUC-7738 is used in combination with an an anti-PD-1 antibody selected from pembrolizumab, cemiplimab, dostarlimab and nivolumab. In particular examples, the NUC-7738 is used in combination with pembrolizumab to treat cancer, such as cutaneous melanoma.

In various examples, the proliferative disease is one in which high levels of extra-cellular PD-L1 (e.g. soluble PD-L1 and/or exosomal PD-L1) contribute to the pathogenesis of the proliferative disease, e.g. cancer. In particular examples, high levels of soluble PD-L1 and/or exosomal PD-L1 inhibits or blocks the host immune system from attacking the cancer cell. In particular examples, high levels of soluble PD-L1 and/or exosomal PD-L1 inhibits or blocks the effect of an immune checkpoint inhibitor given to the patient. Accordingly, in particular examples, NUC-7738 is for use in treating a patient whose cancer cells express high levels of extra-cellular PD-L1 (such as soluble PD-L1 and/or exosomal PD-L1) protein.
In particular examples, the NUC-7738 reduces the expression of soluble PD-L1 and/or exosomal PD-L1 protein. In particular examples, the NUC-7738 reduces the transcription of the mRNA encoding soluble PD-L1 and/or exosomal PD-L1 protein.
In various examples, the proliferative disease is one in which low levels of OX40-L contribute to the pathogenesis of the proliferative disease, e.g. cancer. In particular examples, low levels of OX40-L inhibit or blocks the host immune system from attacking the diseased cell. In particular examples, low levels of OX40-L inhibit or blocks the effect of an immune checkpoint inhibitor given to the patient. Accordingly, in particular examples, NUC-7738 is for use in treating a patient whose cancer cells express low levels of OX40-L protein.

In particular examples, the NUC-7738 increases the expression of OX40-L protein by the cancer cells. In particular examples, the NUC-7738 increases the translation of the mRNA encoding OX40-L.

According to one example the present disclosure provides NUC-7738 for use in the treatment of a proliferative disease, such as cancer by reducing the amount of extra-cellular PD-L1

protein and/or increasing the amount of OX40-L protein produced by the diseased cells. Suitably, the extra-cellular PD-L1 is soluble PD-L1 protein and/or exosomal PD-L1 protein. Suitably, the treatment arises through adaptive immunity (e.g. cellular immunity and/or humoral immunity). In one aspect, the present disclosure refers to the use of NUC-7738 in the manufacture of a medicament for the treatment of a proliferative disease in a patient, wherein the medicament enhances the patient's immune response against the proliferative disease. In another aspect, the present disclosure refers to the use of NUC-7738 in the manufacture of a medicament for the treatment of cancer, wherein the medicament is an immune-sensitiser.

According to another example the present disclosure provides a method of treating a proliferative disease, such as cancer by reducing the amount of extra-cellular PD-L1 protein and/or increasing the amount of OX40-L protein, produced by proliferative disease cells in a patient comprising administering a therapeutically effective amount of NUC-7738 to a patient in need thereof. Suitably, the extra-cellular PD-L1 is soluble PD-L1 protein and/or exosomal PD-L1 protein.

According to another example the present disclosure provides use of NUC-7738 in the manufacture of a medicament for use in a method of treating a proliferative disease, such as cancer by reducing the amount of extra-cellular PD-L1 protein and/or increasing the amount of OX40-L protein, produced by the diseased cells. Suitably, the extra-cellular PD-L1 is soluble PD-L1 protein and/or exosomal PD-L1 protein. Suitably, the proliferative disease is cancer. In a particular example, reducing the amount of extra-cellular PD-L1 protein, such as soluble PD-L1 protein and/or exosomal PD-L1 protein, produced by the cancer cells renders the cancer cells more susceptible to targeting by the host immune system, such as the patient's cellular immune system.

According to one example the present disclosure provides NUC-7738 for use in the treatment of a proliferative disease, such as cancer, by reducing the amount of extra-cellular PD-L1 protein produced by the diseased cells. Suitably, the extra-cellular PD-L1 is soluble PD-L1 protein and/or exosomal PD-L1 protein. Suitably, the treatment arises through adaptive immunity (e.g. cellular immunity and/or humoral immunity).

According to another example the present disclosure provides a method of treating cancer by reducing the amount of extra-cellular PD-L1 protein, such as soluble PD-L1 protein and/or exosomal PD-L1 protein, produced by cancer cells in a patient comprising administering a therapeutically effective amount of NUC-7738 to a patient in need thereof.

According to another example the present disclosure provides use of NUC-7738 in the manufacture of a medicament for use in a method of treating cancer by reducing the amount of extra-cellular PD-L1 protein, such as soluble PD-L1 protein and/or exosomal PD-L1 protein, produced by the cancer cells. In a particular example, reducing the amount of

extra-cellular PD-L1 protein, such as soluble PD-L1 protein and/or exosomal PD-L1 protein, produced by the cancer cells renders the cancer cells more susceptible to targeting by the host immune system, such as the patient's cellular immune system. The ability of NUC-7738 to cause a reduction in the production and release of extra-cellular PD-L1 protein, such as soluble PD-L1 and exosomal PD-L1 protein, supports that NUC-7738 is an immune-sensitiser and can be used as an immune sensitising agent. This role is further supported by the finding that NUC-7738 also causes an increase in the expression and production of OX40-L by the cancer cells, which interacts with OX40 on the surface of T lymphocytes to increase their anti-neoplastic effectiveness.

According to another example the present disclosure provides NUC-7738 for use in the treatment of a proliferative disease by enhancing the patient's immune response against the proliferative disease. Suitably, the NUC-7738 enhances the patient's adaptive immune response. Suitably, NUC-7738 suppresses one or more immune blockers, such as extra cellular PD-L1 and/or OX40-L.

According to another example the present disclosure provides NUC-7738 for use as an immune-sensitiser. Suitably, the present disclosure provides NUC-7738 for use as an immune-sensitiser in the treatment of cancer.

According to another example the present disclosure provides the use of NUC-7738 in the manufacture of an immune-sensitiser medicament. Alternatively, the disclosure provides the use of NUC-7738 in the manufacture of a medicament for use in the treatment of cancer, wherein the NUC-7738 is an immune-sensitiser.

According to another example the present disclosure provides a method of potentiating the immune response to a tumour, the method comprising administering to a patient in need of such treatment a therapeutically effective amount of NUC-7738, alone or in combination with an immune oncology agent as described herein. In a particular example, there is provided a method of potentiating the effect of an an immune oncology agent, the method comprising administering to a patient in need of such treatment a therapeutically effective amount of the immune oncology agent simultaneously or sequentially with NUC-7738. According to another example the present disclosure provides NUC-7738 for use in the treatment of cancer by increasing the amount of OX40-L protein produced by the cancer cells. Suitably, the treatment of cancer arises through adaptive immunity (e.g. cellular immunity and/or humoral immunity).

According to another example the present disclosure provides a method of treating cancer by increasing the amount of OX40-L protein produced by the cancer cells in a patient comprising administering a therapeutically effective amount of NUC-7738 to a patient in need thereof.

According to another example the present disclosure provides use of NUC-7738 in the manufacture of a medicament for use in a method of treating cancer by increasing the amount of OX40-L protein produced by the cancer cells.

In a particular example, increasing the amount of OX40-L protein produced by the cancer cells renders the cancer cells more susceptible to targeting by the host immune system, such as the patient's cellular immune system by promoting effector T cell activation.

According to another example the present disclosure provides NUC-7738 for use in the treatment of cancer by reducing a blockage against targeting the cancer cells by the patient's immune system. Suitably "reducing a blockage" comprises reducing suppression or a suppressor of the patient's immune system.

According to another example the present disclosure provides NUC-7738 for use in the treatment of cancer by enhancing the patient's immune response against the cancer. Suitably, the NUC-7738 augments the patient's immune system against the cancer.

According to another example the present disclosure provides use of NUC-7738 in the manufacture of a medicament for use in a method for treating cancer by enhancing the patient's immune response against the cancer.

According to another example the present disclosure provides a method of treating cancer by enhancing the patient's immune response against the cancer, wherein the method comprises administering to the patient in need thereof a therapeutically effective amount of NUC-7738.

According to another example the present disclosure provides a combination comprising NUC-7738 and an immune oncology agent.

In particular examples, the immune oncology agent is an immune checkpoint inhibitor, a cancer vaccine, antibody therapy, or an adoptive cell therapy such as CAR-T cell.

In particular examples, the immune oncology agent is an immune checkpoint inhibitor

selected from pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. Suitably, the immune checkpoint inhibitor is an anti-PD-1 antibody selected from pembrolizumab, cemiplimab, dostarlimab and nivolumab. In a particular examples, the immune checkpoint inhibitor is pembrolizumab.

According to another example the present disclosure provides a pharmaceutical product comprising NUC-7738 and an immune oncology agent.

In a particular example, the immune oncology agent is an immune checkpoint inhibitor. The immune checkpoint inhibitor may be an antibody against an immune checkpoint protein. Suitably the immune checkpoint inhibitor is an antibody selected from the group consisting of: an anti-PD-L1 antibody, an anti-PD-1 antibody, an anti-CTLA-4 antibody, an anti-LAG-3 antibody, an anti-TIGIT antibody, an anti-BTLA antibody, an anti-TIM-3 antibody, an anti-CD40 antibody and an anti-CD40L antibody.

Suitably the immune checkpoint inhibitor is selected from pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. Suitably, the immune checkpoint inhibitor is an anti-PD-1 antibody selected from pembrolizumab, cemiplimab, dostarlimab and nivolumab. In a particular example the immune checkpoint inhibitor is pembrolizumab.

In one example, the pharmaceutical product may comprise a kit of parts comprising separate formulations of NUC-7738 and an immune checkpoint inhibitor. The separate formulations of NUC-7738 and the immune checkpoint inhibitor may be administered sequentially and/or simultaneously.

In another example the pharmaceutical product is a kit of parts which comprises:

a first container comprising NUC-7738, such as NUC-7738 in association with a pharmaceutically acceptable adjuvant, diluent or carrier; and

a second container comprising an immune checkpoint inhibitor such as an immune checkpoint inhibitor in association with a pharmaceutically acceptable adjuvant, diluent or carrier, and

a container means for containing said first and second containers, optionally wherein the immune checkpoint inhibitor is pembrolizumab.

In one example, the pharmaceutical product may comprise a one or more unit dosage forms (e.g. vials, tablets or capsules in a blister pack). In one example, each unit dose

comprises only one agent selected from NUC-7738 and the immune checkpoint inhibitor. In another example, the unit dosage form comprises both the NUC-7738 compound and the immune checkpoint inhibitor.

According to another example the present disclosure provides a combination comprising NUC-7738 and an immune checkpoint inhibitor as defined herein for use in the treatment of a proliferative disease. Suitably, the proliferative disease is cancer. In particular examples, the proliferative disease is one that is normally treated with an immune checkpoint inhibitor, such as one selected from the group consisting of: pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. Table 1 provides a list of diseases that particular checkpoint inhibitor agents can be used to treat.
The following combinations and diseases are specific examples:

- NUC-7738 in combination with nivolumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), renal cell carcinoma, non-small cell lung cancer, mesothelioma, classical hodgkin lymphoma, squamous cell carcinoma, urothelial carcinoma, colorectal cancer, hepatocellular carcinoma, eosophogeal cancer, gastric cancer and, gastroesophageal junction cancer
- NUC-7738 in combination with durvalumab for use in treating a cancer selected from: urothelial carcinoma, , non-small cell lung-cancer (NSCLC) and extensive stage small cell lung-cancer.
- NUC-7738 in combination with pembrolizumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), non-small cell lung cancer, urothelial carcinoma, classical Hodgkin lymphoma, head and neck squamous cell carcinoma, renal cell carcinoma, colorectal cancer and esophageal carcinoma.
- NUC-7738 in combination with dostarlimab for use in treating a cancer selected from: mismatch repair deficient (dMMR) recurrent or advanced endometrial cancer.
- NUC-7738 in combination with ipilimumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), renal cell carcinoma, mon-small cell lung cancer, mesothelioma, classical hodgkin lymphoma, colorectal cancer, hepatocellular carcinoma and eosophogeal cancer.
- NUC-7738 in combination with cemiplimab for use in treating a cancer selected from: cutaneous squamous cell carcinoma, basal cell carcinoma and non-small cell lung cancer.
- NUC-7738 in combination with atezolizumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), non-small cell lung cancer, small cell lung cancer, urothelial carcinoma, an hepatocellular carcinoma.

- NUC-7738 in combination with avelumab for use in treating a cancer selected from: merkel cell carcinoma, urothelial carcinoma and renal cell carcinoma.

Suitably, the proliferative disease is cancer and the cancer cells express high levels of soluble or exosomal PD-L1. Suitably, the proliferative disease is cancer and the cancer cells express low levels of OX40L.

According to another example the present disclosure provides a method of treating a proliferative disease in a subject in need thereof comprising administering to said subject a combination comprising NUC-7738 and an immune checkpoint inhibitor as defined herein. In particular examples, the proliferative disease is one that is normally treated with an immune checkpoint inhibitor, such as one selected from the group consisting of: pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. Table 1 provides a list of diseases that particular checkpoint inhibitor agents can be used to treat.

The following combinations and diseases are specific examples:

- NUC-7738 in combination with nivolumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), renal cell carcinoma, non-small cell lung cancer, mesothelioma, classical hodgkin lymphoma, squamous cell carcinoma, urothelial carcinoma, colorectal cancer, hepatocellular carcinoma, eosophogeal cancer, gastric cancer and, gastroesophageal junction cancer
- NUC-7738 in combination with durvalumab for use in treating a cancer selected from: urothelial carcinoma, , non-small cell lung-cancer (NSCLC) and extensive stage small cell lung-cancer.
- NUC-7738 in combination with pembrolizumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), non-small cell lung cancer, urothelial carcinoma, classical Hodgkin lymphoma, head and neck squamous cell carcinoma, renal cell carcinoma, colorectal cancer and esophageal carcinoma.
- NUC-7738 in combination with dostarlimab for use in treating a cancer selected from: mismatch repair deficient (dMMR) recurrent or advanced endometrial cancer.
- NUC-7738 in combination with ipilimumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), renal cell carcinoma, mon-small cell lung cancer, mesothelioma, classical hodgkin lymphoma, colorectal cancer, hepatocellular carcinoma and eosophogeal cancer.
- NUC-7738 in combination with cemiplimab for use in treating a cancer selected from: cutaneous squamous cell carcinoma, basal cell carcinoma and non-small cell lung cancer.

- NUC-7738 in combination with atezolizumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), non-small cell lung cancer, small cell lung cancer, urothelial carcinoma, an hepatocellular carcinoma.
- NUC-7738 in combination with avelumab for use in treating a cancer selected from: merkel cell carcinoma, urothelial carcinoma and renal cell carcinoma.

Suitably, the immune checkpoint inhibitor is pembrolizumab, cemiplimab, dostarlimab or nivolumab.

Suitably, the proliferative disease is cancer and the cancer cells express high levels of soluble or exosomal PD-L1. Suitably, the proliferative disease is cancer and the cancer cells express low levels of OX40L.

According to another example the present disclosure provides the use of NUC-7738 in the manufacture of a medicament for treating of a proliferative disease in combination with an immune checkpoint inhibitor as defined herein.

In particular examples, the proliferative disease is one that is normally treated with an immune checkpoint inhibitor, such as one selected from the group consisting of: pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. Table 1 provides a list of diseases that particular checkpoint inhibitor agents can be used to treat.

The following combinations and diseases are specific examples:

- NUC-7738 in combination with nivolumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), renal cell carcinoma, non-small cell lung cancer, mesothelioma, classical hodgkin lymphoma, squamous cell carcinoma, urothelial carcinoma, colorectal cancer, hepatocellular carcinoma, eosophogeal cancer, gastric cancer and, gastroesophageal junction cancer
- NUC-7738 in combination with durvalumab for use in treating a cancer selected from: urothelial carcinoma, , non-small cell lung-cancer (NSCLC) and extensive stage small cell lung-cancer.
- NUC-7738 in combination with pembrolizumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), non-small cell lung cancer,urothelial carcinoma, classical Hodgkin lymphoma, head and neck squamous cell carcinoma, renal cell carcinoma, colorectal cancer and esophageal carcinoma.
- NUC-7738 in combination with dostarlimab for use in treating a cancer selected from: mismatch repair deficient (dMMR) recurrent or advanced endometrial cancer.
- NUC-7738 in combination with ipilimumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), renal cell carcinoma, mon-small cell lung

cancer, mesothelioma, classical hodgkin lymphoma, colorectal cancer, hepatocellular carcinoma and eosophogeal cancer.

- NUC-7738 in combination with cemiplimab for use in treating a cancer selected from: cutaneous squamous cell carcinoma, basal cell carcinoma and non-small cell lung cancer.
- NUC-7738 in combination with atezolizumab for use in treating a cancer selected from: melanoma (including cutaneous melanoma), non-small cell lung cancer, small cell lung cancer, urothelial carcinoma, an hepatocellular carcinoma.
- NUC-7738 in combination with avelumab for use in treating a cancer selected from: merkel cell carcinoma, urothelial carcinoma and renal cell carcinoma.

Suitably, the proliferative disease is cancer and the cancer cells express high levels of soluble or exosomal PD-L1. Suitably, the proliferative disease is cancer and the cancer cells express low levels of OX40L. Suitably the immune checkpoint inhibitor is selected from pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. Suitably, the immune checkpoint inhibitor is an anti-PD-1 antibody selected from pembrolizumab, cemiplimab, dostarlimab and nivolumab. A particularly suitable immune checkpoint inhibitor for use in combination with NUC-7738 for treating cancer, such as cutaneous melanoma, is pembrolizumab.
In particular examples of this example of the disclosure the NUC-7738 and immune checkpoint inhibitor are administered to the subject simultaneously or sequentially.

According to another example the present disclosure provides NUC-7738 as defined herein for use in the treatment of a proliferative disease, wherein the NUC-7738 is for simultaneous or sequential administeration with an immune checkpoint inhibitor as defined herein. Suitably, the immune checkpoint inhibitor is selected from the group consisting of: pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. Suitably, the proliferative disease is one that is normally treated with an immune checkpoint inhibitor, such as pembrolizumab, cemiplimab, dostarlimab or nivolumab.

According to another example the present disclosure provides a method of treating a proliferative disease in a subject in need thereof comprising administering to said subject a combination comprising NUC-7738 and an immune checkpoint inhibitor as defined herein, wherein the NUC-7738 and immune checkpoint inhibitor are administered simultaneously or sequentially. Suitably, the immune checkpoint inhibitor is selected from the group consisting of: pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. Suitably, the immune checkpoint inhibitor is an anti-PD-1

antibody selected from pembrolizumab, cemiplimab, dostarlimab and nivolumab. Suitably, the proliferative disease is one that is normally treated with an immune checkpoint inhibitor, such as pembrolizumab, cemiplimab, dostarlimab or nivolumab.

According to another example the present disclosure provides an immune checkpoint inhibitor as defined herein for use in the treatment of a proliferative disease, wherein the immune checkpoint inhibitor is for simultaneous or sequential administeration with NUC-7738 as defined herein. Suitably, the immune checkpoint inhibitor is selected from the group consisting of: pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. Suitably, the immune checkpoint inhibitor is pembrolizumab, cemiplimab, dostarlimab or nivolumab. Suitably, the proliferative disease is one that is normally treated with an immune checkpoint inhibitor, such as pembrolizumab, cemiplimab, dostarlimab or nivolumab. In one aspect, the present disclosure refers to the use of an immune checkpoint inhibitor in the manufacture of a medicament for the treatment of a proliferative disease such as cancer, wherein the immune checkpoint inhibitor is to be administered with NUC-7738, optionally wherein the immune checkpoint inhibitor and NUC-3378 are to be administered simultaneously or sequentially.

According to another example the present disclosure provides the use of an immune checkpoint inhibitor in the manufacture of a medicament for treating of a proliferative disease in combination with NUC-7738 as defined herein. Suitably, the immune checkpoint inhibitor is selected from the group consisting of: pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab. Suitably, the immune checkpoint inhibitor is pembrolizumab, cemiplimab, dostarlimab or nivolumab. Suitably, the proliferative disease is one that is normally treated with an immune checkpoint inhibitor, such as pembrolizumab, cemiplimab, dostarlimab or nivolumab. In another aspect, the present disclosure refers to the use of NUC-7738 in the manufacture of a medicament for the treatment of cancer, wherein the medicament is to be used in combination with an immune oncology agent, such as an immune checkpoint inhibitor.

In particular examples of this example of the disclosure the NUC-7738 and immune checkpoint inhibitor are administered to the subject simultaneously or sequentially.

Suitably the proliferative disease is cancer. In particular examples, the proliferative disease is one that is normally treated with an immune checkpoint inhibitor, such as pembrolizumab, cemiplimab, dostarlimab or nivolumab.According to another

example the present disclosure provides NUC-7738 for use in the treatment of cancer in a subject whose cancer cells express high levels of soluble or exosomal PD-L1.

According to another example the present disclosure provides the use of NUC-7738 in the manufacture of a medicament for treating cancer in a subject whose cancer cells express high levels of extra-cellular PD-L1 protein, such as soluble or exosomal PD-L1 protein, wherein the treatment comprises administering the NUC-7738 in combination with an immune checkpoint inhibitor as defined herein. Optionally, prior to the treating the patient is tested to determine

whether they have a cancer whose cancer cells express high levels of extra-cellular PD-L1 protein, such as soluble PD-L1 protein or exosomal PD-L1 protein. Suitably, the immune checkpoint inhibitor is pembrolizumab, cemiplimab, dostarlimab or nivolumab.

According to another example the present disclosure provides use of NUC-7738 in the manufacture of a medicament for use in a method of treating cancer by reducing the amount of extra-cellular PD-L1 protein, such as soluble protein and/or exosomal PD-L1 protein, produced by the cancer cells, wherein the method comprises determining whether a subject has a cancer whose cancer cells express high levels of extra-cellular PD-L1 protein, wherein if the subject has a cancer whose cancer cells express high levels of extra-cellular PD-L1 protein, such as soluble PD-L1 protein or exosomal PD-L1 protein the subject is administered a combination of NUC-7738 and an immune checkpoint inhibitor as defined herein. Suitably, the immune checkpoint inhibitor is pembrolizumab, cemiplimab, dostarlimab or nivolumab.
According to another example the present disclosure provides a method of treating cancer in a subject whose cancer cells express high levels of extra-cellular PD-L1 protein, such as soluble PD-L1 protein or exosomal PD-L1 protein, comprising administering to the subject a therapeutically effective amount of NUC-7738 alone or in combination with an immune checkpoint inhibitor. Suitably, the immune checkpoint inhibitor is pembrolizumab, cemiplimab, dostarlimab or nivolumab.

According to another example the present disclosure provides a method of treating cancer in a subject whose cancer cells express high levels of extra-cellular PD-L1, such as soluble PD-L1 protein or exosomal PD-L1 protein, comprising contacting the cancer cells with a therapeutically effective amount of NUC-7738. In particular examples of this example of the disclosure the cancer cells are within a subject or patient. Thus, suitably, the cancer cells are contacted with a therapeutically effective amount of NUC-7738 by administering to the subject a therapeutically effective amount of NUC-7738, alone or in combination with an immune checkpoint inhibitor. Suitably, the immune checkpoint inhibitor is pembrolizumab, cemiplimab, dostarlimab or nivolumab.

Any immune checkpoint inhibitor may be used in the combination therapy defined herein.

In particular examples, the checkpoint inhibitor is selected from the group consisting of: an anti-PD-L1 antibody, an anti-PD-1 antibody, an anti-CTLA-4 antibody, an anti-LAG-3

antibody, an anti-TIGIT antibody, an anti-BTLA antibody, an anti-TIM-3 antibody, an anti-CD40 antibody and an anti-CD40L antibody.
Suitably, the immune checkpoint inhibitor is an anti-PD-1 antibody selected from pembrolizumab, cemiplimab, dostarlimab or nivolumab.

In particular examples, the proliferative disease is cancer characterised by cancer cells that express high levels of extra-cellular PD-L1 protein, e.g. soluble PD-L1 or exosomal PD-L1. In particular examples, the proliferative disease is one that is normally treated with an immune checkpoint inhibitor, such as pembrolizumab, cemiplimab, dostarlimab or nivolumab. Table 1 provides a list of diseases that particular checkpoint inhibitor agents can be used to treat.

According to another example the present disclosure provides a method of determining whether a patient will benefit from treatment with NUC-7738, the method comprising: determining the level of soluble PD-L1 and/or exosomal PD-L1 in a biological sample from the patient; wherein if the level of soluble PD-L1 and/or exosomal PD-L1 produced by the cancer cells in the biological sample is elevated or reduced compared to a reference value the patient will benefit from treatment with NUC-7738.
Suitably, the biological sample is a blood sample or a fraction therefrom (e.g. plasma or serum).

The various examples of the disclosure are based in part on the finding that NUC-7738 is able to reduce the amount of extra-cellular PD-L1 protein, including soluble PD-L1 protein and exosomal PD-L1 protein produced by cancer cells. PD-L1 is known to mask the cancer cell from the immune system and elevated soluble PD-L1 in particular is a biomarker of poor prognosis and resistance to immune checkpoint inhibitors. Accordingly, the ability of NUC-7738 to reduce the amount of extra-cellular PD-L1 protein, such as soluble PD-L1 protein and/or exosomal PD-L1 protein, produced by the cancer cells provides new clinical opportunities for treating and managing cancers in patients. For example, by removing or lessening the camouflage that the cancer cell has from the immune system, thus allowing the host immune system to recognise and attack, include kill, the cancer cell. The ability of NUC-7738 to reduce the levels of extra-cellular PD-L1 protein, such as soluble PD-L1 protein and/or exosomal PD-L1 protein, will serve to enhance the effectiveness of an immune oncology agent, including immune checkpoint inhibitors, which provides for the ability to treat a cancer patient with a combination of NUC-7738 and an immune oncology agent. When the immune oncology agent is an immune checkpoint inhibitor, as described

herein, it may also facilitate the use of a lower dose of the checkpoint inhibitor thus lessening the toxic effect of such agent. Certain cancer patients also develop resistance to immune checkpoint inhibitors which is often associated with an increase in soluble PD-L1 levels. The ability of NUC-7738 to target and reduce the amount of soluble or exosomal PD-L1 is predicted to remove the resistance/block and thus allow the patient to be re-treated with the same or an alternate immune checkpoint inhibitor or to be treated with an immune checkpoint inhibitor that would not otherwise be effective without an NUC-7738-induced reduction in soluble PD-L1. NUC-7738 can therefore also be used to treat patients that are or have developed or have pre-existing resistance to an immune checkpoint inhibitor.

Thus, in particular examples of any of the examples of the disclosure the patient or subject is one that is resistant to or has developed resistance to an immune checkpoint inhibitor. Such a person is one where established doses of the immune checkpoint inhibitor do not provide recognised clinical signals of efficacy. Suitably a subject/patient that has developed resistance to an immune checkpoint inhibitor will have previously shown clinical signals of efficacy, such as maintenance or reduction in cancer mass or reduction in markers of cancer, progression free survival, but the effectiveness of the immune checkpoint inhibitor has then reduced or ceased. This is an indication that the cancer has become resistant to the agent. Suitably a subject/patient that is resistant to (has pre-existing resistance to) an immune checkpoint inhibitor will have markers, such as low mutational burden, microsatellite stable, low tumour infiltrating lymphocytes, high levels of immunosuppressive cytokines, PTEN deletion, indicating pre-existing resistance to immune checkpoint inhibitor therapy (see Liu et al., Am J Clin Dermatol. 20(1): 41–54, 2019).
The various examples of the disclosure are also based upon the finding that NUC-7738 is able to increase the amount of OX40L produced by cancer cells. Stimulating OX40 has shown it to be a candidate for therapeutic immunization strategies for cancer.

OX40 signalling is important for priming and generation of CD4 and CD8 T cell immunity, and in development of T cell memory. OX40 is expressed on T-cells following recognition of foreign or aberrant peptides by T-cell receptors on their cell surface. Binding of OX40 by its ligand OX40L enhances the activation of T-cells and has been shown to augment antitumour immune responses (Weinberg et al. IJ Immunol. 164(4):2160-2169, 2000). The OX40/OX40L axis is therefore recognised as an attractive targets for therapy of autoimmune and inflammatory disease, as well as for the design of vaccination and therapeutic adjuvant strategies for infectious disease and cancer (see e.g. : Deng et al. “OX40 (CD14) and OX40 ligand, important immune checkpoints in cancer”. Onco Targets Ther. 12:7347-7353, 2019;

Roszik et al. “TNFSF4 (OX40L) expression and survival in locally advanced and metastatic melanoma”. Cancer Immunology, Immunotherapy. 68:1493-1500, 2019; Dannull J, Nair S, Su Z, Boczkowski D, DeBeck C, Yang B. “Enhancing the immunostimulatory function of dendritic cells by transfection with mRNA encoding OX40 ligand. Blood. 105:3206–3213, 2005).

Reference to NUC-7738 in any example or example of the disclosure includes NUC-7738 as the (S)- phosphate diastereoisomer or as the (R)-phosphate diastereoisomer or as a mixture of phosphate diastereoisomers; it also includes the compound when in the form of a free base or it may be in the form of a pharmaceutically acceptable salt; and, it also includes a pharmaceutical composition comprising the NUC-7738.

Reference to an immune checkpoint inhibitor in any example or example of the disclosure includes the agent or a pharmaceutically acceptable salt thereof; it also includes a pharmaceutical composition comprising the immune checkpoint inhibitor.

The ability of NUC-7738 to reduce the amount/levels of soluble PD-L1 protein and/or exosomal PD-L1 protein and/or increase the amount/level of OX40L protein provides new therapeutic opportunities for treating cancer.

DESCRIPTION OF THE DRAWINGS

Figure 1. Shows concentration of soluble PD-L1 in melanoma (A375) and lung cancer (A549) cell lines, before and after treatment with NUC-7738, by ELISA.
Figure 2. Shows soluble PD-L1 transcript fold change in melanoma (A375) and lung cancer (A549) cell lines, before and after treatment with NUC-7738.
Figure 3. Shows exosomal PD-L1 from four patients, before and after treatment with NUC-7738. Pre-cycle 1 (untreated), 24 hr Post Cycle 1 and Pre- Cycle 2 (15 days after initial cycle). Cycle in this case is each time a patient receives the drug.
Figure 4. Shows OX40L mRNA expression levels in melanoma (A375) and lung cancer (A549) cell lines treated with NUC-7738.

DETAILED DESCRIPTION OF THE INVENTION

Further features and embodiments of the above defined aspects are described herein below in headed sections. Each section is combinable with any of the above-mentioned aspects in any compatible combination.

Claims

1. Use of NUC-7738 in the manufacture of a medicament for the treatment of a proliferative disease in a patient, wherein the medicament enhances the patient's immune response against the proliferative disease.

2. The use according to claim 1, wherein the medicament reduces the amount of extra-cellular PD-L1 protein and/or increases the amount of OX40-L protein produced by the proliferative disease cells.

3. The use according to claim 2, wherein the extra-cellular PD-L1 protein is soluble PD-L1 and/or exosomal PD-L1.

4. The use according to any one of claims 1 to 3, wherein the treatment of the proliferative disease arises through adaptive immunity (e.g. cellular immunity and/or humoral immunity).

5. The use according to any one of the preceding claims, wherein the proliferative disease is cancer.

6. Use of NUC-7738 in the manufacture of a medicament for the treatment of cancer, wherein the medicament is an immune-sensitiser.

7. The use according to claim 5 or 6, wherein the cancer is selected from: melanoma (including cutaneous melanoma), lung cancer (including NSCLC), breast cancer, colorectal cancer, renal cancer, liver cancer, thyroid cancer, gastric cancer, pancreatic cancer, head and neck cancer, prostate cancer, bladder cancer, lymphoma, ovarian cancer, cervical cancer and endometrial cancer.

8. The use according to any one of the preceding claims, wherein the medicament is to be administered in combination with an immune oncology agent such as an immune checkpoint inhibitor, an antibody therapy, an adoptive cell therapy such as CAR-T therapy, or a cancer vaccine.

9. The use according to claim 8, wherein the immune checkpoint inhibitor is selected from a PD-1 inhibitor, a PD-L1 inhibitor, a LAG-3 inhibitor, CTLA-4 inhibitor, a TIM-3 inhibitor, a TIGIT inhibitor, an OX40 inhibitor, an OX40-L inhibitor or a BTLA inhibitor.

10. The use according to claim 8, wherein the immune checkpoint inhibitor is selected from the group consisting of: BMS-986016/relatlimab, TSR-033, REGN3767, MGD013 (bispecific DART binding PD-1 and LAG-3), GSK2831781, LAG525, MDX-010/ipilimumab, AGEN1884, and CP-675,206/tremelimumab, pembrolizumab, nivolumab, atezolizumab,

avelumab, durvalumab, cemiplimab, dostarlimab, MBG453, TSR-022, LY3321367, tiragolumab (MTIG7192A; RG6058), AB154, MK-7684, BMS-986207, ASP8374, MEDI6469. BMS-986178, SL-279252, INBRX-106 and cudarolimab, or a pharmaceutically acceptable salt or solvate thereof.

11. The use according to any one of the preceding claims, wherein the patient has previously received treatment with an immune checkpoint inhibitor, optionally wherein said treatment has been stopped due to toxicity, relapse or the cancer becoming resistant to the previous treatment.

12. The use according to any one of claims, wherein the treatment with NUC-7738 reduces soluble or exosomal PD-L1 levels by at least about 25% compared the levels obtained in the absence of NUC-7738 treatment.

13. A kit of parts which comprises:

a first container comprising NUC-7738, such as NUC-7738 in association with a pharmaceutically acceptable adjuvant, diluent or carrier; and

a second container comprising an immune oncology agent such as an immune checkpoint inhibitor in association with a pharmaceutically acceptable adjuvant, diluent or carrier, and

a container means for containing said first and second containers, optionally wherein the immune checkpoint inhibitor is pembrolizumab.

14. A method of determining whether a patient will benefit from treatment with NUC-7738, the method comprising: determining the level of soluble PD-L1 and/or exosomal PD-L1 and/or OX40-L in a biological sample from the patient; wherein if the level of soluble PD-L1 and/or exosomal PD-L1 in the biological sample is elevated and/or the level of OX40-L protein in the biological sample is reduced compared to a reference value the patient will benefit from treatment with NUC-7738.

15. Use of an immune checkpoint inhibitor in the manufacture of a medicament for the treatment of a proliferative disease such as cancer, wherein the immune checkpoint inhibitor is to be administered with NUC-7738, optionally wherein the immune checkpoint inhibitor and NUC-3378 are to be administered simultaneously or sequentially.

16. The use according to claim 15, wherein the immune checkpoint inhibitor is selected from the group consisting of: BMS-986016/relatlimab, TSR-033, REGN3767, MGD013 (bispecific DART binding PD-1 and LAG-3), GSK2831781, LAG525, MDX-010/ipilimumab,

AGEN1884, and CP-675,206/tremelimumab, pembrolizumab, nivolumab, atezolizumab, avelumab, durvalumab, MBG453, TSR-022, LY3321367, tiragolumab (MTIG7192A; RG6058), AB154, MK-7684, BMS-986207, ASP8374, MEDI6469. BMS-986178, SL-279252, INBRX-106 and cudarolimab, or a pharmaceutically acceptable salt or solvate thereof.

17. Use of NUC-7738 in the manufacture of a medicament for the treatment of cancer, wherein the medicament is to be used in combination with an immune oncology agent, such as an immune checkpoint inhibitor.

18. The use according to claim 17, wherein the immune oncology agent is selected from pembrolizumab, cemiplimab, dostarlimab, nivolumab, durvalumab, ipilimumab, atezolizumab, and avelumab.

19. The use according to claim 17 or 18, wherein the treatment is of a patient with the cancer and the patient has previously received treatment with an immune checkpoint inhibitor, optionally wherein said treatment has been stopped.