Full title: A parallel group phase I/II marker lesion study to assess the safety, tolerability and efficacy of intravenous or intravesical pembrolizumab in intermediate risk recurrent non-muscle invasive bladder cancer

Short title: PemBla - Pembrolizumab in intermediate risk recurrent non-muscle invasive bladder cancer (NMIBC).

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Conflict of Interest statement

None of the protocol authors have declared a potential conflict of interest

Confidentiality Statement

This document contains confidential information that must not be disclosed to anyone other than the Sponsor, the Trial Office, the Investigator Team, host NHS Trust(s), regulatory authorities, and members of the Research Ethics Committee unless authorised to do so.

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TABLE OF CONTENTS

PROTOCOL SYNOPSIS

Full Title of study:	A parallel group phase I/II marker lesion study to assess the safety, tolerability and efficacy of intravenous or intravesical pembrolizumab in intermediate risk recurrent non-muscle invasive bladder cancer	
Short Title:	Pembrolizumab in intermediate risk recurrent non-muscle invasive bladder cancer (NMIBC)	
Trial Acronym:	PemBla	
Clinical Phase:	Phase I/II	
Study Design:	A parallel group, open label, multi-centre, randomised, phase I/II marker-lesion study of intravesical or intravenous pembrolizumab. The main study will be preceded by a safety run-in with intra-patient dose escalation of intravesical pembrolizumab performed in paired patient cohorts to confirm the safety and tolerability of intravesical pembrolizumab and the dose to be used in the randomised phase.	
	Objectives	Endpoints
Primary:	To assess the safety, tolerability and toxicities of intravesical and intravenous pembrolizumab after TURBT in patients with intermediate risk NMIBC	Incidence and severity of adverse events (NCI CTCAE v4.03)
Secondary:	To provide a preliminary assessment of efficacy of treatment with intravesical pembrolizumab in patients with intermediate risk NMIBC	• Complete response rate of marker-lesion as assessed clinically at TURBT and confirmed on biopsy of tumour bed • Recurrence and progression-free intervals
	To provide a preliminary assessment of efficacy of treatment with intravenous pembrolizumab in patients with intermediate risk NMIBC	• Complete response rate of marker-lesion as assessed clinically at TURBT and confirmed on biopsy of tumour bed • Recurrence and progression-free intervals
Tertiary:	Refer to Section 3 Objectives and Endpoints for the list of Tertiary objectives and endpoints	
Planned enrolment:	Six patients will be enrolled in the safety-run in of intravesical pembrolizumab Thirty patients (fifteen in each of two arms) will be randomised 1:1 to treatment with either intravesical pembrolizumab (Arm A) or intravenous pembrolizumab (Arm B).	
Target Population:	Patients with recurrent intermediate risk NMIBC	
Investigational Medicinal Product(s)	Pembrolizumab Solution for infusion, 200mg, intravenous Pembrolizumab Solution for infusion, 50-200mg, intravesical	
Treatment Duration	Participants in the main study will be in the study for approximately 15 weeks from the time of first TURBT to the end of treatment visit. Participants in the safety-run in period will be in the study for approximately 11 weeks from the time of their TURBT to the end of treatment visit.	
Follow-up duration	Following the end of treatment visit, patients will receive standard care. Patients in the main study will be followed up via the medical care team for 2 years or until disease recurrence, progression or death whichever is sooner.	
End of study	Last Patient Last Visit, and up to 24 months for follow-up.	

SUMMARY SCHEDULE OF EVENTS

Visit Description	**Screening**	**TURBT**	**Pre-treatment**	**On treatment**									**TURBT**	**End of treatment visit**	**Follow-up**
Day	**-44 to -15**	**-14**	**-13 to 1**	**1**	**8**	**15**	**22**	**29**	**36**	**43**	**50**	**64**	**85** +/-7	**92**§ +7	
Informed consent	X														
Demographics & History	X														
Concomitant medication	X			X	X	X	X	X	X	X		X		X	
Physical examination	X			X	X	X	X	X	X	X		X		X	
Height and weight	X														
ECOG PS	X			X	X	X	X	X	X	X		X		X	
Vital signs[a]	X			X	X	X	X	X	X	X		X		X	
Blood for haem and biochem[b]	X			X	X	X	X	X	X	X		X		X	
Blood for TFTs[c]	X						X			X		X		X	
Urinalysis[d]	X			X	X	X	X	X	X	X		X		X	
Pregnancy test	X			X				X		X		X		X	
Blood for germline DNA[e]				X											
Blood for immunoprofiling[f]		X		X	X	X	X	X	X	X	X+	X	X		
Inclusion/exclusion criteria	X														
Resection of bladder tumour(s) TURBT		X									X+		X*		
Tumour assessment		X											X*		
Bladder barbotage		X									X+		X*		
Bladder biopsy[g]		X									X+		X*		
Registration	X														
Randomisation			X*												
AEs		X		X	X	X	X	X	X	X		X		X	
Urine for cytokines[h]				X	X	X	X	X	X	X	X	X	X		
Research Blood **[i]				X					X						
Administration of intravesical Pembrolizumab (Safety run-in and Arm A) [j]				X	X +/-1	X +/-1	X +/-1	X +/-1	X +/-1			X* +/-1			
Administration of intravenous Pembrolizumab (Arm B) [j]				X			X +/-3			X +/-3		X +/-3			
Recurrence data (RFI and PFI)															X
Optional tumour sample at recurrence or progression															X

Visits shaded in grey are only applicable to patients who are due for drug administration on that day

* For patients in the main study only (not patients in safety-run in)

** For patients receiving intravesical treatment only

[+] For patients who have consented to having second non-marker lesions resected only

[§] The end of study visit for patients in the safety run-in will be performed on day 64

[a] Vital signs to include systolic / diastolic blood pressure, pulse rate, respiratory rate, oxygen saturations and temperature. To be performed according to the vital signs scheduling details in section 8.3.

[b] To include FBC, clotting screen, sodium, potassium, urea, creatinine, calcium, phosphate, total protein, albumin, bilirubin, ALP, AST and/or ALT

[c] To include TSH, T3 and T4

[d] To include Blood, White Cells, Protein, Nitrites, pH, culture and sensitivity

[e] 10ml of blood in EDTA tube, taken pre-treatment

[f] 50ml of blood in Na-Heparin tubes

[g] Biopsy of normal bladder epithelium ± tumour bed if complete response of marker lesion

[h] For patients receiving intravesical treatment, urine for cytokines will be collected pre-dose and post-dose (first post-treatment urine sample after the drug has been voided from the bladder) on day 1 and thereafter post-dose only. For patients receiving intravenous treatment, urine for cytokines will be collected pre-dose only.

[i] Blood for research will be taken pre-dose and 2 hours post dose (counted from the completion of syringe/catheter administration of suspension into the bladder) for patients receiving intravesical treatments on day 1, and pre-dose only at cycle 6 (d36).

[j] Trial treatment should begin 14 days after TURBT or as close as possible to this date.

ABBREVIATIONS

ADR	Adverse Drug Reaction
AE	Adverse Event
BCG	Bacillus Calmette-Guerin
CIS	Carcinoma in situ
CR	Complete response
CRF	Case Report Form
CTA	Clinical Trials Authorisation
DKA	Diabetic Ketoacidosis
DLT	Dose limiting toxicity
EAU	European Association of Urologists
ECG	Echocardiogram
ECI	Event of Clinical Interest
EORTC	European Organisation for Research and Treatment of Cancer
FACS	Fluorescence-activated cell sorting
FFPE	Formalin-fixed, paraffin-embedded
HRA	Health Research Authority
HTA	Human Tissue Act
IB	Investigator Brochure
IEPTOC	Independent Early Phase Trial Oversight Committee
IMP	Investigational Medicinal Product
NMIBC	Non-muscle invasive bladder cancer
MMC	Mitomycin C
MA	Marketing Authorisation
MSD	Merck & Co., Inc
MTD	Maximum tolerated dose
PBMC	Peripheral blood mononuclear cells
PD	Pharmacodynamics
PD-1	Programmed cell death protein 1
PD-L1	Programmed death ligand 1
PD-L2	Programmed death ligand 2
PI	Principal Investigator
PFI	Progression-free Interval
REC	Research Ethics Committee
RFI	Recurrence-free interval
RSI	Reference safety information
SAE	Serious Adverse Event
SAR	Serious Adverse Reaction
SOP	Standard Operating Procedure
SPC	Summary of Product Characteristics
LPLV	Last visit of the last patient undergoing the trial
SUSAR	Suspected Unexpected Serious Adverse Drug Reaction
TCR	T cell receptor
TFT	Thyroid function test
TMG	Trial management group
TURBT	Transurethral resection of bladder tumour
ULN	Upper limit of normal

1 INTRODUCTION

1.1 Background

Bladder cancer is the seventh most common cancer in the UK and ninth most common cancer worldwide with 429,800 new cases diagnosed in 2012 [1]. Urothelial (transitional cell) carcinoma is the predominant histological subtype in Europe and the USA where it accounts for 90% of bladder cancers. In the UK in 2011, there were 10,399 people diagnosed with bladder cancer and 5,081 deaths from the disease [1]. It is 2–3 times more common in men than in women and the incidence is strongly related to age with 50% of cases occurring in people aged over 75 and 90% of cases occurring in people aged over 60. Approximately 70% of patients present with non-muscle invasive bladder cancer (NMIBC) that is confined to the mucosa (stage Ta or carcinoma in situ [CIS]) or submucosa (stage T1) [2].

Initial treatment of NMIBC consists of transurethral resection of the bladder tumour (TURBT). Without additional therapy, between 40-80% of tumours will recur within 6-12 months following TURBT and 10-25% will develop muscle invasive, regional or metastatic disease [3]. Instillation of a single dose of intravesical chemotherapy at the time of TURBT has been shown in meta-analyses to significantly reduce recurrence rates [4] and in practice mitomycin C (MMC) is commonly used. Subsequent management is guided by risk stratification based on the pathology results of the resected tumour. Patients can broadly be grouped into low, intermediate or high-risk according to the European Association of Urologists (EAU) 2013 guidelines [5] (Appendix 2), with individual risks of recurrence and progression being established using tables developed by EORTC [6] (Appendix 3). Further instillation of chemotherapy or immunotherapy (typically BCG) into the bladder may be performed for patients at intermediate or high risk.

Despite adjuvant treatment, bladder cancer has a propensity to recur and repeated resection of recurrences and further intravesical chemotherapy or immunotherapy, or in some cases cystectomy, may be required. As such, this has significant cost and quality of life implications but yet management of the disease has not significantly changed for over 20 years. New strategies for treating this disease are therefore urgently required to reduce recurrence and progression rates. Administration of intravesical BCG was first used in the treatment of bladder cancer in 1976 [7] and still forms the mainstay of adjuvant treatment for NMIBC. Although the exact mechanism of effect is not certain, it is generally accepted that it activates non-specific local immunity facilitating recruitment of activated T cells. Infiltration of the bladder with inflammatory cells is seen following treatment [8] accompanied by a significant urinary secretion of cytokines [9]. Whilst intravesical BCG has been shown to delay tumour progression [10], improve overall survival [11] and reduce tumour recurrence [12], failure rates of BCG treatment are up to 40% [13]. In addition it is associated with significant local and systemic side-effects and approximately 20% of patients discontinue treatment due to this [13]. Newer immunotherapeutic agents therefore provide a potentially attractive approach to the management of bladder cancer. This trial will evaluate the safety and efficacy of intravenous and intravesical administration of Pembrolizumab, a PD-1 inhibitor, in the setting of recurrent intermediate risk bladder cancer.

1.2 Rationale for the study

The importance of intact immune surveillance in controlling outgrowth of neoplastic transformation has long been recognised [14]. Bladder tumours are known to possess tumour specific antigens [15] and have high rates of somatic mutation [16], potentially increasing the number of antigens that can be identified by the immune system as foreign. Accumulating evidence shows a correlation between tumour-infiltrating lymphocytes (TILs) in cancer tissue and favourable prognosis in various malignancies including bladder cancer [17] [18] [19]. In particular, high intratumoural CD8 T cell density in urothelial cancers has been associated with better overall and disease specific survival [20] [21].

The PD-1 receptor-ligand interaction is a major pathway hijacked by tumours to suppress immune control. The normal function of PD-1, expressed on the cell surface of activated T-cells under healthy conditions, is to down-modulate unwanted or excessive immune responses, including autoimmune reactions. PD-1 (encoded by the gene *Pdcd1*) is an Ig superfamily member related to CD28 and CTLA-4 which has been shown to negatively regulate antigen receptor signalling upon engagement of its ligands (PD-L1 and/or PD-L2) [22]. PD-1 is expressed on activated lymphocytes including peripheral CD4+ and CD8+ T cells, B-cells, Natural Killer cells, activated monocytes and dendritic cells. Binding of either PD-1 ligand to PD-1 inhibits T-cell activation triggered through the T-cell receptor. Whilst normal human tissues express little PD-L1, a variety of cancers have been shown to express abundant levels of this T-cell inhibitor [23]. Evidence to suggest that the PD-1/PD-L1 pathway may play a critical role in tumour immune evasion has led this to being evaluated as a target for therapeutic intervention in a number of cancers.

Pre-clinical

Studies performed in a range of preclinical models have consistently shown the importance of the PD-1 and PD-L1 interaction and potential anti-tumour effects that can be achieved by blocking this interaction. In mouse models, expression of PD-L1 rendered tumour cells less susceptible to the effects of cytotoxic T cells in vitro and enhanced their invasiveness in vivo, effects that were reversed by anti-PD-L1 Ab [24]. In PD-1 deficient mice, haematogenous spread of melanoma cells was inhibited by PD-1 blockade by enhancing infiltration of tumour-specific T cells and augmenting cytotoxicity [25]. Substantial anti-tumour effects were also shown in vivo in a murine pancreatic cancer model, with PD-L1 blockade promoting CD8+ T cell infiltration into the tumour and inducing local immune activation [26]. In human models, PD-1/PD-L1 blockade has been shown to increase numbers of cytotoxic T lymphocytes in the tumour [27], and enhance cytolytic activity of tumour specific cytotoxic T lymphocytes [28].

Clinical

Targeting the PD-1/PD-L1 pathway has delivered impressive results in trials in a number of malignancies, most notably melanoma and is now becoming an accepted treatment approach. In an early phase study of 135 patients with advanced melanoma, anti-PD1 treatment produced an overall response rate of 38%, with many of the responses being durable [29]. Further studies in melanoma have produced similar results [30] and trials in non-small cell lung cancer [31] [32], renal cell cancer [33] and head and neck cancer [34] have also shown good responses. Initial studies of PD-L1 inhibition in metastatic urothelial cancer have also shown promising results. In a recent Phase I study [35] patients received MPDL3280A, an anti PD-L1 antibody, at a dose of 15 mg/kg intravenously every three weeks for up to a year. MPDL3280A was demonstrated to be well tolerated in this pre-treated population with 54% of the patients treated responding to treatment. However, since only 20% of bladder cancers appear to express PD-L1 [20], treatment with anti-PD-1 antibodies may be effective in a larger proportion of patients, by targeting the interaction of PD-1 with both PD-L1 and PD-L2. Injection of the anti-PD-1 antibody, Pembrolizumab (10 mg/kg intravenously every two weeks), in a population of 33 previously treated urothelial patients gave an overall response of 24%, with acceptable safety and tolerability profile [36]. Fatigue, rash and diarrhoea were the most common drug-related AEs seen and were mostly grade 1-2.

Whilst systemic treatment has not historically been used for treatment of NMIBC due to the limited duration of cytotoxic action of chemotherapy agents and associated toxicity, the potentially durable anti-tumour response generated by anti-PD1 treatments and acceptable toxicity profile creates the possibility for new systemic approaches to treatment that warrant further investigation. In addition, targeted administration via intravesical injection could potentially improve delivery of anti-PD-1 to the tumour site, leading to better clinical responses while reducing toxicity due to systemic administration. Previous studies of intravesical injection of radiolabelled antibodies directed against tumour-associated antigens have successfully showed retention of antibodies within the tumour mass [37], suggesting that this route of delivery is feasible. This study will therefore assess both intravenous and intravesical anti-PD1 antibody treatment in the setting of recurrent NMIBC. The safety of this treatment strategy will be evaluated and a marker lesion approach, described in section 1.4, used to provide an indication of activity. Should response rates meet predefined criteria, larger phase II trials would be developed to further assess the utility of this treatment strategy in NMIBC.

1.3 Investigational Medicinal Product(s) used in the study

Pembrolizumab (previously known as MK-3475) is licensed both in the US and EU at a dose of 2mg/kg every three weeks for the treatment of patients with unresectable or metastatic melanoma. It is a potent and highly selective humanised monoclonal antibody (mAb) of the IgG4/kappa isotype designed to directly block the interaction between PD-1 and its ligands, PD-L1 and PD-L2, releasing PD-1 pathway-mediated inhibition of the immune response. Pembrolizumab also modulates the level of interleukin-2 (IL-2), tumor necrosis factor alpha (TNFα), interferon gamma (IFNγ), and other cytokines. The antibody potentiates existing immune responses only in the presence of antigen and does not nonspecifically activate T-cells.

Various trials have evaluated the safety and clinical efficacy of Pembrolizumab at doses ranging from 1mg/kg to 10mg/kg and administration schedules of every 2 weeks (Q2W) and every 3 weeks (Q3W). No maximum tolerated dose (MTD) has been identified to date and there is evidence of target engagement and objective evidence of tumour size reduction at all dose levels with no clear dose response relationship. Pharmacodynamic data (IL-2 release assay) suggest that peripheral target engagement is durable (>21 days) and pharmacokinetic data analyses show slow systemic clearance, limited volume of distribution and a long half-life (26 days).

Choice of dose

Based on pharmacokinetic analysis from previous studies, for the intravenous arm of the study we will utilise a fixed dose regimen of 200mg Q3W. This has been shown to provide exposures that are consistent with those obtained with a 2mg/kg dose Q3W, which will maintain individual patient exposures in the range established in melanoma as being efficacious whilst also being well tolerated and safe. In addition, utilising a fixed dose regimen will be more convenient for physicians and reduce the potential for dosing errors and wastage.

For the intravesical component of the study we will utilise a once weekly dosing schedule over a period of 6 weeks, as is convention for intravesical therapy. A further maintenance intravesical injection will be given on day 64. Systemic absorption of drugs from the bladder is related to a number of factors with a molecular weight below 300kDa being one major determinant[38]. The approximate molecular weight of Pembrolizumab is 149kDa. As such, a maximum intravesical dose of 200mg will be used to ensure acceptable tolerability should systemic absorption occur. Should the entire dose be systemically absorbed, when administered once weekly the cumulative dose will remain below the 10mg/kg level that has previous been shown to be tolerated when given intravenously on a Q2W schedule.

1.4 Other research interventions

Marker lesion studies

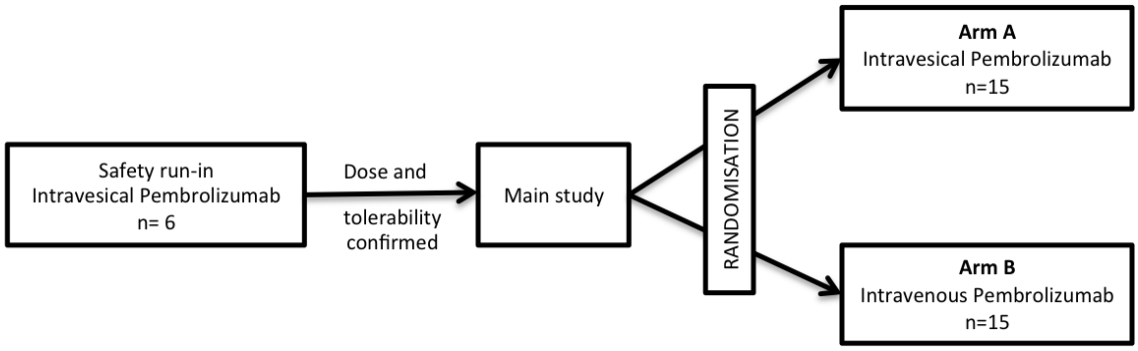
In an effort to develop new, more effective treatments for NMIBC, a number of novel agents have been trialed using marker-lesion studies. In these studies, a single bladder tumour is left unresected whilst the patient receives further intravesical treatment. The bladder is then re-assessed after an interval of time for response of the marker lesion to the treatment, with completion TURBT at that time if the marker lesion persists. By identifying agents that have the ability to eliminate macroscopic disease, drugs that have the potential to eradicate subclinical microscopic disease that is likely to contribute to recurrent disease can be identified for further evaluation. Using a marker-lesion approach enables much more rapid evaluation of agents in a substantially smaller number of patients than would be required to obtain this information from an adjuvant study. The CR rate seen in most marker lesion studies is between 30-50% and it has been shown that primary tumours have a better CR rate (67%) than recurrent tumours (37%) and untreated tumours respond better than those that are pretreated (55% compared with 31% respectively)[39].

Studies using marker lesions including over 1200 patients have been shown to be safe with progression to T2 disease seen in only 7 patients (0.6%), all of whom had high-risk (G3) tumours [39]. No cases of progression have been seen where patients with only intermediate-risk disease have been recruited. As such, patients who are appropriate for marker-lesion studies are those with multiple recurrent intermediate-risk bladder tumours. Given the lack of evidence for the optimum management strategy for these patients, they may potentially benefit from participation in such studies without significantly increased risk. Patients with recurrent high-risk disease may consider radical cystectomy or should undergo repeat TURBT and best available therapy and should therefore not be considered.

Most agents assessed in marker lesion studies have shown low toxicity profiles with mild irritative urinary symptoms being the most common complaint occurring in up to half of the patients [39]. Given the utility and safety of marker-lesion studies for the assessment of intravesical agents, extrapolation of this approach to the assessment of systemic treatments represents a novel but logical progression.

2 TRIAL DESIGN

This is a parallel group, open label, multi-centre, phase I/II marker-lesion study in recurrent intermediate risk NMIBC. Thirty patients (fifteen in each of two arms) will be randomised 1:1 to treatment with either intravesical pembrolizumab (Arm A) or intravenous pembrolizumab (Arm B). The main study will be preceded by a safety run-in phase involving intra-patient dose escalation in six patients to confirm the safety and tolerability of intravesical pembrolizumab and the dose to be used in the randomised phase.



Safety run-in

The safety and tolerability of intravesical Pembrolizumab will be evaluated in six patients following TURBT for NMIBC prior to the main study commencing. Treatment will be administered once weekly according to the following schedule. Intra-patient dose escalation of intravesical Pembrolizumab will be performed in paired patient cohorts with the anticipated doses as shown below.

Figure 1

Cohort	Patient	Dose of Intravesical Pembrolizumab (mg)					
		Day 1	Day 8	Day 15	Day 22	Day 29	Day 36
1	1	50	50	100	100	200	200
	2	50	50	100	100	200	200
2	3	100	100	200	200	200	200
	4	100	100	200	200	200	200
3	5	200	200	200	200	200	200
	6	200	200	200	200	200	200

Patients will be assessed for dose limiting toxicities (DLT) as well as for overall tolerability of the treatment. A DLT is defined as a clinically significant, drug related, grade 4 haematological or $\geq$ grade 3 non-haematological toxicity occurring within 7 days of administration of the first treatment at a given dose for that patient. Treatment start dates will be staggered by at least one week between patients. In addition, treatment of patients in cohort 2 and 3 will not commence until both patients in the preceding cohort have cleared the DLT period for the D15 dose.

If more than 1 patient experiences a DLT at a certain dose, this dose will be declared non-tolerated and further escalation will cease. Patients who have already commenced treatment at the same dose and are tolerating treatment may continue at this dose. If a DLT occurs in either patient at the first dose of cohort 1, or in more than 1 patient at subsequent doses, the TMG will meet to consider the available safety data and determine how to proceed. Recruitment of further patients to a cohort or investigation of intermediate doses or treatment intervals may be considered.

When considering the dose to take forward to the randomised part of the study the TMG will also consider the overall tolerability of the treatment. Patients must be able to receive at least 5 out of the 6 treatments for the regime to be defined as tolerable.

Main study

Once the TMG have considered the available safety data from the safety run-in cohort and confirmed the tolerated intravesical dose, the main study will open to recruitment, provided the TMG is in agreement. The TMG decision will

be documented in writing, and a record will be retained in the Trial Master File. Patients will be randomised to receive either intravesical or intravenous pembrolizumab treatment post TURBT, having had a suitable marker lesion left in situ. For at least five patients in each cohort, a second non-marker lesion will be left in-situ at the time of TURBT that will be resected on day 50.

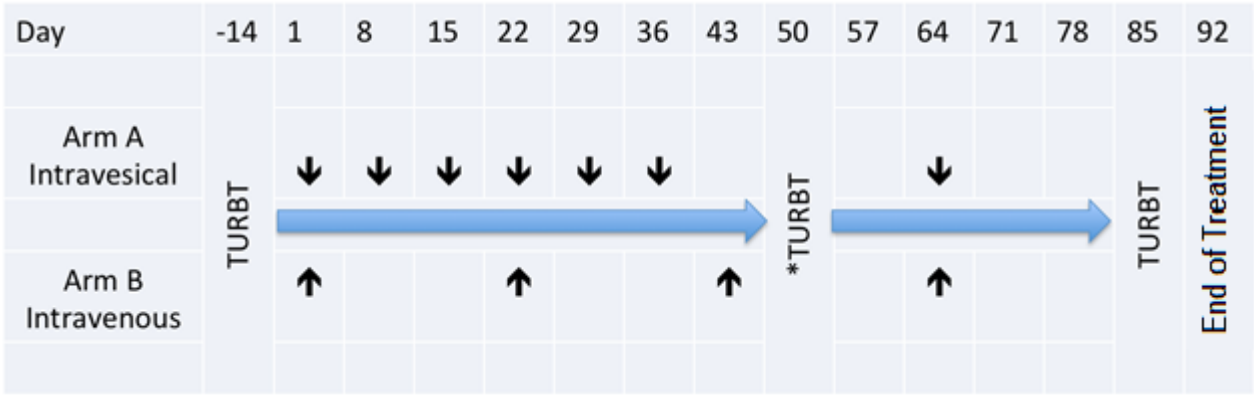


Figure 2: Schedule of treatment administration for main study. *TURBT for resection of second non-marker lesion in at least 5 patients in each arm. Black arrows denote treatment administration

Please refer to the schedule of events and flow chart for details of the study visits and procedures.

2.1 Duration of patient participation

Participants in the main study will be in the study for approximately 15 weeks from the time of first TURBT to the end of treatment visit. Participants in the safety-run in period will be in the study for approximately 11 weeks from the time of their TURBT to the end of treatment visit.

2.2 Post-trial care and follow-up

Following the end of treatment visit, patients will receive standard care. However, patients in the main study will be followed up via the medical care team for 2 years or until disease recurrence, progression or death, whichever is sooner.

3 OBJECTIVES AND ENDPOINTS

Primary Objective	**Endpoints/ Outcome measures**
• To assess the safety, tolerability and toxicities of intravesical and intravenous pembrolizumab after TURBT in patients with intermediate risk NMIBC	• Incidence and severity of adverse events (NCI CTCAE v4.03)
Secondary Objectives	**Endpoints**
• To provide a preliminary assessment of efficacy of treatment with intravesical pembrolizumab in patients with intermediate risk NMIBC	• Complete response rate of marker-lesion as assessed clinically at TURBT and confirmed on biopsy of tumour bed • Recurrence and progression-free intervals
• To provide a preliminary assessment of efficacy of treatment with intravenous pembrolizumab in patients with intermediate risk NMIBC	• Complete response rate of marker-lesion as assessed clinically at TURBT and confirmed on biopsy of tumour bed • Recurrence and progression-free intervals
Tertiary/Exploratory Objectives	**Endpoints**
• Determine correlation between expression of PD-L1 and PD-1+ infiltrating	• Tumour/stromal cell PD-L1 expression and presence of PD-1+ infiltrating

lymphocytes and efficacy of pembrolizumab therapy after TURBT in intermediate risk NMIBC patients	lymphocytes in pre, on and post treatment tumour samples measured by immunohistochemistry and FACS analysis
• Definition of gene expression signatures and genetic profiles capable of predicting efficacy of pembrolizumab treatment in NMIBC patients.	• Gene expression profiling and DNA sequencing on pre-treatment blood and tumour samples
• To evaluate the effects of pembrolizumab treatment on the immunological profile and tumour specific immune responses in patients with intermediate risk NMIBC	• Analysis of TCR repertoire and clonality of infiltrating T cells in resected tumour specimens, urine and normal bladder tissue
• Identification of myeloid or T cell responses in the tumour microenvironment associated with response to treatment	• Analysis of TCR repertoire and clonality of PBMC before, during and after treatment • Analysis of cytokines in blood and urine
• To investigate the pharmacokinetics of intravesical pembrolizumab	• Analysis of blood levels of pembrolizumab during treatment

4 PATIENT SELECTION

Written informed consent must be obtained before any study specific procedures are performed. The Investigator will determine patient eligibility based on the following criteria. Participants must satisfy the eligibility criteria prior to registration, randomisation and pre-dose at Day 1.

4.1 Eligibility criteria

In order to be eligible for participation in this trial, the subject must:

1. Be willing and able to provide written informed consent for the trial and comply with the protocol scheduled follow-up visits and examinations for the duration of the study
2. Be ≥ 18 years of age on day of signing informed consent
3. Have recurrent NMIBC for which adjuvant treatment post TURBT would be a reasonable treatment option
4. Main study only:
    a. Have recurrent, multiple (minimum 2) tumours consistent with NMIBC
    b. Have at least one lesion of between 5-10mm in size clinically that can be left un-resected at TURBT as the marker lesion
    c. Have histologically confirmed low grade transitional cell NMIBC at original and any subsequent diagnosis
5. Have a normal upper urinary tract (as evidenced by ultrasound or CT urography within 2 years prior to randomisation) and no evidence of tumour in prostatic urethra at flexible cystoscopy
6. Have a performance status of 0 or 1 on the ECOG Performance Scale
7. Have adequate organ function as defined in the table below:

Lab Test	Value required
Haemoglobin (Hb)	≥ 9 g/dL without transfusion or EPO dependency
Absolute neutrophil count (ANC)	≥ 1.5 x 10^9/L
Platelet count	≥ 100 x 10^9/L
Total bilirubin	≤ 1.5 times the upper limit of normal (ULN) or direct bilirubin ≤ ULN for subjects with total bilirubin levels > 1.5 x ULN
Serum alanine aminotransferase (ALT) and/or serum aspartate aminotransferase (AST)	≤ 2.5 x ULN

Serum creatinine OR Measured or calculated creatinine clearance[a]	≤ 1.5 x ULN OR ≥ 60ml/min for subject with creatinine levels > 1.5 x institutional ULN
Albumin	≥ 25g/L
International Normalized Ratio (INR) or Prothrombin Time (PT) and Activated Partial Thromboplastin Time (aPTT)	≤ 1.5 x ULN unless subject is receiving anticoagulant therapy as long as PT or APTT is within therapeutic range of intended use of anticoagulants
[a] Creatinine clearance should be calculated as per institutional standard	

8. Female subjects of childbearing potential should have a negative urine or serum pregnancy test at screening and within 72 hours prior to receiving the first dose of study medication. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required
9. Both male and female subjects of childbearing potential (Section 5.1) must be willing to use an adequate method of contraception as outlined in section 5.1 for the course of the study and until 120 days after the last dose of the study medication

4.2 Exclusion criteria:

The subject must be excluded from participating in the trial if the subject:

1. Has received prior radiotherapy to the pelvis
2. Has significant urinary incontinence or known bladder instability
3. Main study only:
    a. Has more than 2 out of 3 of the following present at the current time:
        i. ≥8 tumours
        ii. Tumour ≥3cm in size
        iii. Frequent recurrence (>1/year)
    b. Has a previous history of any of the following: T1 tumour, high grade/G3 tumour, carcinoma in situ, multiple recurrent large (>3cm) Ta, G1 or G2 tumours.
    c. Had a primary tumour of unknown pathological stage or grade
    d. Has disease for which resection of all visible tumours is not possible
4. Is currently participating and receiving study therapy or has participated in a study of an investigational agent and received study therapy or used an investigational device within 28 days of the first dose of trial treatment
5. Has a diagnosis of immunodeficiency or is receiving systemic steroid therapy or any other form of immunosuppressive therapy within 7 days prior to the first dose of trial treatment. Subjects requiring use of inhaled or intranasal corticosteroids or local steroid injections would not be excluded
6. Has a known history of active TB
7. Has received intravesical BCG treatment within 30 days prior to the first dose of trial treatment
8. Has hypersensitivity to pembrolizumab or any of its excipients
9. Has had treatment with any other anti-cancer monoclonal antibody within 28 days prior to enrolment or who has not recovered (i.e. ≤ Grade 1 or at baseline) from adverse events due to agents administered more than 4 weeks earlier
10. Has had treatment with prior chemotherapy, targeted small molecule therapy, or radiation therapy within 2 weeks of administration of study drug or who has not recovered (i.e. ≤ Grade 1 or at baseline) from adverse events due to a previously administered agent.
11. Has a known additional malignancy that is progressing or requires active treatment. Exceptions include basal cell carcinoma of the skin or squamous cell carcinoma of the skin that has undergone potentially curative therapy or in situ cervical cancer
12. Has known active central nervous system (CNS) metastases and/or carcinomatous meningitis. Subjects with previously treated brain metastases may participate provided they are stable (without evidence of progression by imaging for at least four weeks prior to the first dose of trial treatment and any neurological symptoms have returned to baseline), have no evidence of new or enlarging brain metastases, and are not using steroids for a t least 7 days prior to trial treatment. This exception does not include carcinomatous meningitis which is excluded regardless of clinical stability

13. Has active autoimmune disease that has required systemic treatment in the past 2 years (i.e. with use of disease modifying agents or immunosuppressive drugs). Replacement therapy (e.g. thyroxine, insulin or physiologic corticosteroids replacement therapy for adrenal or pituitary insufficiency, etc.) is not considered a form of systemic treatment
14. Has a known history of, or any evidence of active, non-infectious pneumonitis
15. Has an active or intractable infection requiring systemic therapy
16. Has a history or current evidence of any condition, therapy, or laboratory abnormality that might confound the results of the trial, interfere with the subject's participation for the full duration of the trial, or is not in the best interest of the subject to participate, in the opinion of the treating Investigator
17. Has a known psychiatric or substance abuse disorder that would interfere with cooperation with the requirements of the trial
18. Is pregnant or breastfeeding, or expecting to conceive or father children within the projected duration of the trial, starting with the pre-screening or screening visit through to 120 days after the last dose of trial treatment
19. Has received prior therapy with an anti-PD-1, anti PD-L1, or anti-PD-L2 agent
20. Has a known history of Human Immunodeficiency Virus (HIV) (HIV 1/2 antibodies)
21. Has known active Hepatitis B (e.g. HBsAg reactive) or Hepatitis C (e.g. HCV RNA [qualitative] is detected)
22. Has received a live vaccine within 30 days prior to the first dose of trial treatment.

4.3 Protocol deviations and waivers to entry criteria

Protocol adherence is a fundamental part of the conduct of a clinical study. Changes to the approved protocol need prior approval unless for urgent safety reasons.

Investigators must contact OCTO to obtain guidance and/or clarification as necessary if unsure whether the patient satisfies all the entry criteria and to clarify matters of clinical discretion. OCTO will contact the Chief Investigator or clinical coordinators as necessary. Investigators should not request a protocol waiver to enter a patient who does not satisfy the selection criteria.

The Investigator must document and explain any deviations/violations from the approved protocol. The Investigator should promptly report any important violations that might impact patient safety, data integrity or be a possible serious breach (see 21.7 below) to the Trial Office.

4.4 Re-screening if patient does not meet inclusion/exclusion criteria first time round

If a patient does not meet the laboratory haematological and biochemical inclusion/exclusion criteria the first time round, he/she can be re-screened within the 30-day screening period and does not need to be reconsented. Patients who fail at rescreening or who fail any other criteria first time round are ineligible for this recurrence episode but may be rescreened at the time of any subsequent recurrence; in these circumstances, reconsent should be sought.

4.5 Patient registration and randomisation procedure

Potential participants will be identified by their clinical care teams following a diagnosis of recurrent NMIBC at the time of flexible cystoscopy under local anaesthetic, prior to admission for surgical management. Discussion at multidisciplinary (MDT) meetings will aid identification of potential participants. Patients for whom PEMBLA is a suitable option will be approached and given time to consider participation.

A screening log must be kept of <u>all patients</u> considered for the study including any that are subsequently excluded; the reason for exclusion must be recorded on this form. A copy of the screening log should be sent to the Trial Office on request, but without patient identifiers. The original must be retained on site.

Patients will be registered & randomised to the trial in a two part process. Before registering/randomising a patient to the study treatment, the Principal Investigator or designee will confirm eligibility. If in any doubt the Chief Investigator must be consulted before entering the patient. Details of the query and outcome of the decision must be documented on the registration/ eligibility checklist.

4.5.1 Patient Registration (Safety Run-in and Main Study)

The Day -14 TUBRT must not take place until the registration process is complete.

To register a patient to the trial, the site must email the completed Registration Form to the Trial Office: octo-pembla@oncology.ox.ac.uk The patient will then be registered to the trial and assigned a unique trial number by the Trial Office. Patients in the Safety run-in will also be assigned to the appropriate treatment cohort. The site will be informed by email of the patient's unique trial number (and treatment cohort if applicable) by the Trial Office.

The patient's trial number must be specified on all CRFs and SAEs forms submitted for the patient and in all relevant correspondence with the Trial Office.

4.5.2 Patient Randomisation (Main Study Only)

Cycle 1 Day 1 Trial Treatment must not start until this randomisation process is complete, and should begin 14 days after TURBT, and within 7 days of randomisation or as close as possible to this date.

To randomise a patient to the trial, once the TUBRT procedure has taken place and continued eligbility for the trial is confirmed, the site must email the completed Randomisation Form to the Trial Office: octo-pembla@oncology.ox.ac.uk The patient will then be randomised to either Arm A or Arm B trial treatment..

4.5.3 Safety run in phase

The first six patients will not be randomised and will receive intravesical pembrolizumab as part of the safety run-in phase.

4.5.4 Randomisation

Randomisation into the study will only begin after the trial management group has considered the safety analysis of the run-in cohort. Eligible patients will be randomised 1:1 to receive intravenous pembrolizumab or intravesical pembrolizumab following TURBT. Minimisation will be performed by study centre.

5 TRIAL ASSESSMENTS AND PROCEDURES

Please refer to the Schedule of Investigations given at the front of this protocol. Details of all protocol evaluations and investigations must be recorded in the patient's medical record for extraction onto the CRF.

5.1 Informed consent

Potential participants will be given a current, approved version of the patient information sheet and consent form. They will also receive clear verbal information about the study detailing no less than: the nature of the study; the implications and constraints of the protocol; the known side effects and any risks involved in taking part. It will be explained that they will be free to withdraw from the study at any time, for any reason, without prejudice to future care, and with no obligation to give a reason for withdrawal. They will have at least 24 hours to consider the information provided and the opportunity to question the Investigator, their GP or other independent parties before deciding whether to participate.

The Investigator who obtains consent must be suitably qualified and experienced. All delegates must be authorised by the Chief/Principal Investigator to obtain consent. The Investigator is responsible for ensuring that the trial consent procedures comply with current applicable GCP, Regulatory and Ethical requirements. Informed consent discussions and outcomes must be well documented in the medical record. The Investigator must be satisfied that the patient has made an informed decision before taking consent. The patient and the Investigator must personally sign and date the current approved version of the informed consent form in each other's presence. A copy of the information and signed consent form will be given to the participant. The original signed consent form will be retained at the trial site, with copies held in both the medical record and Investigator Site File (ideally the original if local policy permits).

Contraceptive/ Pregnancy counselling

Pembrolizumab may have adverse effects on a foetus in utero. Furthermore, it is not known if pembrolizumab has transient adverse effects on the composition of sperm.

For this trial, male subjects will be considered to be of non-reproductive potential if they have azoospermia (whether due to having had a vasectomy or due to an underlying medical condition).

Female subjects will be considered of non-reproductive potential if they are either:

(1) postmenopausal -defined as at least 12 months with no menses without an alternative medical cause; in women < 45 years of age, a high follicle stimulating hormone (FSH) level in the postmenopausal range may be used to confirm a post-menopausal state in women not using hormonal contraception or hormonal replacement therapy. In the absence of 12 months of amenorrhea, a single FSH measurement is insufficient.

OR

(2) have had a hysterectomy and/or bilateral oophorectomy, bilateral salpingectomy or bilateral tubal ligation/occlusion, at least 6 weeks prior to screening;

OR

(3) has a congenital or acquired condition that prevents childbearing.

Female and male subjects of reproductive potential must agree to avoid becoming pregnant or impregnating a partner, respectively, while receiving study drug and for 120 days after the last dose of study drug by complying with one of the following:

(1) practice abstinence[†] from heterosexual activity;

OR

(2) use (or have their partner use) acceptable contraception during heterosexual activity.

Acceptable methods of contraception are[‡]:

Single method (one of the following is acceptable):

- intrauterine device (IUD)
- vasectomy of a female subject's male partner
- contraceptive rod implanted into the skin

Combination method (requires use of two of the following):

- diaphragm with spermicide (cannot be used in conjunction with cervical cap/spermicide)
- cervical cap with spermicide (nulliparous women only)
- contraceptive sponge (nulliparous women only)
- male condom or female condom (cannot be used together)
- hormonal contraceptive: oral contraceptive pill (oestrogen/progestin pill or progestin-only pill), contraceptive skin patch, vaginal contraceptive ring, or subcutaneous contraceptive injection

†Abstinence (relative to heterosexual activity) can be used as the sole method of contraception if it is consistently employed as the subject's preferred and usual lifestyle and if considered acceptable by local regulatory agencies and ERCs/IRBs. Declaration of abstinence for the duration of exposure to the IMP, periodic abstinence (e.g., calendar, ovulation, sympto-thermal, post-ovulation methods, etc.) and withdrawal are not acceptable methods of contraception.

‡If a contraceptive method listed above is restricted by local regulations/guidelines, then it does not qualify as an acceptable method of contraception for subjects participating at sites in this country/region.

Subjects should be informed that taking the study medication may involve unknown risks to the foetus (unborn baby) if pregnancy were to occur during the study. In order to participate in the study, subjects of childbearing potential must adhere to the contraception requirement (described above) from the day of study medication initiation (or 14 days prior to the initiation of study medication for oral contraception) throughout the study period and for 120 days after the last dose of trial drug. If there is any question that a subject of childbearing potential will not reliably comply with the requirements for contraception, that subject should not be entered into the study.

Use in Pregnancy
If a subject inadvertently becomes pregnant while on treatment with pembrolizumab, the subject will immediately be removed from the study. The site will contact the subject at least monthly and document the subject's status until the pregnancy has been completed or terminated. The outcome of the pregnancy will be reported to the Trial Office without delay and within 24 hours if the outcome is a serious adverse event (e.g., death, abortion, congenital anomaly, or other disabling or life-threatening complication to the mother or newborn).
The study Investigator will make every effort to obtain consent to follow the outcome of the pregnancy and report the condition of the foetus or newborn to the Trial Office. If a male subject impregnates his female partner, the study personnel at the site must be informed immediately and the pregnancy reported to the Trial Office as described above and in Section 13.

5.2 Screening evaluations

For all parts of the study the following screening and eligibility assessments must be performed/obtained within the 30 days before the patient undergoes TURBT:

- Written informed consent
- Demographic details including age, sex, self-reported race/ethnicity, history of occupational exposure to aromatic amines
- Medical History to include cancer history, prior cancer therapies and procedures, smoking history, clinically significant past medical history and concomitant medical conditions
- Concomitant medications
- Physical examination
- Height and weight
- ECOG performance status
- Vital signs: systolic / diastolic blood pressure (BP), pulse rate, respiratory rate, oxygen saturations and temperature
- Haematology – Full blood count (FBC), clotting screen
- Biochemistry – sodium, potassium, urea, creatinine, calcium, phosphate, total protein, albumin, bilirubin, alkaline phosphatase (ALP), AST and/or ALT, thyroid function tests (TSH, T3 and T4)
- Urinalysis (Blood, White Cells, Protein, Nitrites, pH), culture and sensitivity
- Pregnancy test (for female subjects of childbearing potential only): serum or urine Human Chorionic Gonadotropin (HCG) test. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required
- Overall assessment of patient eligibility according to inclusion/exclusion criteria

5.3 Evaluations during the study

Day -14 - TURBT
A cystoscopy and TURBT will be performed on day -14 of the study in accordance with local hospital protocols. This should be performed no later than 30 days following diagnosis of recurrent NMIBC; if outside of this window, please contact the PemBla Trial Office for advice. The following will also be performed:

- Blood for immunoprofiling
- Bladder barbotage (instillation and recovery of 4 x 50ml instillations of normal saline into the bladder) prior to resection
- A biopsy of the normal bladder epithelium, to be sent to the central laboratory for research purposes.

In addition, for patients in the Safety Run-in Study only:

- All tumour(s) should be resected and a sample sent to the local laboratory for histological review and grading. Any tissue excess to diagnostic requirements will be sent to the central laboratory for immunoprofiling, gene expression profiling and DNA sequencing.

In addition, for patients in the Main Study only - all patients:

- At the time of TURBT an appropriate marker lesion should be selected. This should be a solitary lesion measuring between 5-10mm which can be left un-resected without being disturbed/touched during

resection of the other lesions. If this is not technically possible all tumours should be resected and the patient excluded from the study.

- The size, location and morphology of the marker lesion should be documented in a bladder diagram. Where possible, photographs should be taken. Other non-marker lesion tumours should also be documented on the bladder diagram.
- For patients with 3 or more tumours who have consented to having a second non-marker lesion left in-situ, this should be identified, recorded on the bladder diagram and left un-resected.
- All other lesions should be resected. A sample of one of the resected tumours should be sent to the local laboratory for tumour grading and fixation for subsequent PD-1/PD-L1 testing. All other resected tumour samples will be sent to the central laboratory for immunoprofiling, gene expression profiling and DNA sequencing.

Day -13 - Day-1

During this period, the pathology results from the resected tumours should be reviewed. For the main study only, if any of the tumours are found to be high grade/G3 or CIS, the patient will be excluded from further participation in the study and arrangements should be made to resect the marker lesion and any other non-marker lesion that was left at the earliest opportunity. Subject to on-going eligibility, the patient will be randomised to either intravesical or intravenous Pembrolizumab.

Evaluations on day 1 (All patients)

- Baseline AEs
- Review of concomitant medications
- Physical examination
- ECOG performance status
- Vital signs - systolic / diastolic BP, pulse rate, respiratory rate, oxygen saturations and temperature
- Haematology – Full blood count (FBC), clotting screen
- Biochemistry – sodium, potassium, urea, creatinine, calcium, phosphate, total protein, albumin, bilirubin, alkaline phosphatase (ALP), AST and/or ALT
- Blood for germline DNA
- Urinalysis (Blood, White Cells, Protein, Nitrites, pH), culture and sensitivity
- Pregnancy test (for female subjects of childbearing potential only): serum or urine Human Chorionic Gonadotropin (HCG) test. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required
- Blood samples for immunoprofiling
- Blood samples for research assessment (intravesical arm only) pre-dose and 2hr post dose
- Urine sample for cytokines - Intravesical patients pre-dose and post-dose sample (after the drug has been voided from the bladder) will be collected (visits thereafter only post-dose will be collected). For intravenous treatment patients, urine for cytokines collected pre-dose only.
- Administration of study agent and monitoring for any immediate adverse events

Evaluations on day 8 and 15 (Patients receiving intravesical treatment only)

- AEs
- Review of concomitant medications
- Physical examination
- ECOG performance status
- Vital signs - systolic / diastolic BP, pulse rate, respiratory rate, oxygen saturations and temperature
- Haematology – Full blood count (FBC), clotting screen
- Biochemistry – sodium, potassium, urea, creatinine, calcium, phosphate, total protein, albumin, bilirubin, alkaline phosphatase (ALP), AST and/or ALT
- Urinalysis (Blood, White Cells, Protein, Nitrites, pH), culture and sensitivity
- Blood samples for immunoprofiling
- Urine sample for cytokines – post dose only
- Administration of study agent and monitoring for any immediate adverse events

Evaluations on day 22 (Patients receiving intravenous or intravesical treatment)

- AEs
- Review of concomitant medications
- Physical examination
- ECOG performance status
- Vital signs - systolic / diastolic BP, pulse rate, respiratory rate, oxygen saturations and temperature
- Haematology – Full blood count (FBC), clotting screen
- Biochemistry – sodium, potassium, urea, creatinine, calcium, phosphate, total protein, albumin, bilirubin, alkaline phosphatase (ALP), AST and/or ALT, thyroid function tests (TSH, T3 and T4)
- Urinalysis (Blood, White Cells, Protein, Nitrites, pH), culture and sensitivity
- Blood samples for immunoprofiling
- Urine sample for cytokines - Intravenous patients pre-dose only, Intravesical post-dose only.
- Administration of study agent and monitoring for any immediate adverse events

Evaluations on day 29 and 36 (Patients receiving intravesical treatment only)

- AEs
- Review of concomitant medications
- Physical examination
- ECOG performance status
- Vital signs - systolic / diastolic BP, pulse rate, respiratory rate, oxygen saturations and temperature
- Haematology – Full blood count (FBC), clotting screen
- Biochemistry – sodium, potassium, urea, creatinine, calcium, phosphate, total protein, albumin, bilirubin, alkaline phosphatase (ALP), AST and/or ALT
- Urinalysis (Blood, White Cells, Protein, Nitrites, pH), culture and sensitivity
- Blood samples for immunoprofiling
- Blood samples for research assessment - (d36 only) pre dose only
- Urine sample for cytokines - post dose only
- Pregnancy test (for female subjects of childbearing potential only): serum or urine Human Chorionic Gonadotropin (HCG) test. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required (Day 29 only)
- Administration of study agent and monitoring for any immediate adverse events

Evaluations on day 43 (Patients receiving intravenous treatment only)

- AEs
- Review of concomitant medications
- Physical examination
- ECOG performance status
- Vital signs - systolic / diastolic BP, pulse rate, respiratory rate, oxygen saturations and temperature
- Haematology – Full blood count (FBC), clotting screen
- Biochemistry – sodium, potassium, urea, creatinine, calcium, phosphate, total protein, albumin, bilirubin, alkaline phosphatase (ALP), AST and/or ALT, thyroid function tests (TSH, T3 and T4)
- Urinalysis (Blood, White Cells, Protein, Nitrites, pH), culture and sensitivity
- Blood samples for immunoprofiling
- Urine sample for cytokines – pre-dose only
- Pregnancy test (for female subjects of childbearing potential only): serum or urine Human Chorionic Gonadotropin (HCG) test. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required
- Administration of study agent and monitoring for any immediate adverse events

Evaluations on day 50 (Patients who have consented to resection of second non-marker lesion only)

- Patients who consented to having a second non-marker lesion left at the time of the original TURBT will undergo a cystoscopy for resection of the non-marker lesion which will be sent for research
- Bladder barbotage and a biopsy of normal bladder epithelium will be performed and sent for research
- Blood samples for immunoprofiling
- Urine sample for cytokines – to be taken pre-operatively

Evaluations on day 64 (Main study only – all patients)

- AEs
- Review of concomitant medications
- Physical examination
- ECOG performance status
- Vital signs - systolic / diastolic BP, pulse rate, respiratory rate, oxygen saturations and temperature
- Haematology – Full blood count (FBC), clotting screen
- Biochemistry – sodium, potassium, urea, creatinine, calcium, phosphate, total protein, albumin, bilirubin, alkaline phosphatase (ALP), AST and/or ALT, thyroid function tests (TSH, T3 and T4)
- Urinalysis (Blood, White Cells, Protein, Nitrites, pH), culture and sensitivity
- Blood samples for immunoprofiling
- Urine sample for cytokines - Intravenous patients pre-dose only, Intravesical post-dose only.
- Pregnancy test (for female subjects of childbearing potential only): serum or urine Human Chorionic Gonadotropin (HCG) test. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required
- Administration of study agent and monitoring for any immediate adverse events

Evaluations on day 85 (Main study only – all patients)

On day 85 of the main study a cystoscopy and TURBT will be performed in accordance with local hospital protocols. In addition:

- The size and morphology of the marker lesion should be documented in a bladder diagram with photographs where possible. Any new lesions should also be documented.
- Bladder barbotage should be performed prior to resection of any residual marker lesion
- If still visible, the marker lesion should be resected and sent to the central laboratory for immunoprofiling
- A biopsy will be performed of normal bladder epithelium and sent to the central laboratory for research purposes
- If the marker lesion is no longer visible, biopsies of the site of the previous tumour bed should also be performed and sent to the local laboratory for histological assessment and the central laboratory for research purposes
- Blood samples should be taken for immunoprofiling
- Urine sample for cytokines – taken pre-operatively

5.4 End of study/treatment evaluations

5.4.1 Safety run-in

The end of study visit for patients on the safety run-in part of the study will be performed on day 64 (or within 7 days thereafter). Evaluations will include:

- AEs
- Review of concomitant medications
- Physical examination
- ECOG performance status
- Vital signs - systolic / diastolic BP, pulse rate, respiratory rate, oxygen saturations and temperature
- Haematology – Full blood count (FBC), clotting screen
- Biochemistry – sodium, potassium, urea, creatinine, calcium, phosphate, total protein, albumin, bilirubin, alkaline phosphatase (ALP), AST and/or ALT, thyroid function tests (TSH, T3 and T4)
- Urinalysis (Blood, White Cells, Protein, Nitrites, pH), culture and sensitivity
- Blood samples for immunoprofiling
- Urine sample for cytokines
- Pregnancy test (for female subjects of childbearing potential only): serum or urine Human Chorionic Gonadotropin (HCG) test. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required

5.4.2 Main study (Intravenous and Intravesical treatment)

The end of treatment visit for the main study will be performed on day 92 (or within 7 days thereafter) for all patients. Evaluations will include:

- AEs
- Review of concomitant medications
- Physical examination
- ECOG performance status
- Vital signs - systolic / diastolic BP, pulse rate, respiratory rate, oxygen saturations and temperature
- Haematology – Full blood count (FBC), clotting screen
- Biochemistry – sodium, potassium, urea, creatinine, calcium, phosphate, total protein, albumin, bilirubin, alkaline phosphatase (ALP), AST and/or ALT, thyroid function tests (TSH, T3 and T4)
- Urinalysis (Blood, White Cells, Protein, Nitrites, pH), culture and sensitivity
- Pregnancy test (for female subjects of childbearing potential only): serum or urine Human Chorionic Gonadotropin (HCG) test. If the urine test is positive or cannot be confirmed as negative, a serum pregnancy test will be required

Follow-up evaluations

After the end of the treatment visit, patients will be followed up as per local protocols for standard of care at the discretion of the Investigator. This would ordinarily consist of cystoscopy every three months. Safety reporting will continue for 90 days post end of treatment. Data on the recurrence-free interval and progression-free interval will be collected for 2 years or until disease recurrence, progression, death or closure of the trial, whichever is sooner.

Collection of an optional tumour sample at the time of recurrence or progression for research purposes will be performed if the patient provides consent for this. The optional tumour sample would be collected at the time of a standard care surveillance cystoscopy or resection if the patient had a repeat TURBT in standard care. It will be made clear at the time of consent that the patient can change their mind at any point.

5.5 Evaluations on early withdrawal

If a patient is withdrawn from the treatment or the study early, the end of treatment evaluations in section 5.4 should be performed as soon as possible and within 28 days. Any/all remaining marker lesions should be resected as soon as possible as per standard care i.e. final TURBT should be brought forward.

6 EARLY PATIENT WITHDRAWAL

Treatment Withdrawal

During the course of the trial, a patient may withdraw early from treatment. This may happen for a number of reasons, including:

- Unacceptable toxicity
- AE/SAEs requiring discontinuation
- Loss to follow-up
- Significant protocol deviation or inability to comply with trial procedures
- Clinical decision
- Patient decision

Patients may choose to stop treatment and/or study assessments but may remain on study follow-up. When the patient stops treatment early, the 'End of Treatment' Form needs to be completed, and any other relevant CRFs (for example SAE Form). The reason for withdrawing from treatment early should be clearly documented in the medical records. Excepting withdrawal of consent, patients will be followed-up for 2 years or until disease recurrence, progression or death, whichever is sooner.

Consent Withdrawal

Consent withdrawal means that a patient has expressed a wish to withdraw from the study completely. Under these circumstances, the site needs to document all relevant discussions in the patient notes and notify the Trial Office, which will allow the office to mark all future CRFs as not applicable.

Under these conditions, Investigators are still required to follow up any SAEs until resolution.

6.1 Patient evaluability and replacement

During the safety run in phase patients who do not receive at least 5 out of the 6 scheduled treatments, except for reasons of drug-related toxicity, will be replaced such that there are 6 patients evaluable for tolerability. During the main study all participants who receive one dose of either intravenous or intravesical pembrolizumab will be evaluable. Patients who are not evaluable for the primary endpoint, i.e. any patient that withdraws from the main study before receiving treatment with the study drug, will be replaced.

7 SAMPLES FOR LABORATORY ANALYSIS

7.1 Samples to be analysed in local Trust's laboratories

Diagnostic Laboratories

Bloods samples for haematology and biochemistry analysis and urine samples for culture and sensitivity will be labelled with standard patient identifiers and sent to the hospital diagnostic laboratory. Results will be processed in the standard way and entered into the routine hospital reporting system. Samples will be stored, held, reported and subsequently destroyed in accordance with standard local laboratory practice.

Pathology

A sample of one of the resected tumours from the baseline TURBT should be sent to the local laboratory for tumour grading and fixation for subsequent PD-1/PD-L1 testing. In addition, in the case of complete clinical response at the day 85 TURBT, a biopsy of the tumour bed will be performed and sent to the local laboratory for histological analysis. Samples should be labelled, processed and reported according to local hospital protocols.

7.2 Samples to be sent to and analysed in Central Analytical laboratories

Detailed instructions for sample handling can be found in the Sample Handling Manuals for the trial.

PD-L1 expression analysis

The following samples will be sent to Qualtek for analysis

- Formalin fixed paraffin embedded (FFPE) tumour sample for PD-L1 expression analysis (5 unstained slides) Tumour samples will be fixed in formalin and paraffin embedded at site

Research Samples for translational research at Oxford

All other samples gifted for research will be delivered into the custodianship of Prof Vincenzo Cerundolo and managed by his team at the MRC Human Immunology Unit laboratory at the Weatherall Institute of Molecular Medicine. These will include:

- Blood samples for germline DNA analysis
- Blood samples for isolation of peripheral blood mononuclear cells (PBMC) and cytokine measurement
- Fresh tumour samples for DNA sequencing, gene expression profiling, T cell receptor (TCR) analysis and functional studies
- Fresh biopsies of normal bladder epithelium for immunoprofiling
- Fresh biopsies of tumour bed in event of CR for immunoprofiling
- Urine for cytokine measurement
- Recovered bladder barbotage instillations for isolation and characterisation of lymphocytes and/or tumour cells
- Blood samples for research taken from patients receiving intravesical treatment pre-dose and 2 hours post-dose for the first treatment, and pre-dose at cycle 6 (d36). Research use to be determined as directed by the custodian, expected analysis to be pharmacokinetic assay of the systemic absorption of pembrolizumab or other before/ after time critical analyses.

7.3 Pharmacodynamic and translational assays

Pharmacodynamic and translational assays may include but are not limited to the following:

- Immunoprofiling
  To assess the extent of T cell activation mediated by pembrolizumab and changes in the immunological

profile in response to treatment, PBMC will be isolated from blood before, during and after treatment. Tumour infiltrating lymphocytes will also be extracted from tumour samples using established protocols. Fluorescence-activated cell sorting (FACS) and mass cytometry (CyTOF) will be used to identify T cell subsets in peripheral blood and tumour, T cells will be isolated and the TCR repertoire will be analysed. Cytokines will also be measured from blood and urine samples.

- DNA sequencing
  Whole genome sequencing will be performed on blood and tumour samples. Gene expression profiling will also be performed.

7.4 Summary of samples/assays to be taken during the study

Assay/sample handling and storage will be managed according to separate written instructions.

7.5 Labelling and confidentiality of samples sent

All samples sent to analytical laboratories will be labelled with the trial code, trial patient number, Year of birth, and date/time taken. Should a laboratory receive any samples carrying patient personal identifying data the recipient must immediately obliterate this information and re-label with a trial label.

7.6 Clinical reporting of exploratory research assay results

The results of the research assays are exploratory and are not intended to influence the individual patient's medical care. Findings will not be reported routinely to the responsible clinician except in the unlikely event that the result might be beneficial to the patient's clinical management.

7.7 Samples for Biobanking

Samples are not being collected specifically for biobanking, however patients will be asked to consent for the transfer of any surplus tumour biopsy and blood samples from the trial central laboratories to a licensed biobank for future use.

7.8 Trial sample custodianship and retention at end of study

All research samples collected during this trial are considered a gift to the University of Oxford. The trial Academic Lead Professor Enzo Cerundolo is the responsible trial sample custodian. He is authorised to oversee trial sample retention, release for exploratory assays within the terms of the trial ethics approval. He will comply with the sponsor and applicable HTA guidelines and regulatory requirements. Any decision to vary from the planned exploratory analyses should be guided by the Chief Investigator, TMG and other trial stakeholders and documented by the Custodian.

Laboratories are instructed to retain any surplus samples pending instruction from the sample custodian on use, storage or destruction. It is possible that new or alternative assays may be of future scientific interest. At the end of the research study any surplus samples may be retained for use in other projects that have received ethical approval. Hence, any surplus study samples may be transferred to a licensed tissue bank where they will be managed in accordance with applicable host institution policies and the Human Tissue Act (HTA) requirements.

7.9 Withdrawal of consent for sample collection and/or retention

A patient may withdraw consent to provide samples for research at any time without giving a reason. The Investigator must ensure that their wishes are recorded in the medical record and will inform the Trial Office accordingly. The Investigator should discuss with patients the valuable use of samples that have already been provided and under circumstances where these samples have already been processed, it would not be possible to destroy such samples.

8 INVESTIGATIONAL MEDICINAL PRODUCTS (IMPS)

8.1 Name of IMPs

The investigational medicinal product being used in this trial is pembrolizumab. It will be supplied as a solution for Infusion

8.2 Treatment dose and duration

Study Drug	Dose	Dose frequency and schedule	Route of administration	Administer over	Use
Pembrolizumab	200mg	Q3W x 4 doses	IV infusion	30 minutes	Experimental
Pembrolizumab	50-200mg*	Q1W* x 6 doses Further dose D64	Intravesical	5 minutes	Experimental

* Subject to recommendation by the TMG following analysis of safety and tolerability data from the safety run-in cohort.

Dose modifications for drug related toxicity are permitted as detailed in section 8.5.

8.3 Management of drug administration

Trial treatment should begin 14 days after TURBT, and within 7 days of registration/randomisation or as close as possible to this date. Subsequent treatments should be administered according to the schedule shown in Figure 1 for the safety run-in cohort, and Figure 2 for the main study, in Section 2. For patients receiving intravenous treatment, pembrolizumab may be administered up to 3 days before or after the scheduled date for administrative reasons. For patients receiving intravesical treatment, pembrolizumab may be administered up to 1 day before or after the scheduled date if required for administrative reasons.

8.3.1 Intravesical treatment

The Pembrolizumab suspension should be made up in a syringe with a luer lock on the end. A urinary catheter will be inserted into the urethra under aseptic conditions according to local hospital protocol and the bladder drained completely. Using a catheter adapter with the syringe, the pembrolizumab suspension will then be instilled into the bladder via the catheter over a period of no more than 5 minutes. After instillation the catheter will be removed and the patient instructed to retain the instilled suspension in the bladder for a period of 2 hours. During this period care should be taken to ensure that the instilled suspension has sufficient contact with the whole mucosal surface of the bladder. The patient should be encouraged to mobilise or if lying down to rotate between prone, supine, left lateral and right lateral positions every 15 minutes. After 2 hours the patient should void the instilled suspension directly into a toilet.

The patient should be advised to limit their fluid intake for 4 hours prior to instillation and until bladder evacuation is permitted (i.e. 2 hours after instillation).

For the patient's first instillation of intravesical treatment in all parts of the study, and at any increased dose level for that patient during the safety run-in, the patient should be observed for 2 hours after administration of pembrolizumab with blood pressure, pulse, respiratory rate, oxygen saturations and temperature to be monitored every 15 minutes for first 30 minutes and then every 30 minutes until the bladder is voided. For subsequent instillations vital signs should be performed before treatment and thereafter as clinically indicated.

After bladder evacuation, the next first voided urine sample should be collected for immunoprofiling.

Intravenous treatment

Pembrolizumab will be administered as an intravenous infusion using an infusion pump over 30 minutes with a window of -5 and + 10 minutes through a peripheral line or in-dwelling catheter containing a sterile, non-pyrogenic, low protein binding 0.2 to 5 micron in-line or add on filter. No other drugs should be administered through the same infusion line. Whenever possible, the lowest infusion rate should be used that will allow completion of the infusion within the 30 minutes. Use 30ml normal saline to flush the infusion line at the end of the infusion.

Management of infusion reactions

Across clinical studies with pembrolizumab in approximately 5000 patients, severe infusion-related reactions have been reported in less than 0.1% of patients. For severe infusion reactions, the infusion should be stopped and pembrolizumab permanently discontinued. Patients with mild or moderate infusion reactions may continue to receive pembrolizumab with close monitoring; premedication with an antipyretic and/or antihistamine may be considered according to local protocols. See section 9.1 for further guidance.

8.4 Special precautions

Not applicable

8.5 Dose modification

Adverse events (both non-serious and serious) associated with pembrolizumab exposure may represent an immunologic aetiology. These adverse events may occur shortly after the first dose or several months after the last dose of treatment. For all patients in the study, whether receiving intravenous or intravesical treatment, pembrolizumab must be withheld for drug-related toxicities and severe or life-threatening AEs as per the table below. See section 9.1 for supportive care guidelines, including use of corticosteroids.

General instructions:

1. Corticosteroid taper should be initiated upon AE improving to Grade 1 or less and continue to taper over at least 4 weeks.
2. For situations where pembrolizumab has been withheld, pembrolizumab can be resumed after AE has been reduced to Grade 1 or 0 and corticosteroid has been tapered. Pembrolizumab should be permanently discontinued if AE does not resolve within 12 weeks of last dose or corticosteroids cannot be reduced to ≤10 mg prednisone or equivalent per day within 12 weeks.
3. For severe and life-threatening irAEs, IV corticosteroid should be initiated first followed by oral steroid. Other immunosuppressive treatment should be initiated if irAEs cannot be controlled by corticosteroids.

Immune-related AEs	Toxicity grade or conditions (CTCAE)	Action taken to pembrolizumab	Management with corticosteroid and/or other therapies	Monitor and follow-up
Pneumonitis	Grade 2	Withhold	• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper	• Monitor participants for signs and symptoms of pneumonitis • Evaluate participants with suspected pneumonitis with radiographic imaging and initiate corticosteroid treatment • Add prophylactic antibiotics for opportunistic infections
	Grade 3 or 4, or recurrent Grade 2	Permanently discontinue		
Diarrhea / Colitis	Grade 2 or 3	Withhold	• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper	• Monitor participants for signs and symptoms of enterocolitis (ie, diarrhea, abdominal pain, blood or mucus in stool with or without fever) and of bowel perforation (ie, peritoneal signs and ileus). • Participants with ≥ Grade 2 diarrhea

	Grade 4	Permanently discontinue		suspecting colitis should consider GI consultation and performing endoscopy to rule out colitis. • Participants with diarrhea/colitis should be advised to drink liberal quantities of clear fluids. If sufficient oral fluid intake is not feasible, fluid and electrolytes should be substituted via IV infusion.
AST / ALT elevation or Increased bilirubin	Grade 2	Withhold	• Administer corticosteroids (initial dose of 0.5- 1 mg/kg prednisone or equivalent) followed by taper	• Monitor with liver function tests (consider weekly or more frequently until liver enzyme value returned to baseline or is stable
	Grade 3 or 4	Permanently discontinue	• Administer corticosteroids (initial dose of 1-2 mg/kg prednisone or equivalent) followed by taper	
Type 1 diabetes mellitus (T1DM) or Hyperglycemia	Newly onset T1DM or Grade 3 or 4 hyperglycemia associated with evidence of β-cell failure	Withhold	• Initiate insulin replacement therapy for participants with T1DM • Administer anti-hyperglycemic in participants with hyperglycemia	• Monitor participants for hyperglycemia or other signs and symptoms of diabetes.
Hypophysitis	Grade 2	Withhold	• Administer corticosteroids and initiate hormonal replacements as clinically indicated.	• Monitor for signs and symptoms of hypophysitis (including hypopituitarism and adrenal insufficiency)
	Grade 3 or 4	Withhold or permanently discontinue[1]		
Hyperthyroidism	Grade 2	Continue	• Treat with non-selective beta-blockers (eg, propranolol) or thionamides as appropriate	• Monitor for signs and symptoms of thyroid disorders.
	Grade 3 or 4	Withhold or permanently discontinue[1]		
Hypothyroidism	Grade 2-4	Continue	• Initiate thyroid replacement hormones (eg, levothyroxine or liothyroinine) per standard of care	• Monitor for signs and symptoms of thyroid disorders.

Nephritis and Renal dysfunction	Grade 2	Withhold	• Administer corticosteroids (prednisone 1-2 mg/kg or equivalent) followed by taper.	• Monitor changes of renal function
	Grade 3 or 4	Permanently discontinue		
Myocarditis	Grade 1 or 2	Withhold	• Based on severity of AE administer corticosteroids	• Ensure adequate evaluation to confirm etiology and/or exclude other causes
	Grade 3 or 4	Permanently discontinue		
All other immune-related AEs	Intolerable/ persistent Grade 2	Withhold	• Based on type and severity of AE administer corticosteroids	• Ensure adequate evaluation to confirm etiology and/or exclude other causes
	Grade 3	Withhold or discontinue based on the type of event. Events that require discontinuation include and not limited to: Gullain-Barre Syndrome, encephalitis		
	Grade 4 or recurrent Grade 3	Permanently discontinue		

1. Withhold or permanently discontinue pembrolizumab is at the discretion of the investigator or treating physician.

NOTE:

For participants with Grade 3 or 4 immune-related endocrinopathy where withhold of pembrolizumab is required, pembrolizumab may be resumed when AE resolves to ≤ Grade 2 and is controlled with hormonal replacement therapy or achieved metabolic control (in case of T1DM).

Intravesical pembrolizumab

The following additional specific guidance applies to patients receiving intravesical pembrolizumab.

In the event of grade 2 haematuria, dysuria, urinary retention, urinary frequency/urgency, or bladder spasm that persists beyond 72 hours following intravesical treatment, the next scheduled instillation should be omitted. Symptoms must have resolved to a maximum of grade 1 before treatment resumes at the next scheduled timepoint. Irritative bladder symptoms can be managed by anticholinergic or antispasmodic therapy. In patients experiencing haematuria, it is important to perform a urine culture to exclude bacterial cystitis.

If the patient has a confirmed urinary tract infection treatment should be omitted to allow for resolution of the UTI with antibiotics. If catheterisation is traumatic, treatment should be omitted and subsequent cycles given as scheduled. Any patient who misses more than one dose of intravesical pembrolizumab should be discontinued from treatment.

8.6 Management of overdose

For the purposes of this trial, an overdose will be defined as any dose exceeding the prescribed dose for pembrolizumab. Whilst no specific information is available on the treatment of overdose of pembrolizumab, doses of up to 10mg/kg have been safely administered intravenously in other studies. In the event of overdose, the subject should be observed closely for signs of toxicity and appropriate supportive treatment should be provided if clinically indicated.

All reports of overdose with or without an adverse event must be reported within 24 hours to the Trial Office as an Event of Clinical Interest on an SAE Form (see section 12.6.1). The Trial Office will report the event to MSD within the agreed timescales.

9 OTHER TREATMENTS (NON-IMPS)

9.1 Support medication

Subjects should receive appropriate supportive care measures as deemed necessary by the treating Investigator. Suggested supportive care measures for the management of adverse events with potential immunologic aetiology are outlined below. Where appropriate, these guidelines include the use of oral or intravenous treatment with corticosteroids as well as additional anti-inflammatory agents if symptoms do not improve with administration of corticosteroids. 1mg/kg per day of methylprednisolone (or oral equivalent) is suggested as an initial treatment dose for most adverse events of potential immune aetiology. Note that several courses of steroid tapering may be necessary as symptoms may worsen when the steroid dose is decreased. For each disorder, attempts should be made to rule out other causes such bacterial or viral infections, which might require additional supportive treatment. The treatment guidelines are intended to be applied when the Investigator determines the events to be related to pembrolizumab.

Note: if after the evaluation the event is determined not to be related, the Investigator does not need to follow the treatment guidance (as outlined below). Refer to section 8.5 for dose modification.

It may be necessary to perform additional procedures such as bronchoscopy, endoscopy, or skin biopsy as part of evaluation of the event.

- **Pneumonitis**:
    - For **Grade 2 events**, treat with systemic corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks.
    - For **Grade 3-4 events**, immediately treat with intravenous steroids. Administer additional anti-inflammatory measures, as needed.
    - Add prophylactic antibiotics for opportunistic infections in the case of prolonged steroid administration.
- **Diarrhoea/Colitis**:

Subjects should be carefully monitored for signs and symptoms of enterocolitis (such as diarrhoea, abdominal pain, blood or mucus in stool, with or without fever) and of bowel perforation (such as peritoneal signs and ileus).

- All subjects who experience diarrhoea/colitis should be advised to drink liberal quantities of clear fluids. If sufficient oral fluid intake is not feasible, fluid and electrolytes should be substituted via IV infusion. For Grade 2 or higher diarrhoea, consider GI consultation and endoscopy to confirm or rule out colitis.
- For **Grade 2 diarrhoea/colitis,** administer oral corticosteroids.
- For **Grade 3 or 4 diarrhoea/colitis,** treat with intravenous steroids followed by high dose oral steroids.
- When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks.

- **Type 1 diabetes mellitus (if new onset, including diabetic ketoacidosis [DKA]) or ≥ Grade 3 Hyperglycaemia, if associated with ketosis (ketonuria) or metabolic acidosis (DKA)**
  - For **T1DM** or **Grade 3-4** Hyperglycaemia
    - Insulin replacement therapy is recommended for Type I diabetes mellitus and for Grade 3-4 hyperglycaemia associated with metabolic acidosis or ketonuria.
    - Evaluate patients with serum glucose and a metabolic panel, urine ketones, glycosylated haemoglobin, and C-peptide.
- **Hypophysitis**:
  - For **Grade 2** events, treat with corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks. Replacement of appropriate hormones may be required as the steroid dose is tapered.
  - For **Grade 3-4** events, treat with an initial dose of IV corticosteroids followed by oral corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks. Replacement of appropriate hormones may be required as the steroid dose is tapered.
- **Hyperthyroidism or Hypothyroidism**:

  Thyroid disorders can occur at any time during treatment. Monitor patients for changes in thyroid function (at the start of treatment, periodically during treatment, and as indicated based on clinical evaluation) and for clinical signs and symptoms of thyroid disorders.
  - **Grade 2** hyperthyroidism events (and **Grade 2-4** hypothyroidism):
    - In hyperthyroidism, non-selective beta-blockers (e.g. propranolol) are suggested as initial therapy.
    - In hypothyroidism, thyroid hormone replacement therapy, with levothyroxine or liothyroinine, is indicated per standard of care.
  - **Grade 3-4** hyperthyroidism
    - Treat with an initial dose of IV corticosteroid followed by oral corticosteroids. When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks. Replacement of appropriate hormones may be required as the steroid dose is tapered.
- **Hepatic**:
  - For **Grade 2** events, monitor liver function tests more frequently until returned to baseline values (consider weekly).
    - Treat with IV or oral corticosteroids
  - For **Grade 3-4** events, treat with intravenous corticosteroids for 24 to 48 hours.

- When symptoms improve to Grade 1 or less, a steroid taper should be started and continued over no less than 4 weeks.

- **Renal Failure or Nephritis**:
  - For **Grade 2** events, treat with oral corticosteroids.
  - For **Grade 3-4** events, treat with intravenous corticosteroids.
  - When symptoms improve to Grade 1 or less, steroid taper should be started and continued over no less than 4 weeks.

- **Stevens-Johnson syndrome (SJS) and Toxic Epidermal Necrolysis (TEN)**
  - If patient has signs or symptoms of SJS or TEN withhold pembrolizumab and refer the patient for specialist care for assessment and treatment
  - If SJS or TEN is confirmed permanently discontinue treatment with pembrolizumab

- **Immune-mediated myocarditis management**
  - For suspected immune-mediated myocarditis, ensure adequate evaluation to exclude other aetiologies, and administer corticosteroids as appropriate.

- **Management of Infusion Reactions**: Signs and symptoms usually develop during or shortly after drug infusion and generally resolve completely within 24 hours of completion of infusion.

  The table below shows treatment guidelines for subjects who experience an infusion reaction associated with administration of pembrolizumab.

NCI CTCAE Grade	Treatment	Premedication at subsequent dosing
Grade 1 Mild reaction; infusion interruption not indicated; intervention not indicated	Increase monitoring of vital signs as medically indicated until the subject is deemed medically stable in the opinion of the Investigator.	None
Grade 2 Requires infusion interruption but responds promptly to symptomatic treatment (e.g., antihistamines, NSAIDS, opiates, IV fluids); prophylactic medications indicated for < =24 hrs	**Stop Infusion and monitor symptoms.** Additional appropriate medical therapy may include but is not limited to: • IV fluids • Antihistamines • NSAIDS • Paracetamol • Opiates Increase monitoring of vital signs as medically indicated until the subject is deemed medically stable in the opinion of the Investigator. If symptoms resolve within one hour of stopping drug infusion, the infusion may be restarted at 50% of the original infusion rate (e.g., from 100 mL/hr to 50 mL/hr). Otherwise dosing will be held until symptoms resolve and the subject should be premedicated for the next scheduled dose. **Subjects who develop Grade 2 toxicity despite adequate premedication should be permanently discontinued from further trial treatment administration.**	Subject may be premedicated 1.5h (± 30 minutes) prior to infusion of pembrolizumab with: Chlorphenamine 4 mg PO or 10mg IV Paracetamol 1000g PO
Grades 3 or 4 Grade 3: Prolonged (i.e., not rapidly responsive to symptomatic medication and/or brief interruption of infusion); recurrence of	**Stop Infusion.** Additional appropriate medical therapy may include but is not limited to: • IV fluids • Antihistamines • NSAIDS • Paracetamol	No subsequent dosing

NCI CTCAE Grade	Treatment	Premedication at subsequent dosing
symptoms following initial improvement; hospitalization indicated for other clinical sequelae (e.g., renal impairment, pulmonary infiltrates) Grade 4: Life-threatening; inotrope or ventilatory support indicated	• Opiates • Oxygen • Inotropes • Corticosteroids • Adrenaline Increase monitoring of vital signs as medically indicated until the subject is deemed medically stable in the opinion of the Investigator. Hospitalization may be indicated. **Subject is permanently discontinued from further trial treatment administration.**	

9.2 Concomitant medication and non-drug therapies

Concomitant medication may be given as medically indicated taking account of prohibited therapies (section 9.3). All patients will be asked to provide a complete list of prescription and over-the-counter medications that have been taken within the previous 4 weeks prior to the first treatment visit. They must also inform the Investigator about any new medication started while in the trial.

Details (including indication, doses, frequency and start/stop dates) of concomitant medication taken during the trial until the completion of the end of study visit must be recorded in the medical record and the appropriate CRF.

9.3 Prohibited therapies

Patients should not be prescribed any other anti-cancer or investigational therapies while participating in this study. In addition, the following advice should be noted:

10. Live vaccines should not be given within 30 days prior to the first dose of the trial treatment, while participating in the trial and for 6 months after the end of treatment. Examples of live vaccines include, but are not limited to, the following: measles, mumps, rubella, chicken pox, yellow fever, rabies, BCG, and oral typhoid vaccine. Seasonal influenza vaccines are generally killed virus vaccines and are allowed; however intranasal influenza vaccines are live attenuated vaccines, and are not allowed.
11. Glucocorticoids for any purpose other than to modulate symptoms from an adverse event suspected to be of immunologic aetiology should be avoided (excepting inhaled or intranasal corticosteroids or local steroid injections). The use of physiological replacement doses of corticosteroids may be approved after consultation with the Trial Office.

Subjects who, in the assessment by the Investigator, require the use of any of the aforementioned treatments for clinical management should be removed from the trial.

The use of intravesical BCG should be avoided for 30 days following the last dose of study drug.

9.4 Drug Interactions

No formal pharmacokinetic drug interaction studies have been conducted with pembrolizumab. Subjects should maintain a normal diet unless modifications are required to manage an AE such as diarrhoea, nausea or vomiting.

10 DRUG MANAGEMENT

10.1 Drug supplies

Pembrolizumab will be supplied free of charge by Merck as summarised below

Product Name & Potency	Dosage Form
Pembrolizumab 100 mg/ 4mL	Solution for Infusion

All supportive medication is to be sourced and funded locally.

10.2 Drug ordering

Drug supply details will be provided by the Trial Office prior to shipping, and all subsequent queries should be directed to the PEMBLA trial team. If a vial of pembrolizumab is accidentally destroyed, i.e. by dropping the vial or through contamination, the pharmacist should contact the Trial Office on 01865 227190 or email octo-pembla@oncology.ox.ac.uk immediately so that replacement patient supplies may be arranged.

Initial supplies of pembrolizumab are sent out by Merck (MSD) after they have been informed by the Trial Office that all approvals are in place. Subsequent supplies will be provided in batches organised by the Trial Office. Pharmacies will be responsible for monitoring and requesting stock as required, following the re-ordering instructions in the Pharmacy Manual. Contact the Trial office on 01865227190 or by email on octo-pembla@oncology.ox.ac.uk to request an order to be placed. Standard delivery time is within 5 working days of the order being placed.

10.3 IMP Receipt

Receipt of trial medication must be recorded by an authorised person at the trial site. IMP shipments will arrive with an 'Acknowledgement of Receipt' form that should be emailed to octo-pembla@oncology.ox.ac.uk to confirm receipt.

If supplies are damaged on arrival, contact the PemBla Trial Office. Damaged supplies should be destroyed on site and a drug destruction form completed.

10.4 Handling and storage

Clinical trial supplies must be stored in a secure, limited-access location under the storage conditions specified on the label. Clinical trial supplies may not be used for any purpose other than that stated in the protocol. If a temperature excursion occurs, please quarantine the affected drug as per local procedures and contact the PemBla Trial Office on 01865 227190 or email octo-pembla@oncology.ox.ac.uk

10.5 Dosing dispensing

Pembrolizumab Solution for Infusion is a sterile, non-pyrogenic aqueous solution supplied in a single-use Type I glass vial containing 100mg/4ml of Pembrolizumab. The product is a preservative-free solution which is essentially free of extraneous particulates. Solution for Infusion vials should be stored at refrigerated conditions (2-8°C) and in the original box to ensure the drug product is protected from light.

Pembrolizumab infusion solutions should be prepared in 0.9% Sodium Chloride (normal saline) and the final concentration of pembrolizumab in the infusion solution should be between 1mg/ml and 10mg/ml. For intravesical infusions the final volume of infusion instilled will be 40ml.

Pembrolizumab solutions may be stored at room temperature for a cumulative time of up to 4 hours. This includes room temperature storage of reconstituted drug product solution in vials, room temperature storage of admixture solutions in the IV bags and the duration of infusion. In addition, IV bags may be stored under refrigeration at 2°C to 8°C for up to 20 hours. If refrigerated, allow the IV bags to come to room temperature prior to use.

For further information, including particulars for intravesical administration, please see the pharmacy manual.

10.6 Drug accountability

Full patient-specific drug accountability records must be maintained for pembrolizumab. Hospitals may amend the Drug Dispensing Logs provided, or use their own documentation if it captures all the information requested on the Drug Dispensing Logs and is given prior approval by the Trials Office. The drug dispensing and inventory logs should be kept up to date and must contain the following information; patient identifier, date and quantity received at site, date and quantity dispensed, date and quantity returned/destroyed at site. At the conclusion of the study the overall numbers of drug shipped to the centre, the number dispensed and the number destroyed will be provided by the pharmacy. An account must be given of any discrepancy.

10.7 Drug destruction

Any unused drug remaining in a used vial should not be used for another infusion of the same subject or different subject and should be disposed of at site according to local hospital policy. During the trial or upon completion or termination of the study, written permission must be gained from the Trial Office before any unused and/or partially

used and any expired investigational product is destroyed. Once permission is gained, local hospital destruction policy can be followed. A dated certificate of disposal should be completed and retained in the Pharmacy File. As per Section 10.3, if supplies are damaged on arrival, contact the PemBla Trial Office. Upon permission from the trial office, damaged supplies should be destroyed on site and a drug destruction form completed.

10.8 Occupational safety

Vein extravasation and accidental spillages of IMP should be dealt with according to hospital policy.

The product is not expected to pose an occupational safety risk to site staff under normal conditions of use and administration.

10.9 Product quality complaints:

Any quality complaints or comments concerning the pembrolizumab clinical drug supplies for the study should be sent to the Trial Office in the first instance: octo-pembla@oncology.ox.ac.uk

11 EVALUATION OF RESPONSE`

11.1 Tumour assessment

A clinical assessment of the extent of disease will be performed at the time of cystoscopy on day -14 and day 85 for patients within the main study. Disease response is not assessed at day 50 for applicable participants having resection of a second lesion. Data will also be collected during the follow-up from routine cystoscopy assessments.

Baseline evaluations
The location, size and morphology of all lesions prior to resection including identification of the marker lesion should be documented in a bladder diagram with photographs where possible at the time of the initial TURBT (day -14). For patients who have consented to undergo a second TURBT the second non-marker lesion should be identified on the bladder diagram.

Evaluations at end of treatment
Repeat cystoscopy will be performed at the end of treatment on day 85. The location, size and morphology of the marker lesion and any new or recurrent lesions should be documented in a bladder diagram with photographs where possible. Transurethral resection will be performed for the marker lesion if persistent and any new or recurrent lesions at this time. If the marker lesion is not visible, biopsies of the tumour bed will be performed.

11.2 Tumour response

Tumour response will be classified as follows:

Pathological complete response: Clinical resolution of the marker lesion at day 85 with no pathological evidence of malignancy on biopsies of the tumour bed

Clinical complete response: Clinical resolution of the marker lesion at day 85 without negative biopsies of the tumour bed.

Non-response: Persistence of the marker lesion

Progression: Development of any of the following

- T1 disease (lamina propria invasion)
- $\geq$ T2 disease (muscle invasive)
- Lymph node(N1+) or distant metastases (M1)
- High grade (G3) disease or CIS

Recurrence: Appearance of one or more new lesions

Tumour response should be classified as 'non evaluable' (NE), only when it is not possible to classify it under another response category e.g. when baseline and/or follow-up assessment is either not performed or not performed appropriately.

11.3 Other definitions of outcome:

Toxic death: Any death to which drug toxicity is thought to have a major contribution.
Early death: Death during the first three weeks of treatment that is not a toxic death.

12 SAFETY REPORTING

The Investigator will monitor each patient for clinical and laboratory evidence of adverse events on a routine basis throughout the study. Adverse event monitoring starts on the day of the TURBT procedure (Day -14) until 90 days post treatment, or 30 days following administration of the last dose of study medication if the subject initiates a new anticancer therapy. Should an Investigator become aware of any study drug related SAEs following this period these must also be reported as stated below. All reportable AEs will be followed to a satisfactory conclusion during the treatment period. Any reportable drug-related AEs that are unresolved at the end of treatment visit are to be followed up by the Investigator until resolution or stabilisation. SAEs that are considered to be definitely unrelated to the trial intervention will not be followed up and monitored beyond the end of treatment.

All AEs reported to the Trial Office will be processed according to internal SOPs. The Trial Office may request additional information for any AE as judged necessary.

12.1 Adverse Event Definitions

An Adverse Event or Experience (AE) is any untoward medical occurrence in a study subject temporally associated with the administration of an investigational medicinal product (IMP) or a comparator product, whether or not considered related to the IMP or a comparator product. An AE can therefore be any unfavourable and unintended sign, symptom, disease (new or exacerbated) and/or significant abnormal laboratory or physiological observation temporally associated with the use of a medicinal product.

A Serious Adverse Event (SAE) is any AE, regardless of dose, causality or expectedness, that:

• **Results in death**	
• **Is life-threatening**	This refers to an event in which the subject was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.
• **Requires in-patient hospitalisation or prolongs existing inpatient hospitalisation**	In general, hospitalisation signifies that the subject has been admitted (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalisation are AEs. If a complication prolongs hospitalisation or fulfils any other serious criteria, the event is serious. When in doubt as to whether hospitalisation occurred or was necessary, the AE should be considered serious.
• **Results in persistent or significant incapacity or disability**	This means a substantial disruption of a person's ability to conduct normal life functions. It does not include experiences of relatively minor medical significance or accidental trauma (e.g. sprained ankle), which do not constitute a substantial disruption.
• **Is a congenital anomaly or birth defect**	
• **Is any other medically other**	Defined as an event that may jeopardise the patient or may require

medically important event	intervention to prevent one of the outcomes listed above. Any new primary cancer must be reported as an SAE.

An Adverse Drug Reaction (ADR) is an AE which is considered to be causally related to any dose of the IMP. This means that a causal relationship between the IMP and the AE is at least a reasonable possibility, i.e., the relationship cannot be ruled out.

An Unexpected Drug Reaction is an adverse drug reaction, the nature or severity of which, is not consistent with applicable product information (referring to information in SPC or IB).

A Suspected Unexpected Serious Adverse Drug Reaction (SUSAR) is a serious adverse drug reaction, the nature or severity of which is not consistent with the applicable product information (e.g. Investigator's Brochure for an unapproved investigational product or SPC for an approved product).

12.2 Clinical laboratory abnormalities and other abnormal assessments as AEs and SAEs

Abnormal laboratory findings (e.g., clinical chemistry, haematology, urinalysis) or other abnormal assessments (e.g., ECGs, X-rays and scans) that are judged by the Investigator as clinically significant will be recorded as AEs or SAEs if they meet the definitions given above. By definition, **all Grade 3 and or 4 laboratory abnormalities should be reported as SAEs.**

Clinically significant abnormal laboratory findings or other abnormal assessments that are detected during the study or are present at baseline and significantly worsen following the start of the study will be reported as AEs or SAEs. However, clinically significant abnormal laboratory findings or other abnormal assessments that are associated with the disease being studied, unless judged by the Investigator as more severe than expected for the patient's condition, or that are present or detected at the start of the study and do not worsen, will not be reported as AEs or SAEs.

The Investigator will exercise his or her medical and scientific judgment in deciding whether an abnormal laboratory finding or other abnormal assessment is clinically significant.

12.3 Determining adverse event causality

A Serious Adverse Reaction (SAR) is a SAE that may be related to trial treatment. The assessment of "relatedness" must be determined by a medically qualified individual and is primarily the responsibility of the PI at site or agreed designee. SAEs that will be considered related will include any SAE that is documented as possibly, probably or definitely related to protocol treatment. The assessment of relatedness is made using the following:

Classification	Relationship	Definition
Drug-related	**Definitely related**	• Starts within a time related to the study drug administration *and* • No obvious alternative medical explanation.
	Probably related	• Starts within a time related to the study drug administration *and* • Cannot be reasonably explained by known characteristics of the patient's clinical state.
	Possibly related	1. Starts within a time related to the study drug administration *and* 2. A causal relationship between the study drug and the adverse event is at least a reasonable possibility.
Not drug-related	**Probably not related**	3. The time association or the patient's clinical state is such that the study drug is not likely to have had an association with the observed effect.
	Definitely not related	• The AE is definitely not associated with the study drug administered.

The Investigator must endeavour to obtain sufficient information to confirm the causality of the adverse event (i.e. relation to surgery, study drug, background treatment, other illness, progressive malignancy etc.) and give their

opinion of the causal relationship between each AE and study drug. This may require instituting supplementary investigations of significant AEs based on their clinical judgement of the likely causative factors and/or include seeking a further specialist opinion.

12.4 Reference safety information (RSI) for assessment of expectedness

The reference safety information (RSI) for the trial is section 7.1 of the Investigator Brochure for pembrolizumab which lists the expected side effects associated with the use of intravenous pembrolizumab.

A copy of the current approved version of the IB must be held in the Site File for reference. Any change or update to the IB during the trial will be made via substantial amendment.

Local side effects of intravesical treatment

Cystitis and haematuria are the most common local side effects of current intravesical therapy and typically resolve within 48 hours after instillation. Bladder spasm, urinary frequency or urgency and urinary retention may also occur. Haematuria frequently occurs with cystitis following intravesical treatment and may be related to the extent of the previous TURBT. In rare instances, catheterisation and bladder irrigation of clots or cystoscopy may be required. Table 1 below lists the most common adverse reactions seen in the SWOG study 8795 using intravesical BCG or MMC.

Adverse Event	TICE BCG n = 222		MMC n = 220	
	All grades	Grade ≥ 3	All grades	Grade ≥ 3
Dysuria	115 (52%)	6 (3%)	77 (35%)	5 (2%)
Urgency/Frequency	112 (50%)	5 (2%)	63 (29%)	7 (3%)
Haematuria	85 (38%)	6 (3%)	56 (25%)	5 (2%)
Flu-like symptoms	54 (24%)	1 (<1%)	29 (13%)	0
Fever	37 (17%)	1 (<1%)	7 (3%)	0
Pain (not specified)	37 (17%)	4 (2%)	22 (10%)	1 (<1%)
Haemorrhagic cystitis	19 (9%)	3 (1%)	10 (5%)	0
Chills	19 (9%)	0	2 (1%)	0
Bladder cramps	18 (8%)	0	9 (4%)	0
Nausea	16 (7%)	0	12 (5%)	0
Incontinence	8 (4%)	0	3 (1%)	0
Myalgia/arthralgia	7 (3%)	0	0	0
Diaphoresis	7 (3%)	0	1 (<1%)	0
Rash	6 (3%)	1 (<1%)	16 (7%)	2 (1%)

Table 1. Adverse events seen in SWOG study 8795 associated with use of intravesical BCG or MMC

12.5 Suspected Unexpected Serious Adverse Drug Reactions (SUSARs)

All SUSARS must be reported to the responsible Authority and main REC by the Trial Office within the required timelines:

- Fatal or life threatening SUSARs will be reported within 7 days of the Trial Office receiving the initial report. Any additional information will be reported within eight days of sending the first report.
- All other SUSARs will be reported within 15 days of the Trial Office receiving the initial report

In addition, other safety issues qualify for expedited reporting where they might materially alter the current risk assessment of an IMP or be sufficient to change IMP administration or the overall conduct of the trial.

12.6 Expedited reporting of SAEs

The following SAE reporting requirements apply regardless of the Investigator's assessment of the causality or expectedness of the SAE. All SAEs should be reported on the trial SAE report form (see SAE report form and completion guidelines).

If a Serious Adverse Event occurs that requires reporting, a Serious Adverse Event reporting form should be completed and emailed within 24 hours of becoming aware of the event to:

Pharmacovigilance Office, OCTO
Email: octo-safety@oncology.ox.ac.uk
Tel no: +44 (0) 01865 227182

OCTO will ensure SAEs are forwarded to MSD Global Safety within 2 working days of learning of the information. If the SAE has not been reported to OCTO within the specified timeframe, a reason for lateness must be provided when sending the SAE Report Form.

Investigators should also adhere to their local Trust policy for incident and SAE reporting in research.

12.6.1 Events of Clinical Interest

Selected non-serious and serious adverse events are also known as Events of Clinical Interest (ECI) and must be reported within 24 hours to the Trial Office as an Event of Clinical Interest on an SAE Form. OCTO will ensure ECIs are forward to the MSD Global Safety within the correct timeframe. ECIs will be reported from the day of the TUBRT procedure (Day -14) through to 90 days following cessation of study treatment, or 30 days following cessation of treatment if the subject initiates a new anti-cancer therapy.

Events of clinical interest for this trial include:

1. An overdose of MSD product, as defined in Section 8.6
2. An elevated AST or ALT lab value that is greater than or equal to 3X the upper limit of normal and an elevated total bilirubin lab value that is greater than or equal to 2X the upper limit of normal and, at the same time, an alkaline phosphatase lab value that is less than 2X the upper limit of normal, as determined by way of protocol-specified laboratory testing or unscheduled laboratory testing.*
   *Note: These criteria are based upon available regulatory guidance documents. The purpose of the criteria is to specify a threshold of abnormal hepatic tests that may require an additional evaluation for an underlying aetiology.

12.7 Follow-up of Serious Adverse Events

A follow-up report must be completed when the SAE resolves, is unlikely to change, or when additional information becomes available. If the SAE is a suspected SUSAR then follow up information must be provided as requested by the Trial Office.

If new or amended information on a reported SAE becomes available, the Investigator should report this on a new SAE form using the completion guidelines. If using the original form to notify further information, you must initial and date all new or amended information so that all changes are clearly identified.

SAEs that are considered to be definitely unrelated to the trial intervention will not be followed up and monitored beyond the end of treatment.

12.8 Reporting Adverse Events on the CRF

All AEs, including Serious AEs must be recorded on the case report forms (CRF) for that patient (unless otherwise specified in section 12.9. The information provided will include date of onset, event diagnosis (if known) or sign/symptom, severity, time course, duration and outcome and relationship of the AE to study drug. Any concomitant medications or any other therapy used to treat the event must be listed. The Investigator will provide an "other" cause for serious AEs considered to be unrelated to the study drug. Sites should ensure data entered into the CRF is consistent with the SAE report information where applicable.

Each separate AE episode must be recorded. For example, if an AE resolves completely or resolves to baseline and then recurs or worsens again, this must be recorded as a separate AE. For AEs to be considered intermittent, the events must be of similar nature and severity.

AEs may be spontaneously reported by the patient and/or in response to an open question from study personnel or revealed by observation, physical examination, or other diagnostic procedures. Any clinically relevant deterioration in laboratory assessments or other clinical finding is considered an AE.

Terms and Grading of Events
All adverse events and toxicities must be graded according to the NCI Common Terminology Criteria for adverse events (NCI-CTCAE) Version 4.03.

12.9 Events exempt from being reported as AE/ SAEs

Progression of underlying disease
Disease recurrence and progression are efficacy endpoints for this study and will be captured on the CRF. Adverse events including hospitalisation that are clearly consistent with disease recurrence or progression will not be reported as individual AE/SAEs. Clinical symptoms of recurrence or progression will only be reported as adverse events if the symptom cannot be determined as exclusively due to the underlying malignancy, or does not fit the expected pattern of progression for the disease under study. Every effort should be made to document the objective recurrence or progression of underlying malignancy.

Death on study
A death due to the disease under study is to be recorded on the Death CRF form. All other deaths must be reported as SAEs.

Elective admissions and supportive care
Elective admissions to hospital for patient convenience or for planned procedures or investigations or treatment as specified in this protocol and standard supportive care are not SAEs, and do not require SAE reporting.

Expected toxicity of standard treatment
The following adverse events related to surgery are considered expected if grade $\leq$ 3 and are exempt from expedited reporting but should be reported using the appropriate CRF:

- Bladder discomfort or pain
- Urinary frequency
- Haematuria
- Urinary retention requiring catheterisation
- Infection of the bladder requiring antibiotics
- Delayed bleeding requiring removal of clots or further surgery
- Damage to the ureters requiring additional therapy
- Injury to the urethra causing delayed scar formation
- Perforation of the bladder requiring a temporary urinary catheter or open surgical repair

12.10 Informing Investigators of new safety information

The Trial Office or the Chief Investigator will ensure that all Investigators are kept informed in a timely manner, as new safety profile information becomes available. Investigators are responsible for briefing their study team and onward transmission to their R&D office as appropriate.

13 PREGNANCY

Pregnancies (in a participant or partner) occurring during treatment or within 120 days of last dose of pembrolizumab must be reported within 24 hours of becoming aware of it using the Pregnancy Notification Form. The Investigator must ensure that all patients are aware at the start of a clinical trial of the importance of reporting all pregnancies (in themselves and their partners) that occur during this period.

Women who become pregnant should be withdrawn from treatment immediately.

All reported pregnancies should be followed up through the participants General Practitioner or their Consultant Oncology until the outcome is known, additional consent for partners of trial patients will be sought to do this using the Pregnancy Information Sheet and Consent Form. Pregnancy outcome must be recorded in the medical record and in the follow-up section of the Pregnancy Notification Form. If the outcome of the pregnancy meets any of the criteria for seriousness, it must also be reported as an SAE. Examples of pregnancy outcomes that are SAEs include reports of:

- congenital anomalies or developmental delay, in the foetus or the child.

- foetal death and spontaneous abortion.
- suspected adverse reactions in the neonate that are classified as serious

14 DEFINING THE END OF TRIAL

For this study the end of the trial is defined as "The last visit of the last patient undergoing the trial (LPLV) and up to 24 months for follow-up".
The study will be stopped when:

- The stated number of patients to be recruited is reached.
- The stated primary and secondary objectives of the study are achieved.

The sponsor and the Chief Investigator reserve the right to terminate the study earlier at any time. In terminating the study, they must ensure that adequate consideration is given to the protection of the participants' best interests.

15 STATISTICAL CONSIDERATIONS

15.1 Sample size and power

The primary aim of this study is safety, tolerability and toxicity and therefore the sample size has been chosen to give reasonable information on these endpoints. In considering the secondary endpoint of response, using Ahern's single stage phase II design with significance level 0.05 (one sided), power 0.8, we have set 20% as the highest level of efficacy at which we would not continue to another trial. Based upon existing intravesical therapies a complete response rate of 55% may be achievable. The 20% threshold requires a minimum of 13 patients in each arm of the trial, and if there are 6 successes in an arm then a 55% complete response rate is plausible. Thirty patients will be randomised in total, to allow for drop out - No adjustment will be made for the multiple testing caused by having two arms.

16 STATISTICAL ANALYSIS PLAN

A detailed statistical analysis plan will be available from the time the first patient is recruited and will be finalised before any analysis is undertaken. The analysis plan will be written in accordance with current Standard Operating Procedures and will be finalised and agreed by the trial statistician, a second statistician and the CI.

The data from the safety run-in phase will be described including the variables that define the DLTs and the safety and toxicity variables.

All analyses in the main trial will be on a modified intention-to-treat basis. This means that patients will be analysed as they are randomised irrespective of the treatment actually received. The intention-to-treat population will include all patients who have given their informed consent and for whom there is confirmation of successful allocation of a randomisation number. This will be modified by omitting patients without the endpoint/outcome measure.

It is therefore important that every effort is made to encourage patients, including those patients who do not receive/complete their allocated treatment, to attend for follow-up clinic visits to avoid bias in the analysis of the results.

For the main trial, baseline characteristics will be summarised (number and frequency) for all patients by randomisation group. Serious adverse events and adverse events will be summarised. Complete response rate will be given for each treatment with a 90% confidence interval (CI). Kaplan-Meier curves for the recurrence-free interval (RFI) and for progression-free interval (PFI) will be presented for each treatment group, with RFI and PFI estimates given at 6 months, 12 months, 18 months, and 2 years along with 90% CI.

16.1 Inclusion in analysis

All patients will be included in the safety analysis. All randomized patients with outcome data will be included in all analysis (modified intention to treat). Full details of patients groups for analysis will be detailed in the separate Statistical Analysis Plan.

16.2 Subgroup analysis

No patient subgroup analysis is planned.

16.3 Interim Analyses

Interim analysis will be carried out for dose escalation as described in section 2.

16.4 Procedures for reporting any deviation(s) from the original statistical plan

Any deviations from the Statistical Analysis Plan will be noted in the final statistical report.

16.5 Final analysis

Based upon projected accrual rates, this trial is expected to complete recruitment within 24 months of opening to recruitment. Final analysis will be after all patients have been followed up for at least 24 months with a publication planned within a year.

17 TRIAL COMMITTEES

17.1 Trial Management Group (TMG)

The Chief Investigator will chair a TMG responsible for overseeing the successful conduct and publication of the trial. The TMG will review safety and dose escalations. It will provide regular progress reports as required by the applicable steering committees and governance bodies.

MSD will be informed of the outcome of any TMG meetings and the MSD Medical Advisor will be involved in end-of-study discussions.

17.2 Data and Safety Monitoring

There is no Data and Safety Monitoring Committee (DSMC). SAEs upon receipt are reviewed by an independent nominated clinician as part of the trial office SAE Standard Operating Procedure. Cumulative reports of all AEs & SAEs are reviewed by the TMG and an Independent Trial Steering Committee (IEPTOC) will be in place to monitor the safety of the trial on an ongoing basis (IEPTOC meets, on average, 6-monthly).

17.3 Trial Steering Committee

As an early phase study in the OCTO portfolio, the Independent Early Phase Trials Oversight Committee (IEPTOC) will provide overall supervision of the safe and effective conduct of the trial according to its terms of reference. At least annually it will review trial progress against agreed milestones, adherence to protocol, patient safety and consider new information. The IEPTOC has the authority to recommend study closure where appropriate.

18 DATA MANAGEMENT

18.1 Database considerations

Data management will be performed via a web-based, bespoke trial database (OpenClinica). OpenClinica is a dedicated and validated clinical trials database designed for electronic data capture. See: http://www.openclinica.org. The Trial Office will provide sites with instructions and a video link for training purposes.

The participants will be identified by a unique trial specific number and/or code in any database. The name and any other identifying detail will NOT be included in any trial data electronic file.

18.2 Case reports forms (CRFs)

The Investigator and study site staff will ensure that data collected on each subject is recorded in the CRF as

accurately and completely as possible. All appropriate laboratory data, summary reports and Investigator observations will be transcribed into the CRFs from the relevant source data held in the site medical record(s). CRFs entries will not contain any source data (unless otherwise specified in the completion instructions provided by the Trial Office). It is important to ensure that:

- the relevant CRFs are completed.
- all CRF data are verifiable in the source documentation or the discrepancies must be explained.
- CRF sections are completed in a timely fashion, as close to the visit or event being recorded as possible.
- data queries are resolved and documented by authorised study staff in a timely fashion. The reason for the change or correction should be given where appropriate.
- as much data as possible is entered and cleaned in preparation for each study database lock point.

Note: 'in a timely fashion' means within no more than 14 working days of the initial event and within 28 days of receipt of a data query unless otherwise specified.

The above considerations also apply to patients who are withdrawn early. If a patient withdraws from the study, the reason must be noted on the appropriate form and the patient must be followed-up as per protocol.

18.3 Accounting for missing, unused, or spurious data.

Attempts will be made to find any missing data and supplement this where possible after consultation with the Investigator. The completeness and correctness of the data will be monitored as per the monitoring plan. During the review of CRF's by the trial statistician a check of the expected use of all data is made.

19 CLINICAL STUDY REPORT

All clinical data will be presented at the end of the study as data listings. These will be checked to confirm the lists accurately represents the data collected during the course of the study. The trial data will then be locked and a final data listing produced. The clinical study report will be based on the final data listings. The locked trial data may then be used for analysis and publication.

20 STUDY SITE MANAGEMENT

20.1 Study site responsibilities

The Principal Investigator (the PI or lead clinician for the study site) has overall responsibility for conduct of the study, but may delegate responsibility where appropriate to suitably experienced and trained members of the study site team. All members of the study site team must complete the Staff Contact Responsibility Sheet provided prior to undertaking any study duties. The PI must counter sign and date each entry in a timely manner, authorising staff to take on the delegated responsibilities.

20.2 Study site set up and activation

The Principal Investigator leading the investigational study site is responsible for providing all required core documentation. Mandatory Site Training organised by the Trial Office must be completed before the site can be activated. The Trial Office will check to confirm that the site has all the required study information/documentation and is ready to recruit. The site will then be notified once they are activated on the trial database and able to enter patients.

20.3 Study documentation

The Trial Office will provide an Investigator File and Pharmacy File to each investigational site containing the documents needed to initiate and conduct the study. The Trial Office must review and approve any local changes made to any study documentation including patient information and consent forms prior to use. Additional documentation generated during the course of the trial, including relevant communications must be retained in the site files as necessary to reconstruct the conduct of the trial.

21 REGULATORY AND ETHICAL CONSIDERATIONS

The Sponsor and Investigators will ensure that this protocol will be conducted in compliance with the UK Clinical Trials Regulations[1] and the applicable policies of the sponsoring organisation. Together, these implement the ethical principles of the Declaration of Helsinki (1996) and the regulatory requirements for clinical trials of an investigational medicinal product under the European Union Clinical Trials Directive.

21.1 Ethical conduct of the trial and ethics approval

The protocol, patient information sheet, consent form and any other information that will be presented to potential trial patients (e.g. advertisements or information that supports or supplements the informed consent) will be submitted to an appropriate Research Ethics Committee (REC), HRA (where required), regulatory authorities (MHRA), and host insitutions (s) for written approval. Principal Investigators will be approved by the REC.

21.2 Regulatory Authority approval

This study will be conducted under a UK Medicines and Healthcare Products Regulatory Agency (MHRA) Clinical Trials Authorisation (CTA). Approval to conduct the study will be obtained from the Responsible Authority prior to initiating the study.

21.3 NHS Research Governance

Investigators are responsible for ensuring they obtain local Trust management agreement to conduct the trial in accordance with local arrangements and policies.

21.4 Protocol amendments

Amendments are changes made to the research following initial approval. A 'substantial amendment' is an amendment to the terms of the Responsible Authority application (if applicable), the REC application, or to the protocol or any other supporting documentation, that is likely to affect to a significant degree:

- the safety or physical or mental integrity of the subjects of the trial;
- the scientific value of the trial;
- the conduct or management of the trial; or
- the quality or safety of the investigational medicinal product(s) used in the trial.

Non-substantial amendments are those where the change(s) involve only minor logistical or administrative aspects of the study.

All amendments will be generated and managed according to the Trial Office standard operating procedures to ensure compliance with applicable regulation and other requirements. Written approval will be obtained, where necessary, from the Research Ethics Committee (REC), HRA, regulatory authorities (MHRA), and host insitutions (s) for all amendments to the original approved documents.

Written confirmation of all applicable REC, HRA, regulatory and local approvals must be in place prior to implementation by Investigators. The only exceptions are for changes necessary to eliminate an immediate hazard to study patients (see below).

It is the Investigator's responsibility to update patients (or their authorised representatives, if applicable) whenever new information (in nature or severity) becomes available that might affect the patient's willingness to continue in the trial. The Investigator must ensure this is documented in the patient's medical notes and the patient is re-consented if appropriate.

21.5 Urgent safety measures

The Sponsor or Investigator may take appropriate urgent safety measures to protect trial participants from any immediate hazard to their health or safety. Urgent safety measures may be taken without prior authorisation. The

1 The Medicines for Human Use (Clinical Trials) Regulations (S.I. 2004/1031) and any subsequent amendments to it.

trial may continue with the urgent safety measures in place. **The Investigator must inform the Trial Office IMMEDIATELY if the study site initiates an urgent safety measure:**

The notification must include:

- Date of the urgent safety measure;
- Who took the decision; and
- Why the action was taken.

The Investigator will provide any other information that may be required to enable the Trial Office to report and manage the urgent safety measure in accordance with the current regulatory and ethical requirements for expedited reporting and close out. The Trial office will follow written procedures to implement the changes accordingly.

21.6 Temporary halt

The sponsor and Investigators reserve the right to place recruitment to this protocol on hold for short periods for administrative reasons **or** to declare a temporary halt. A temporary halt is defined a formal decision to:

- interrupt the treatment of subjects already in the trial for safety reasons;
- stop recruitment on safety grounds; or
- stop recruitment for any other reason(s) considered to meet the substantial amendment criteria, including possible impact on the feasibility of completing the trial in a timely manner.

The Trial Office will report the temporary halt via an expedited substantial amendment procedure. The trial may not restart after a temporary halt until a further substantial amendment to re-open is in place. If it is decided not to restart the trial this will be reported as an early termination.

21.7 Serious Breaches

The Medicines for Human Use (Clinical Trials) Regulations require the Sponsor to notify any "serious breaches" to the MHRA within 7 days of the sponsor becoming aware of the breach. A serious breach is defined as “A breach of GCP or the trial protocol which is likely to effect to a significant degree:

- the safety or physical or mental integrity of the subjects of the trial; or
- the scientific value of the trial”

Investigators must notify the Trials Office within one working day if any serious breach of GCP is suspected. The Trial Office will review the event and, if appropriate will report a serious breach to the REC, Regulatory Authority and the NHS host organisation within 7 days of the Trial Office becoming aware of the breach.

21.8 Trial Reports

This protocol will comply with all current applicable Regulatory Authority, Research Ethics Committee and Sponsor reporting requirements.

The Trial Office will determine which reports need to be circulated to Principal Investigators and other interested parties. Study sites are responsible for forwarding trial reports they receive to their local Trust as required.

22 EXPENSES AND BENEFITS

Study sites will provide expenses to patients as per local practice.

23 QUALITY ASSURANCE

23.1 Risk assessment

A risk assessment and a monitoring plan will be prepared before the study opens and will be reviewed throughout the study if necessary in the light of significant changes while the study is ongoing or in response to outcomes from monitoring activities. Monitoring plans will be amended as appropriate.

23.2 Monitoring

Regular monitoring will be performed according to the monitoring plan. Data will be evaluated for compliance with the protocol, completeness and accuracy. The Investigator and institutions involved in the study will permit study-related monitoring and provide direct on-site access to all study records and facilities if required. They will provide adequate time and space for the completion of monitoring activities.

Study sites will be monitored centrally by checking incoming data for compliance with the protocol, consistency, completeness and timing. The case report data will be validated using appropriate set criteria, range and verification checks. The study site must resolve all data queries in a timely manner. All queries relating to key outcome and safety data and any requiring further clarification will be referred back to the study site for resolution. For other non-critical data items, OCTO staff may resolve data queries centrally providing the correct answer is clear. Such changes will be clearly identified in the CRF and the study site informed.

Study sites will also be monitored remotely and/or by site visit as necessary to ensure their proper conduct of the trial. OCTO staff will be in regular contact with site personnel to check on progress and deal with any queries that they may have. Monitoring reports will be sent to the site in a timely fashion. The Investigator is expected to action any points highlighted through monitoring and must ensure that corrective and preventative measures are put into place as necessary to achieve satisfactory compliance.

23.3 Audit and Regulatory Inspection

All aspects of the study conduct may be subject to internal or external quality assurance audit to ensure compliance with the protocol, GCP requirements and other applicable regulation or standards. It may also be subject to a regulatory inspection. Such audits or inspections may occur at any time during or after the completion of the study. Investigators and their host Institution(s) should understand that it is necessary to allow auditors/inspectors direct access to all relevant documents, study facilities and to allocate their time and the time of their staff to facilitate the audit or inspection visit. Anyone receiving notification of a Regulatory Inspection that will (or is likely to) involve this trial must inform the Trial Office without delay.

24 RECORDS RETENTION & ARCHIVING

During the clinical trial and after trial closure the Investigator must maintain adequate and accurate records to enable the conduct of a clinical trial and the quality of the research data to be evaluated and verified. All essential documents must be stored in such a way that ensures that they are readily available, upon request for the minimum period required by national legislation or for longer if needed. The medical files of trial subjects must be retained in accordance with applicable national legislation and the host institution policy.

Retention and storage of laboratory records for clinical trial samples must also follow these guidelines. Retention and storage of central laboratory records supporting PD endpoints and the disposition of samples donated via the trial must also comply with applicable legislation and Sponsor requirements.

It is the University of Oxford's policy to store data for a minimum of 5 years. Investigators may not archive or destroy study essential documents or samples without written instruction from the Trial Office.

25 PATIENT CONFIDENTIALITY

Personal data recorded on all documents will be regarded as confidential, and to preserve each patient's anonymity, only their initials and date of birth will be recorded on the CRFs.

The Investigator site must maintain the patient's anonymity in all communications and reports related to the research. The Investigator site team must keep a separate log of enrolled patients' personal identification details as necessary to enable them to be tracked. These documents must be retained securely, in strict confidence. They form part of the Investigator Site File and are not to be released externally.

26 STUDY FUNDING

This trial is being run by the Oncology Clinical Trials Office (OCTO) at the University of Oxford and is being funded by the Oxford NIHR Biomedical Research Centre, CRUK Oxford Centre and an educational grant from the Merck Sharp & Dohme Corp Investigator Studies Programme. Any additional NHS clinical service support costs of patient care whilst on study should be met by the host study site.

27 SPONSORSHIP AND INDEMNITY

27.1 Sponsorship

The Sponsor will provide written confirmation of Sponsorship and authorise the trial commencement once satisfied that all arrangements and approvals for the proper conduct of the trial are in place. A separate study delegation agreement, setting out the responsibilities of the Chief Investigator and Sponsor will be put in place between the parties.

27.2 Indemnity

The University has a specialist insurance policy in place which would operate in the event of any participant suffering harm as a result of their involvement in the research (Newline Underwriting Management Ltd, at Lloyd's of London). NHS indemnity operates in respect of the clinical treatment that is provided.

27.3 Contracts/Agreements

This trial is subject to the Sponsor's policy requiring that written contracts/agreements are agreed formally by the participating bodies as appropriate. A Clinical Trial Agreement (CTA) will be placed between the Sponsor and participating organisations prior to site activation.

The Sponsor will also set up written agreements with any other external third parties involved in the conduct of the trial as appropriate.

28 PUBLICATION POLICY

The Sponsor will retain ownership of all data arising from this trial. The intention is to publish this research in a specialist peer reviewed scientific journal on completion of the study. The results may also be presented at scientific meetings and/or used for a thesis. The Investigators will be involved in reviewing drafts of the manuscripts, abstracts, press releases and any other publications arising from the trial and retain final editorial control. All publications/presentations should be shared with MSD prior to any presentations/submissions, allowing time for review (ideally 2 weeks for abstracts or posters and 4 weeks for manuscripts). Authors will acknowledge the study sponsor and funding bodies as appropriate.

29 REFERENCES

1. Cancer Research UK Bladder cancer statistics. http://www.cancerresearchuk.org/health-professional/cancer-statistics/statistics-by-cancer-type/bladder-cancer, Accessed June 2015.

2. Kirkali, Z. *et al.* Bladder cancer: Epidemiology, staging and grading, and diagnosis. *Urology* **66,** 4–34 (2005).

3. Treatment of non-muscle invasive bladder cancer. *uptodate.com* at <http://www.uptodate.com/contents/treatment-of-non-muscle-invasive-bladder-cancer?source=see_link&sectionName=RISK+STRATIFICATION&anchor=H10#H10>, Accessed June 2015

4. Sylvester, R. J., Oosterlinck, W. & van der Meijden, A. P. M. A single immediate postoperative instillation of chemotherapy decreases the risk of recurrence in patients with stage Ta T1 bladder cancer: a meta-analysis of published results of randomized clinical trials. *The Journal of Urology* **171,** 2186–90 (2004).

5. Babjuk, M. *et al.* EAU guidelines on non-muscle-invasive urothelial carcinoma of the bladder: update 2013. *Eur. Urol.* **64,** 639–653 (2013).

6. Sylvester, R. J. *et al.* Predicting recurrence and progression in individual patients with stage Ta T1 bladder cancer using EORTC risk tables: a combined analysis of 2596 patients from seven EORTC trials. *Eur. Urol.* **49,** 466– 477 (2006).

7. Morales, A., Eidinger, D. & Bruce, A. W. Intracavitary Bacillus Calmette-Guerin in the treatment of superficial bladder tumors. *The Journal of Urology* **116,** 180–183 (1976).

8. Mitropoulos, D. N. Novel insights into the mechanism of action of intravesical immunomodulators. *In Vivo* **19,** 611–621 (2005).

9. Taniguchi, K. *et al.* Systemic immune response after intravesical instillation of bacille Calmette-Guérin (BCG) for superficial bladder cancer. *Clin. Exp. Immunol.* **115,** 131–135 (1999).

10. Silvester, R. J., Van der Meijden, A. & Lamm, D. L. *Intravesical bacillus Calmette-Guérin reduces the risk of progression in patients with superficial bladder cancer: a meta-analysis of the published results of ….* (J Urol, 2002).

11. Patard, J.-J. *et al.* Intravesical Bacillus Calmette-Guerin Treatment Improves Patient Survival in T1G3 Bladder Tumours. *Eur. Urol.* **41,** 635–642 (2002).

12. Shelley, M. *et al. Intravesical Bacillus Calmette-Guérin in Ta and T1 bladder cancer*. (John Wiley & Sons, Ltd, 2000). doi:10.1002/14651858.CD001986

13. Witjes, J. A. Management of BCG Failures in Superficial Bladder Cancer: A Review. *Eur. Urol.* **49,** 790–797 (2006).

14. Disis, M. L. Immune regulation of cancer. *J. Clin. Oncol.* **28,** 4531–4538 (2010).

15. Bubenik, J., Perlmann, P., Helmstein, K. & Moberger, G. Immune response to urinary bladder tumours in man. *International Journal of Cancer* **5,** 39–46 (1970).

16. Cancer Genome Atlas Research Network. Comprehensive molecular characterization of urothelial bladder carcinoma. *Nature* **507,** 315–322 (2014).

17. Thompson, R. H. *et al.* PD-1 is expressed by tumor-infiltrating immune cells and is associated with poor outcome for patients with renal cell carcinoma. *Clin Cancer Res* **13,** 1757–1761 (2007).

18. Fukunaga, A. *et al.* CD8+ Tumor-Infiltrating Lymphocytes Together with CD4+ Tumor-Infiltrating Lymphocytes and Dendritic Cells Improve the Prognosis of Patients with Pancreatic Adenocarcinoma. *Pancreas* **28,** e26 (2004).

19. Zhang, L. *et al.* Intratumoral T Cells, Recurrence, and Survival in Epithelial Ovarian Cancer. *N Engl J Med* **348,** 203–213 (2003).

20. Faraj, S. F. *et al.* Assessment of Tumoral PD-L1 Expression and Intratumoral CD8+ T Cells in Urothelial Carcinoma. *Urology* **85,** 703.e1–703.e6 (2015).

21. Sharma, P. *et al.* CD8 tumor-infiltrating lymphocytes are predictive of survival in muscle-invasive urothelial carcinoma. *Proc. Natl. Acad. Sci. U.S.A.* **104,** 3967–3972 (2007).

22. Keir, M. E., Butte, M. J., Freeman, G. J. & Sharpe, A. H. PD-1 and Its Ligands in Tolerance and Immunity. *http://dx.doi.org/10.1146/annurev.immunol.26.021607.090331* **26,** 677–704 (2008).

23. Dong, H. *et al.* Tumor-associated B7-H1 promotes T-cell apoptosis: A potential mechanism of immune evasion. *Nature Medicine* **8,** 793–800 (2002).

24. Iwai, Y. *et al.* Involvement of PD-L1 on tumor cells in the escape from host immune system and tumor immunotherapy by PD-L1 blockade. *Proc. Natl. Acad. Sci. U.S.A.* **99,** 12293–12297 (2002).

25. Iwai, Y., Terawaki, S. & Honjo, T. PD-1 blockade inhibits hematogenous spread of poorly immunogenic tumor cells by enhanced recruitment of effector T cells. *Int. Immunol.* **17,** 133–144 (2005).

26. Nomi, T. *et al.* Clinical significance and therapeutic potential of the programmed death-1 ligand/programmed death-1 pathway in human pancreatic cancer. *Clin Cancer Res* **13,** 2151–2157 (2007).

27. Wong, R. M. *et al.* Programmed death-1 blockade enhances expansion and functional capacity of human melanoma antigen-specific CTLs. *Int. Immunol.* **19,** 1223–1234 (2007).

28. Blank, C. *et al.* Blockade of PD-L1 (B7-H1) augments human tumor-specific T cell responses in vitro. *International Journal of Cancer* **119,** 317–327 (2006).

29. Hamid, O. *et al.* Safety and Tumor Responses with Lambrolizumab (Anti–PD-1) in Melanoma. *N Engl J Med* **369,** 134–144 (2013).

30. Topalian, S. L. *et al.* Survival, durable tumor remission, and long-term safety in patients with advanced melanoma receiving nivolumab. *J. Clin. Oncol.* **32,** 1020–1030 (2014).

31. Garon, E. B. *et al.* Pembrolizumab for the Treatment of Non–Small-Cell Lung Cancer. *N Engl J Med* 150419053123009 (2015). doi:10.1056/NEJMoa1501824

32. Gettinger, S. N., Shepherd, F. A., Antonia, S. J. & Brahmer, J. R. *First-line nivolumab (anti-PD-1; BMS-936558, ONO-4538) monotherapy in advanced NSCLC: Safety, efficacy, and correlation of outcomes with PD-L1* (J Clin Oncol, 2014).

33. Motzer, R. J. *et al.* Nivolumab for Metastatic Renal Cell Carcinoma: Results of a Randomized Phase II Trial. *J. Clin. Oncol.* **33,** 1430–1437 (2015).

34. Chow, L. Q. *et al.* LBA31A Phase 1B study of Pembrolizumab in patients with human papilloma virus positive and negative head and neck cancer. *Ann Oncol* **25,** mdu438.32 (2014).

35. Powles, T. *et al.* MPDL3280A (anti-PD-L1) treatment leads to clinical activity in metastatic bladder cancer. *Nature* **515,** 558–562 (2014).

36. Plimack, E. R., Gupta, S., Bellmunt, J., Berger, R., Montgomery, B., Gonzalez, E. J., et al. (2014). LBA23A Phase 1B study of Pembrolizumab in patients with advanced urothelial tract cancer. *Annals of Oncology*, *25*(suppl 4), mdu438.24. doi:10.1093/annonc/mdu438.24

37. Hughes, O. D. *et al.* Targeting superficial bladder cancer by the intravesical administration of copper-67-labeled anti-MUC1 mucin monoclonal antibody C595. *JCO* **18,** 363–370 (2000).

38. Crawford, E. D. Intravesical therapy for superficial cancer: need for more options. *JCO* **20,** 3185–3186 (2002).

39. Gofrit, O. N., Zorn, K. C., Shikanov, S. & Steinberg, G. D. Marker Lesion Experiments in Bladder Cancer—What Have We Learned? *The Journal of Urology* **183,** 1678–1685 (2010).

APPENDIX 1: ECOG PERFORMANCE SCALE

Activity Performance Description	Score
Fully active, able to carry out all on all pre-disease performance without restriction.	0
Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g. light housework, office work.	1
Ambulatory and capable of all self-care, but unable to carry out any work activities. Up and about more than 50% of waking hours.	2
Capable of only limited self-care. Confined to bed or chair more than 50% of waking hours.	3
Completely disabled. Cannot carry out any self-care. Totally confined to bed or chair.	4

APPENDIX 2: EAU RISK CATEGORIES

Risk Category	Definition
Low-risk tumours	Primary, solitary, Ta, LG/G1, <3cm, no CIS
Intermediate-risk tumours	All cases between categories of low and high risk
High-risk tumours	Any of the following: • T1 tumours • HG/G3 tumours • CIS • Multiple and recurrent and large (>3cm) Ta,G1, G2 tumours (all these conditions must be present)

Adapted from EAU guidelines[5]

APPENDIX 3: EORTC RISK TABLES

Factor	Recurrence score	Progression score
Number of tumours		
1	0	0
2 to 7	3	3
≥ 8	6	3
Tumour size		
≤ 3cm	0	0
≥ 3cm	3	3
Prior recurrence rate		
Primary	0	0
≤ 1/year	2	2
> 1/year	4	2
T category		

Ta	0	0
T1	1	4
CIS		
No	0	0
Yes	1	6
Grade		
G1	0	0
G2	1	0
G3	2	5
Total score	0-17	0-23

Probability of recurrence and progression according to total score:

Recurrence score	**Probability of recurrence at 1 year (%) (95% CI)**	**Probability of recurrence at 5 years (%) (95% CI)**
0	15 (10-19)	31 (24-37)
1-4	24 (21-26)	46 (42-49)
5-9	38 (35-41)	62 (58-65)
10-17	61 (55-67)	78 (73-84)

Progression score	**Probability of progression at 1 year (%) (95% CI)**	**Probability of progression at 5 years (%) (95% CI)**
0	0.2 (0-0.7)	0.8 (0-1.7)
2-6	1.0 (0.4-1.6)	6 (5-8)
7-13	5 (4-7)	17 (14-20)
14-23	17 (10-24)	45 (35-55)