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Investigating side effects of checkpoint inhibitors and surveillance imaging in high-risk melanoma patients

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Abstract

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Historically, the prognosis for metastatic cutaneous malignant melanoma (CMM) was very poor, and postoperative follow-up relied mainly on clinical examination, as early detection of recurrence offered limited benefit due to lack of effective systemic therapies. The introduction of targeted therapies and immune checkpoint inhibitors (CPIs) has markedly improved outcomes in advanced CMM patients. Consequently, many countries have added whole-body imaging to follow-up protocols after radical surgery for high-risk CMM, although recommendations vary due to limited scientific evidence.

CPIs have become a cornerstone in the treatment of several solid malignancies. Despite their efficacy, they may cause immune-related adverse events (IRAEs). Most IRAEs are low grade and manageable, but some are long-lasting and a few even fatal.

Papers I–III are multicenter retrospective cohort studies including approximately 600 patients with advanced solid tumors treated with CPIs. Paper I evaluated a simplified frailty score based on age, performance status, and comorbidity (Charlson Comorbidity Index) as a predictor of IRAEs. Frail patients had a significantly higher risk of developing any-grade and multiple IRAEs, but not severe (grade 3–4) toxicity or treatment discontinuation. The individual score components were not independently associated with IRAEs.

Paper II examined the association between multiple IRAEs and treatment efficacy, measured by progression-free survival (PFS) and overall survival (OS). Patients with multiple IRAEs experienced significantly improved PFS and OS compared with those experiencing one or no IRAEs, suggesting that multiple IRAEs may serve as a surrogate marker of treatment efficacy.

Paper III compared toxicity profiles of PD-1 and PD-L1 inhibitors in a real-world setting. PD-L1 inhibitors were associated with a significantly lower risk of grade ≥2 IRAEs and treatment discontinuation due to toxicity, supporting a more favorable tolerability profile.

Paper IV presents an interim analysis of a nationwide randomized phase III trial (TRIM, NCT03116412) evaluating scheduled whole-body imaging in stage IIB–III CMM after radical surgery. At a median follow-up of 33.6 months, no significant differences were observed in relapse-free, distant metastasis-free, or overall survival between standard and imaging-based follow-up. However, follow-up remains relatively short, and few patients have completed five years of follow-up.

Keywords: checkpoint inhibitor, immune related adverse events, surveillance imaging, cutaneous malignant melanoma

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Att våga är att förlora fotfästet en stund.
Att inte våga är att förlora sig själv.
Søren Kierkegaard

To my family

List of Papers

This thesis is based on the following papers, which are referred to in the text by their Roman numerals.

I. Olsson Ladjevardi C, Koliadi A, Rydén V, Inan El-Naggar A, Digkas E, Valachis A*, Ullenhag GJ* (*shared senior position). (2023) Predicting immune-related adverse events using a simplified frailty score in cancer patients treated with checkpoint inhibitors: A retrospective cohort study. *Cancer Medicine,* 12(12):13217-13224

II. Olsson Ladjevardi C, Koliadi A, Rydén V, Inan El-Naggar A, Digkas E, Valachis A*, Ullenhag GJ* (*shared senior position). (2024) Multiple immune-related adverse events secondary to checkpoint inhibitor therapy in patients with advanced cancer: association with treatment effectiveness. *Frontiers in Oncology, 14:1399171*

III. Cecilia Olsson Ladjevardi, Marcus Skribek, Anthoula Koliadi, Viktoria Rydén, Ali Inan El-Naggar, Evangelos Digkas, Antonios Valachis*, Gustav J Ullenhag* (*shared senior position). (2024) Differences in immune-related toxicity between PD-1 and PD-L1 inhibitors: a retrospective cohort study. *Cancer Immunology, Immunotherapy, 74:14.*

IV. Cecilia Olsson Ladjevardi*, Ylva Naeser*,….Gustav Ullenhag, (*joint first authors). (2025) Physical examinations and whole-body imaging versus physical examinations alone during follow-up after radical surgery of stage IIB–C and III cutaneous malignant melanoma (TRIM): an interim analysis of a multicenter, randomized, phase 3 trial in Sweden. *The Lancet Oncology, 26:1501-10*

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Contents

Introduction 11

Cutaneous malignant melanoma 12
- Epidemiology 12
- Aetiology 13
- Risk factors 15
- Staging and prognosis 16
- Treatment of resectable CMM 16
  - Surgery 16
  - Neoadjuvant systemic therapy 17
  - Adjuvant systemic therapy 18
- Treatment of generalized disease 20
  - Immunotherapy 20
  - Targeted therapy 21
  - Chemotherapy 22
  - Additional treatment options 22

Cancer immunology 24
- Background 24
- Immunosurveillance and immune escape 24
- The cancer immunity cycle 25

Cancer immunotherapy 28
- Checkpoint inhibitors 28
  - CTLA-4 inhibition, mechanism of action 29
  - PD-1/PD-L1 inhibition, mechanism of action 31
  - Other immune checkpoints 33

Toxicity and efficacy of checkpoint inhibitors 35
- Incidence of IRAEs due to type of CPI 35
- Incidence of IRAEs in relation to dose 37
- Organ specific IRAEs in relation to type of CPI 38
- Organ specific IRAEs in relation to tumor type 39
- Timeline of IRAE onset 40
- Treatment of IRAEs and impact on efficacy 41
- Impact of antibiotic use on efficacy of checkpoint inhibitors 42
- Predictive markers for treatment response 43
- Predictive markers of IRAEs 44

Aims of the thesis....46

Patients and methods....47
Paper I, II and III....47
Paper IV....48

Results....49
Paper I and II....49
Study cohort characteristics....49
Paper I....49
Frequency, outcomes and prediction of IRAEs....49
Paper II....49
Characteristics of multiple IRAEs and impact on efficacy....49
Paper III....51
Characteristics of the study cohort....51
Occurrence of IRAEs with PD-1 versus PD-L1 inhibitors....51
Paper IV....52

Discussion....53

Conclusions....56

Future perspectives....57

Populärvetenskaplig sammanfattning på svenska
(Summary in Swedish)....59

Acknowledgements....62

References....65

Abbreviations

AJCC	American Joint Committee of Cancer
APC	Antigen presenting cells
CCI	Charlson Comorbidity Index
CLND	Complete lymph node dissection
CMM	Cutaneous malignant melanoma
CPI	Checkpoint inhibitor
ctDNA	Circulating tumor DNA
CTLA-4	Cytotoxic T lymphocyte-associated protein 4
DC	Dendritic cell
DMFS	Distant metastasis free survival
dMMR	Defect mismatch repair
EMA	European medicines agency
EMR	Electronic medical records
EVF	Event-free survival
FDA	Food and Drug Administration
FMT	Fecal microbiota transplantation
HCC	Hepatocellular carcinoma
IARC	World Health Organization International Agency for Research on Cancer
IFN	Interferon
IL-2	Interleukin 2
IL-6	Interleukin 6
IRAE	Immune related adverse events
ITB	Immortal time bias
LAG-3	Lymphocyte activation gene-3
LRFS	Locoregional relapse free survival
mAbs	Monoclonal antibodies
MHC	Major histocompatibility complex
MSI-H	Microsatellite instability high
NK	Natural killer
NSCLC	Non-small cell lung cancer
ORR	Overall response rate
OS	Overall survival

PD-1	Programmed death 1
PD-L1	Programmed death-ligand 1
PFS	Progression free survival
PMT-O	Pooled median time to onset
PMT-R	Pooled median time to resolution
PS	Performance status
RCC	Renal cell carcinoma
RFS	Recurrence free survival
TCR	T-cell receptor
TH	T-helper
TMB	Tumor mutation burden
TME	Tumor microenvironment
TNF	Tumor necrosis factor
Tregs	Regulatory T cells
TTO	Time to onset
T-VEC	Talimogene laherparepvec
UV	Ultraviolet
VEGF	Vascular endothelial growth factor

Introduction

The incidence of cutaneous malignant melanoma (CMM) continues to rise globally. Historically, the prognosis for metastatic CMM was very poor and postoperative follow-up relied mainly on clinical examinations, as early detection of recurrence provided little benefit given the limited treatment options.

During the past decade, the introduction of targeted therapies and immunotherapies has markedly improved survival in advanced disease. Consequently, many countries have introduced whole-body imaging in the follow-up protocols after radical surgery of high-risk CMM, although protocols vary widely due to limited scientific evidence. Even though distant metastases might be detected earlier, especially in asymptomatic patients, in patients undergoing surveillance with whole-body imaging, the impact on prognosis and survival is unclear.

Immunotherapies with checkpoint inhibitors (CPIs), were first approved for patients with metastatic CMM in 2011. Today indications range from a broad variety of malignancies and in different clinical settings. Although highly effective, CPIs may cause side effects called immune related adverse events (IRAEs), of varying severity. Knowledge regarding risk factors for developing of IRAEs is scarce. While the development of IRAEs is associated with improved treatment efficacy, the impact of multiple IRAEs and treatment outcomes is less studied. Furthermore, the CPIs targeting programmed cell death protein 1 (PD-1) and programmed death-ligand 1 (PD-L1) act through similar, but not identical, mechanisms, with clinical trials showing slight differences in both efficacy and toxicity, though real-world studies are lacking.

This doctoral project addresses these knowledge gaps by evaluating whether a simplified frailty score predicts IRAEs in patients treated with CPIs (paper I), by examining the association between multiple IRAEs and treatment outcome (paper II), by comparing efficacy and toxicity of PD-1 versus PD-L1 inhibitors in a real-world setting (paper III) and by assessing whether adding whole-body imaging in the follow-up after radical surgery of CMM improves survival by earlier detection of recurrence (paper IV).

Cutaneous malignant melanoma

Epidemiology

The incidence of CMM has been steadily rising across North America, Europe and Oceania since the 1960s [1-5]. Globally, an estimated 320 000 new cases and 59 000 melanoma-related deaths were reported in 2022, making melanoma the 17th most common malignancy worldwide. The highest age-standardized incidence rates are observed in Australia-New Zealand, at 36 per 100 000 person-years, followed by Northern Europe and North America at 18 versus 16 per 100 000 person-years [6]. If this upward trend continues, the number of diagnosed cases is projected to increase by 50% by 2040 [7].

In the Nordic countries CMM incidence is highest in Denmark, Norway and Sweden, while it is lower in Finland and Iceland [6]. In Sweden, CMM was the fourth most common malignancy among women and third most common among men in 2024 [8]. Over the past decade, the incidence has risen rapidly, with a 60% increase from 3800 cases in 2014 to 6000 cases in 2024 [9]. Despite this, the overall mortality rate has remained steady since several years [10].

Interestingly, a recent cohort study analyzing melanoma cases in Sweden between 1990 and 2022 found a significant downward trend in both melanoma incidence and mortality among individuals aged 30–49 years. This trend may reflect the positive impact of improved ultraviolet (UV) protection, public health campaigns and the availability of effective treatments. However, the authors also highlight that shifting population demographics could contribute to these findings [11].

The median age at diagnosis of CMM in Sweden during 2020-2024 was 67 years for women and 71 years for men [10]. Most CMMs are thin (≤1 mm) at the time of diagnosis, reflecting early detection in many cases. In 2024, the incidence of CMM was slightly higher in men than in women, with 3200 new cases reported in men and 2800 in women [9]. Additionally, men have a worse prognosis compared to women, even after adjusting for more advanced disease at diagnosis [12, 13].

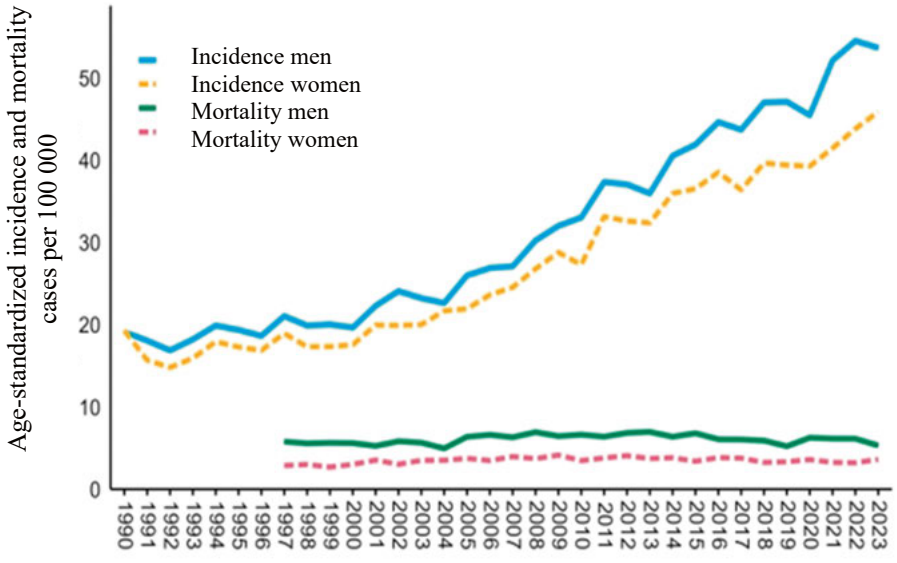

Figure 1. Age-standardized incidence and mortality from invasive malignant melanoma in Sweden for men and women, 1990-2023. Reprinted from The Swedish Melanoma Register Report 2025 with permission.

Aetiology

Melanocytes are neural crest-derived cells in the basal epidermis that produce melanin which protects the skin from UV radiation. UV-induced DNA damage stimulates keratinocytes to release melanocyte-stimulating hormone, which binds melanocortin 1 receptor on melanocytes, increasing melanin production. The development of CMM results from genetic alterations affecting key regulatory pathways [14].

Malignant melanoma has one of the highest somatic mutational burdens among cancers [15]. Those associated with chronically sun-exposed skin exhibit a greater mutational burden than CMMs on less sun-exposed areas. These mutations predominantly activate two main signaling pathways: The RAS/RAF/MEK/ERK signaling pathway (also called MAPK pathway) and the phosphoinositol-3-kinase (P13K)/AKT pathway. The MAPK pathway is the most frequently dysregulated with up to 90% of CMMs showing aberrant activation of this pathway. It regulates gene expressions involved in cell proliferation, differentiation and survival, while the P13K pathway is involved in cellular homeostasis [14, 16].

In CMM, BRAF mutations, present in approximately 50% of cases, are the leading cause of MAPK pathway activation, most commonly V600E and

V600K. V600E is more frequent in younger patients and women and is not classically related to chronic sun-exposure, whereas V600K is linked to cumulative sun exposure and increasing age [17]. NRAS mutations occur in 15-30% of melanomas, with a higher prevalence in amelanotic and nodular subtypes [16, 17]. KIT mutations (2-8%) are more common in mucosal, acral and chronically sun exposed CMMs [16, 18]. The tumor suppressor gene NF1 is mutated in about 10-15% of CMMs, predominantly in those arising from chronically sun-damaged skin and may co-occur with BRAF and NRAS mutations [14, 16].

TERT promotor mutations enhance telomere maintenance, preventing cell aging and apoptosis. The prevalence of this mutation is associated with poor prognosis. Mutations in the tumor suppressor gene CKDN2A can be found in patients with hereditary melanoma. These mutations also significantly increase the risk of pancreatic carcinoma. The TP53 gene normally acts as a tumor suppressor. Somatic mutations in TP53 are associated with tumor progression. PTEN is another tumor suppressor gene and mutations in PTEN are commonly detected in the vertical growth phase and in metastatic CMM [16, 17].

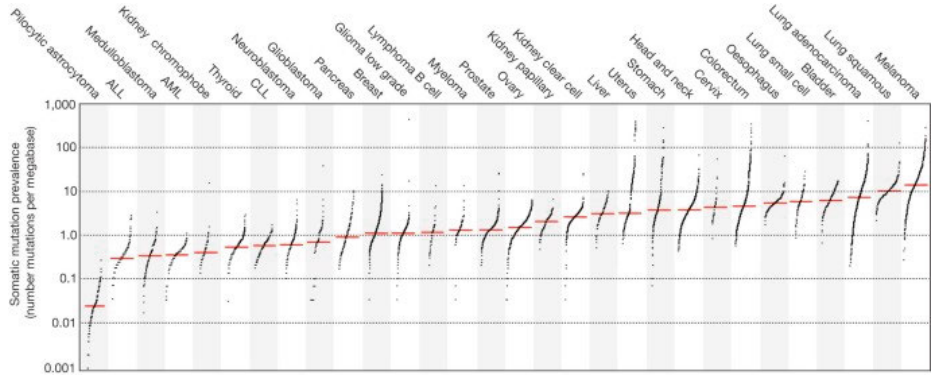

Figure 2. The prevalence of somatic mutations across different cancer types. Every dot represents a sample whereas the red horizontal lines are the median numbers of mutations in the respective cancer types. The vertical axis (log scaled) shows the number of mutations per megabase whereas the different cancer types are ordered on the horizontal axis based on their median numbers of somatic mutations. Abbreviations: ALL, acute lymphoblastic leukaemia; AML, acute myeloid leukaemia; CLL, chronic lymphocytic leukaemia. Reprinted from the article "Signatures of mutational processes in human cancer" with permission from publisher [15].

Melanoma progression is further driven by interactions with the tumor microenvironment (TME) and the immune system. Overexpression of proteins that promote invasion and tissue infiltration, facilitates local spread and metastasis [14, 17]. The gut microbiota may also influence carcinogenesis [19]. Additionally, melanoma cells evade immune detection and

destruction through multiple mechanisms, enabling tumor survival and progression [20]. These processes are discussed in greater detail in the chapter *Cancer Immunology*.

Risk factors

The risk factors for development of CMM can be broadly categorized as either patient-related or environmental. The primary environmental risk factor is exposure to UV radiation, particularly UV-B rays, from natural sunlight [17]. The pattern and timing of sun exposure significantly influence the risk, with intense, intermittent exposure being more strongly associated with CMM development than chronic, continuous exposure [21, 22]. Childhood or adolescence sunburns further amplify this risk, and individuals who have experienced more than five severe sunburns face an estimated twofold enhance in their likelihood of developing CMM [23]. Additionally, studies have demonstrated an association between the use of sunbeds and a heightened risk of melanoma, prompting the World Health Organization International Agency for Research on Cancer (IARC) to classify UV radiation from tanning beds as a human carcinogen in 2009 [24]. Moreover, occupational exposure to pesticides has been associated with an increased risk of developing acral melanoma [25].

The patient-related risk factors for the development of CMM include the following:

- Phenotypic characteristics: Fair skin, blond or red hair, freckling and blue or green eyes [26].
- Naevi: A high number of naevi (<50-100) as well as presence of large naevi (>5mm), significantly increase risk [26].
- Genetic predispositions: Mutations in the CDKN2A gene are the most common in patients with a history of CMM, accounting for approximately 20% of families with a history of CMM. Mutations in TP53 are also implicated [17]. Other inherited conditions associated with a heightened risk include xeroderma pigmentosum, familial retinoblastoma, Lynch syndrome type II and Li-Fraumeni cancer syndrome [14].
- Family history: A family history of melanoma is a significant risk factor [26].
- Immunosuppression: Immunosuppression due to diseases, such as AIDS, and the use of immunosuppressive therapy following organ transplantation, elevate the risk [17].

- History of previous skin cancers: Individuals with a history of non-melanoma skin cancer or previous melanoma (including melanoma in situ) are at increased risk [17, 26].

Staging and prognosis

The staging of CMM is based on the TNM-system, as defined by the American Joint Committee of Cancer (AJCC), with the most recent update published in 2017 as the AJCC eight edition (AJCC8) [27, 28]. The T-stage is determined by the primary tumor thickness (Breslow) and the presence or absence of ulceration. The N-stage describes the extent of locoregional metastases, and the M-stage reflects the presence of distant metastases, categorized based on the anatomic site of metastases into four subgroups.

Stage groups combine the T, N and M classifications and are denoted by Roman numerals I-IV. Subgroups are IA-IB (stage I), IIA-IIC (stage II), and IIIA-IIID (stage III). Stage IV indicates distant metastases, regardless of T or N stage, and has no subgroups.

Prognosis after radical surgery depends mainly on the tumor stage [27, 29] and guides both adjuvant therapy and follow-up. Melanoma with a high risk of recurrence after surgery is often defined as stage IIB, IIC and all stage III. The risk of recurrence at 10 years for stage IIB and IIC is 33% vs 37%. For stage III, recurrence rates vary widely, ranging from 30% to 71%, and increase progressively with each substage (IIIA-D). The data are based on a cohort of patients where the vast majority were diagnosed before the introduction of adjuvant treatments [29].

Treatment of resectable CMM

For many years, surgery with curative intent was the standard treatment for patients with resectable CMM, including those with oligometastatic stage IV disease. However, in 2018 systemic adjuvant treatment was introduced in Sweden, and in early 2023 the Swedish national guidelines were updated to recommend neo-adjuvant treatment for patients in specific stage-groups [30].

Surgery

Clinically suspected CMM is primary excised with a margin of 2-5 mm. If the pathology report confirms the diagnosis of CMM, a wide local excision is subsequently performed according to tumor thickness: 1 cm for tumors $\leq$ 2.0 mm and 2 cm for tumors > 2.0 mm, as recommended by the Swedish national

guidelines [31]. Ongoing trials are evaluating narrower margins. The international MelMarT-study randomizes patients with melanomas >1 mm to 10 vs 20 mm margin (completion expected in 2026) [32]. The Swedish WiseOrWide trial includes patients with tumors ≤ 1 mm and ≥ 1.5 mm clear primary margins, randomizing to no further surgery or 1 cm re-excision [33].

In situations with synchronous regional metastases (nodal or non-nodal), a 10 mm wide excision is recommended regardless of the primary tumor's thickness.

Sentinel lymph node biopsy is recommended for patients with tumors > 1 mm (≥T2) and adds prognostic, but more importantly, predictive value [31, 34]. Complete lymph node dissection (CLND) is reserved for clinically evident lymph node metastases. For patients with a positive sentinel node biopsy but no clinical nodal disease, CLND is not recommended, as it offers no survival benefit compared to active ultrasound surveillance of the nodal basin [35, 36].

Neoadjuvant systemic therapy

The phase II OpACIN trial randomized 20 patients with macroscopic stage III CMM to ipilimumab (3 mg/kg) plus nivolumab (1 mg/kg) given either fully adjuvant (4 cycles) or split (2 neoadjuvant + 2 adjuvant). Neo-adjuvant therapy showed better outcomes but high toxicity in both arms [37]. In the subsequent phase II OpACIN-neo (n=89), different preoperative dosing regimens of ipilimumab and nivolumab were evaluated, with the aim of identifying a less toxic yet equally effective schedule. Patients underwent surgery at week six. Ipilimumab 1 mg/kg plus nivolumab 3 mg/kg for two neoadjuvant doses emerged as the superior regimen [38]. A survival update from 2023 reported that the median recurrence free survival (RFS) or overall survival (OS) were not reached in either trial. In OpACIN (69 months median follow-up), the estimated 5-year RFS and OS were 70% and 90% (neoadjuvant) vs 60% and 70% (adjuvant) respectively. In OpACIN-neo (47 months median follow-up), the estimated 3-year RFS and OS for the overall cohort were 82% and 92% respectively. Among patients with a pathologic response, the estimated 3-year RFS was 95% versus 37% in non-responders [39].

The phase II SWOG S1801 trial (n=313) randomized patients with resectable macroscopic stage III or IV melanoma to 3 cycles of pembrolizumab before surgery followed by 15 cycles postoperatively (neoadjuvant-adjuvant group), or surgery followed by 18 cycles of pembrolizumab (adjuvant group). Two-year event-free survival (EVF) was significantly higher in the neoadjuvant-adjuvant group compared to the adjuvant group, 72% versus 49%, with similar toxicity between the arms [40]. Updated results from the 3-year EVF confirm sustained benefit for the neoadjuvant-adjuvant group [41]. In the phase III NADINA trial (n=423) patients with resectable macroscopic stage

III melanoma received two cycles of ipilimumab (80 mg) plus nivolumab (240 mg) before surgery (neoadjuvant arm), or surgery followed by 12 cycles of nivolumab (adjuvant arm). Only non/partial responders in the neoadjuvant arm received adjuvant therapy. Twelve-month EVF was 83.7% in the neoadjuvant group and 57.2% in the adjuvant group. Grade ≥ 3 adverse events occurred in 29.7% of patients in the neoadjuvant group versus 14.7% in the adjuvant group [42]. The 2-year EVF, presented at the ESMO conference 2025, was 77.3% in the neoadjuvant group and 55,7% in the adjuvant group [43].

After publication of SWOG S1801 in early 2023, the Swedish melanoma guidelines were updated to include recommendations for neoadjuvant immunotherapy in resectable stage III/IV CMM [31].

Adjuvant systemic therapy

Immunotherapy

Adjuvant nivolumab and pembrolizumab following radical surgery for stage III (AJCC 7) CMM are approved by United States Food and Drug Administration (FDA) and European Medicines Agency (EMA), based on two large phase III trials. In the CheckMate 238 trial, patients with complete resection of stage IIIB-C or IV melanoma were randomly assigned to receive either nivolumab or ipilimumab for up to a year. The 12-month RFS was 70.5% in the nivolumab group and 60.8% in the ipilimumab group. Treatment related grade 3 or 4 adverse events occurred in 45.9% of those in the ipilimumab group and 14.4% in the nivolumab group [44]. At a minimum follow-up of nearly 9 years, RFS with nivolumab remained superior to ipilimumab (HR=0.76; 95% CI, 0.63-0.9; 9-year rates of 44% vs. 37%). Nine-year OS rates were 69% vs 65% for the nivolumab vs ipilimumab group (HR=0.87; 95%CI, 0.69-1.11). Fewer patients in the nivolumab group than in the ipilimumab group received subsequent systemic therapy (37.3 % vs. 44.6%) [45]. In the KEYNOTE-054 trial 1019 patients with complete resection of stage III (AJCC 7) CMM were randomized to receive pembrolizumab or placebo for up to a year. Long-term follow-up demonstrates a sustained benefit for the adjuvant compared to the placebo group with 7-year RFS of 50% (95% CI, 46-55%) versus 36% (95% CI, 32-41%). Distant metastasis free survival at 7 years was 54% (95% CI, 50-59%) in the pembrolizumab group and 42% (95% CI, 37-46%) in the placebo group [46].

Adjuvant PD-1 inhibition in stage II B and C (AJCC 8) has been evaluated in two large phase III trials. The results formed the basis for regulatory approval of adjuvant treatment by the FDA and EMA. In KEYNOTE-716 postoperative pembrolizumab significantly improved 3-year RFS compared with

placebo (76,2% vs 63,4%; HR=0.62; 95% CI, 0.49-0.79) after a median follow-up of 39 months. Three-year distant metastasis free survival (DMFS) was also higher in the pembrolizumab group (84.4% vs 74.7%; HR=0.62; 95% CI 0.49-0.79). The median RFS or DMFS was not reached in either treatment group [47]. In CheckMate 76K nivolumab demonstrated a significant RFS benefit over placebo. Initial results showed 1-year RFS rates of 89% versus 79% [48], and updated analyses confirmed sustained benefit with 3-year RFS of 71% versus 61% respectively [49].

According to the Swedish national guidelines, adjuvant treatment with a PD-1 inhibitor is recommended for patients with stage IIIC-D melanoma and should be considered following radical surgery of stage IV disease. In contrast, adjuvant therapy for stage IIC and IIIB melanoma is not recommended outside of clinical trials due to immature data and unclear treatment benefit. However, patients with stage IIC and IIIB may be offered treatment within the Grand SLAM trial (ClinicalTrials.gov Identifier: NCT06488482) [31].

Targeted therapy

Adjuvant treatment with the BRAF inhibitor dabrafenib in combination with the MEK inhibitor trametinib in patients with BRAF V600 mutations was investigated in the phase III trial COMBI-AD and compared with placebo. Patients with stage III (AJCC 7) CMM were included. Five-year RFS was improved with dabrafenib plus trametinib (52% vs 36% HR=0.51; 95% CI 0.42-0.61). Overall survival at five years was also higher in the combination group (65% vs 54% HR=0.55; 95% CI 0.44-0.70) [50]. However, in the final analysis, reported in late 2024, after nearly 10 years of follow-up, no significant improvement in OS was observed in the overall study population treated with dabrafenib plus trametinib. However, in subgroup analysis, an OS benefit (HR=0.75; CI, 0.58-0.96) was demonstrated in patients with BRAF V600E mutated tumors [51].

Radiotherapy

Adjuvant radiotherapy to the lymph node basin after lymphadenectomy reduces the risk of locoregional recurrence but does not improve OS [52]. According to the Swedish national guidelines, adjuvant radiotherapy may be considered for patients at a high risk of recurrence when adjuvant systemic treatment cannot be administered, or for patients with a high risk of relapse who have no surgical options in the event of recurrence [31].

Treatment of generalized disease

Treatment of generalized disease consists of immunotherapy, targeted treatment and chemotherapy. In certain situations, treatment with isolated hyperthermic perfusion or c-kit inhibitor, may be an option.

Immunotherapy

Ipilimumab was in 2011 the first CPI approved by the FDA, for treatment of unresectable stage III and stage IV CMM. This is further described in the *Cancer Immunology* section under the subheading *CTLA-4 inhibitors* (page 29).

In 2014 the FDA approved the PD-1 inhibitors nivolumab and pembrolizumab for the treatment of advanced CMM, making CMM the first cancer type for which PD-1 inhibitors received regulatory approval. The study designs and results that formed the basis for the approval are described in the *Cancer Immunology* section under the subheading *PD-1/PD-L1 inhibitors* (page 31). These agents may be used regardless of BRAF mutational status or tumor PD-L1 expression.

The efficacy of combined immunotherapy with ipilimumab and nivolumab was evaluated in the phase III CheckMate 067 trial in previously untreated patients with unresectable stage III or stage IV CMM. Patients were randomized to receive nivolumab alone, ipilimumab alone or the combination of nivolumab plus ipilimumab. Median progression free survival (PFS) was 11.5 months in the combination therapy group, compared with 2.9 months for ipilimumab alone and 6.9 months for nivolumab alone. Among patients with PD-L1-positive tumors, median PFS was similar in the nivolumab monotherapy and combination therapy groups. In contrast, in patients with PD-L1-negative tumors (<1%) PFS was significantly longer with combination therapy than with nivolumab alone. High-grade IRAEs were more frequent with combination treatment (55%), than with nivolumab (16%) or ipilimumab (27%) [53]. In the final analysis, melanoma specific survival at 10 years was 52% in the combination therapy group, 44% in the nivolumab group and 23% in the ipilimumab group. Overall survival was significantly improved with both combination therapy and nivolumab alone, compared with ipilimumab alone. However, no statistically significant difference was observed between combination therapy and nivolumab monotherapy [54].

Combined treatment with relatlimab and nivolumab is discussed in the *Cancer Immunology* section under the subheading *Other immune checkpoints* (page 33).

Targeted therapy

Vemurafenib – Cobimetinib

Vemurafenib was evaluated in the phase III BRIM-3 trial in previously untreated metastatic CMM patients with tumors harboring the BRAF V600E mutation. Patients were randomized to receive either vemurafenib or dacarbazine. Response rates were 48% with vemurafenib and 5% with dacarbazine. Crossover from dacarbazine to vemurafenib after disease progression was recommended [55]. The most common grade 3-4 adverse events associated with vemurafenib were cutaneous toxicities including cutaneous squamous-cell carcinoma, keratoacanthoma and rash [56]. Final analyses demonstrated sustained benefit, with significantly longer median OS, censored at crossover, in the vemurafenib group compared with the dacarbazine group (13.6 vs 9.7 months) [57].

The efficacy of combination therapy with vemurafenib and cobimetinib was evaluated in the phase III co-BRIM study, in which patients received either cobimetinib or placebo in combination with vemurafenib. Extended follow-up at five years demonstrated a significantly improved median OS of 22.5 months (95% CI, 20.3-28.8) with combination therapy and 17.4 (95% CI, 15.0-19.8) months with vemurafenib monotherapy [58].

Vemurafenib is approved both as monotherapy and in combination with cobimetinib.

Dabrafenib – Trametinib

Trametinib was investigated in the phase III METRIC trial in patients with advanced melanoma. Treatment with trametinib was compared with the chemotherapy dacarbazine, with crossover to trametinib permitted upon disease progression. Median PFS was significantly longer in the trametinib arm than in the dacarbazine arm (4.8 vs 1.5 months). Reported adverse events included peripheral edema, rash and diarrhea, as well as reduced cardiac ejection fraction and ocular toxicity [59]. Dabrafenib was evaluated in the phase III BREAK-3 trial, in which patients were randomized to receive dabrafenib or dacarbazine. Median PFS was significantly improved with dabrafenib compared with dacarbazine (5.1 vs 2.7 months). The most frequently reported adverse events were cutaneous toxicity, fever, arthralgia, headache and fatigue [60].

The combination of dabrafenib plus trametinib was evaluated in the phase III COMBI-v trial, in which patients received either dabrafenib plus trametinib or vemurafenib as first-line treatment. Both median PFS and 1-year OS were significantly improved with dabrafenib plus trametinib (11.4 vs 7.3 months and 12 % vs 65%, respectively) [61]. Treatment with dabrafenib plus trametinib was also assessed in the phase III COMBI-d trial, where patients were randomized to receive dabrafenib plus trametinib or dabrafenib plus placebo.

Three-year PFS was 22% with the combination therapy and 12% for dabrafenib monotherapy, and 3-year OS was 44% versus 32%, respectively [62].

Dabrafenib and trametinib are approved for use as monotherapy or in combination and the latter is standard of care. In Sweden, MEK inhibitor monotherapy is not recommended.

Encorafenib-binimetinib

The efficacy of encorafenib plus binimetinib treatment was evaluated in the phase III COLUMBUS trial. Patients with unresectable stage III or stage IV CMM, either previously untreated or with disease progression following first-line immunotherapy, were randomized to receive encorafenib plus vemurafenib, encorafenib monotherapy, or vemurafenib monotherapy. At the 7-year follow-up, PFS was significantly longer with encorafenib plus binimetinib compared with both monotherapy arms. The 7-year PFS rats were 21.2 % for encorafenib plus binimetinib, 15.8 % for encorafenib monotherapy, and 6.4 % for vemurafenib. Overall survival at 7-years was significantly improved with encorafenib plus binimetinib compared with vemurafenib (27.4% vs 18.2%), while no significant difference was observed compared with encorafenib monotherapy (27.4 vs 31.7%) [63]. Encorafenib and binimetinib are approved for use only in combination.

Chemotherapy

Chemotherapy is currently used as a second- or third-line treatment in CMM patients. No studies have demonstrated an OS benefit with chemotherapy. Dacarbazine was previously the standard chemotherapeutic agent for melanoma and is administered intravenously. Temozolomide is an oral analogue of dacarbazine that also penetrates the blood-brain barrier. Both agents have low response rates of 10-20% [64, 65]. Their efficacy has been directly compared in a phase III trial, which showed a small but statistically significant improvement in PFS with temozolomide (1,9 months) compared to dacarbazine (1,5 months), while there was no significant difference in OS [66]. Additionally, quality of life assessments favoured temozolomide over dacarbazine [66, 67]. Accordingly, the Swedish national guidelines recommend temozolomide as the preferred chemotherapeutic option [31].

Additional treatment options

For patients with at least one injectable metastasis and no evidence of visceral organ involvement, treatment with talimogene laherparepvec (T-VEC), an oncolytic virus encoding granulocyte colony stimulating factor (GM-CSF), is

approved. The approval is based on a phase III trial demonstrating significantly higher rates of durable responses in patients treated with T-VEC intratumorally compared with those receiving the cytokine GM-CSF subcutaneously [68]. Another therapeutic option for cutaneous metastases is electrochemotherapy, which involves intratumoral administration of bleomycin or cisplatin, followed by application of electric pulses [69]. In patients with intransit or satellite metastases confined to the limbs, isolated limb perfusion, most commonly using melphalan, may be considered [70]. Additionally, stereotactic radiosurgery can be an effective treatment option in patients with brain oligometastases [71].

Cancer immunology

Background

Throughout history, there have been reports of spontaneous cancer regression, with one of the theories attributing this phenomenon to immunological factors [72]. In 1890 William B. Coley, now recognized as the "Father of Cancer Immunotherapy", pioneered a groundbreaking treatment by inducing erysipelas using a cocktail of heat-killed bacteria known as "Coley's Toxins", to treat osteosarcoma patients. Coley reported remarkable success and published many reports of tumor regression following treatment with his toxins [73]. This can be considered the start of cancer immunotherapy, a revolutionary field that harnesses the body's own immune system to recognize and eliminate cancer cells. Coley's work laid the foundation for modern immunotherapeutic approaches, including CPIs, other monoclonal antibodies and cancer vaccines, which have since transformed oncology and significantly improved patient outcomes.

Immunosurveillance and immune escape

The concept of cancer immunosurveillance, introduced in the 1950s by Burnet and Thomas, describes the immune system´s ability to detect and destroy malignantly transformed cells [20]. Building on this idea, Duann et al. expanded the framework in 2002 with the concept of cancer immunoediting, which highlights the dynamic interplay between the immune system and tumor cells. Cancer immunoediting involves three distinct phases: elimination, equilibrium, and escape. In the elimination phase, if successful, the immune system detects and eradicates developing tumors, halting the process. If some tumor cells survive elimination, they enter the equilibrium phase, where they coexist in a dynamic balance with the immune system, a state that can persist for years. Finally, in the escape phase, cancer cells evolve mechanisms to evade immune detection and destruction, leading to uncontrolled proliferation and the development of clinically detectable cancer [20].

In 2000 Hanahan and Weinberg launched six hallmarks describing the transformation of normal cells into cancer cells. These are: maintain growth

signals, insensitivity to antigrowth signals, evading apoptosis, limitless replicative potential, sustained angiogenesis, tissue invasion and metastasis [74]. The ability to evade immune destruction was recognized as an emerging hallmark of cancer in 2011, marking a significant advancement in our understanding of tumor biology. This concept underscores how cancer cells develop sophisticated mechanisms to avoid detection and elimination by the immune system, enabling them to survive and proliferate despite the body's natural defenses [75]. In the same update, deregulating cellular metabolism was also introduced as an emerging hallmark, while tumor promoting inflammation as well as genome instability were described as enabling mechanisms [75]. Since then, the hallmarks have been updated twice, and over time, the framework has evolved from a simple list of tumor traits to a complex systems model describing cancer as an evolving ecosystem involving genetic, epigenetic, immune, microbial and environmental interactions [76, 77].

The cancer immunity cycle

A widely accepted model describes the mechanisms of interaction between the immune system and developing cancer. Known as the cancer immunity cycle, this process consists of seven sequential events that enable the immune system to recognize and eliminate cancer cells. The process starts when the immune system detects antigens from dead cancer cells. Neoantigens generated by oncogenesis are released and captured by dendritic cells (DCs) for processing, a step that requires additional immune-stimulating signals to overcome peripheral tolerance to tumor antigens (step 1). These signals may include proinflammatory cytokines and factors released by the gut microbiota or dying tumor cells. Once processed, DCs present tumor antigens to T cells via major histocompatibility complex (MHC) I or MHC II molecules (step 2). In the third step effector T cells in the lymph node are primed and activated against the tumor-specific antigen. At this stage, the balance between effector T cells and regulatory T cells determines the nature of the immune response. Following activation, effector T cells migrate toward the tumor site (step 4) and infiltrate the tumor bed (step 5). Once inside, they recognize cancer cells by binding their T-cell receptor (TCR) to tumor antigens presented on MHC I molecules (step 6), ultimately leading to killing of the tumor cell (step 7). This final stage in the cycle not only eliminate cancer cells but also triggers the release of additional tumor-associated antigens, amplifying the immune response in subsequent cycles [78].

An updated version of the cancer-immunity cycle was published in 2023 [79]. While the fundamental structure of the cycle remains intact, several

important new concepts were introduced. Among these is the classification of a tumor's immunological phenotype, or "immunotype," which includes three distinct categories: immune-inflamed, immune-excluded, and immune-desert. The update also introduces a microenvironmental cancer-immunity subcycle, positioned between step 5 (T cell infiltration into the tumor bed) and step 6 (binding of the T cell receptor to tumor antigens presented on MHC class I molecules). This subcycle consists of three sequential phases: first, activated T cells accumulate within and migrate through the tumor stroma; second, they interact with intratumoral immune cells; and third, they sustain their effector functions within the TME. However, various factors within the TME, such as inhibitory immune cells, stromal barriers, metabolic disturbances, and T cell dysfunction, can interfere with this process and ultimately disrupt the progression of the cancer-immunity cycle [79].

However, in patients with cancer, this cycle is often disrupted. There are several possible reasons for defective immune response. Cancer antigens may go undetected or be misclassified as "self" prompting a regulatory T-cell response rather than an effector T-cell attack. Additionally, effector T cells may struggle to infiltrate tumors, fail to home to the tumor site effectively, or encounter a TME that actively suppresses immune activity. These immune evasion strategies allow cancer cells to persist and proliferate despite the presence of an immune response, highlighting the complexity of immune dysfunction in cancer progression [78].

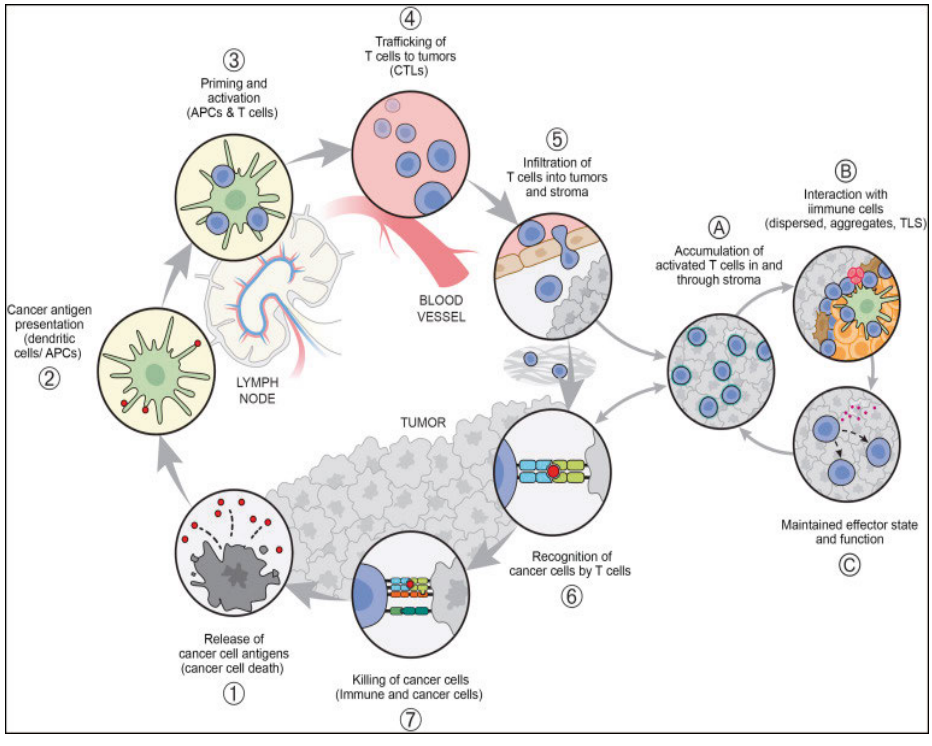

Figure 3. The cancer immunity cycle and the tumor microenvironment cancer-immunity subcycle with its sequential steps illustrated. Abbreviations: APCs = antigen presenting cells, CTLs = cytotoxic T lymphocytes, TLS = tertiary lymph node structure. Reprinted from the article "The cancer-immunity cycle: Indication, genotype, and immunotype" 2023, Vol.56/Issue 10, p.2188-225, DOI: 10.1016/j.immuni.2023.09.011, licensed under CC BY-NC-ND

Cancer immunotherapy

Cancer immunotherapy is an established therapeutic approach that complements existing treatments such as chemotherapy, surgery, radiotherapy and targeted therapy. This therapy works by activating the patient's own immune system to generate a specific and precise response, ultimately leading to the elimination of cancer cells. There are various forms of immunotherapy, including monoclonal antibodies (mAbs), such as bispecific mAbs and immune CPIs, small molecule immunotherapies, adoptive cell therapies (e.g., chimeric antigen receptor T cells [CAR-T], tumor infiltrating lymphocytes [TIL] and CAR-NK cells), oncolytic viruses and cancer vaccines [80]. In recognition of their groundbreaking work in cancer treatment using immune CPIs, James P. Allison and Tasuku Honjo were awarded the 2018 Nobel Prize in Physiology or Medicine.

Checkpoint inhibitors

As previously described, the cancer-immunity cycle provides a framework for understanding T cell involvement in the anti-tumor immune response, encompassing priming and activation in lymph nodes, trafficking into the TME, and ultimately, tumor elimination. Antigen-presenting cells (APCs) play a crucial role by identifying and presenting tumor neoantigens to naïve T cells. Effective T cell activation requires two key signals: first, the TCR must bind to the MHC displaying the antigen on APCs; second, the co-stimulatory receptor CD28 on the T cell must engage with the B7 ligand on APCs. Upon activation, T cells proliferate, enhance their effector functions, and generate memory T cells. At the same time, inhibitory checkpoint molecules are upregulated to regulate T cell activity, preventing excessive immune responses and reducing the risk of autoimmunity. Many different immune checkpoints have been found, cytotoxic T lymphocyte-associated protein 4 (CTLA-4) and programmed death 1 (PD-1) are the two most studied T-cell inhibitory molecules [81].

CTLA-4 inhibition, mechanism of action

CTLA-4 is expressed on activated T cells and regulatory T cells (Tregs). As a homolog of CD28, it binds to B7 ligands, leading to the inhibition of T cell proliferation and interleukin 2 (IL-2) production. CTLA-4 binds to B7 ligands with greater affinity and avidity than CD28, thus enabling to outcompete CD28 ligation. Blocking CTLA-4 with an anti-CTLA-4 antibody disrupts this inhibitory checkpoint, thereby enhancing T cell-mediated anti-tumor responses as demonstrated in Figure 4. CTLA-4 is believed to suppress T cell activation at an early stage of the immune response in the lymph node, while PD-1 primarily regulates T cell activity at later stages within peripheral tissues [81].

CTLA-4 inhibitors

As earlier mentioned, in 2011, the FDA approved the first CPI, ipilimumab, a monoclonal antibody targeting CTLA-4, for the treatment of CMM after demonstration of improved OS in patients with unresectable stage III or IV disease in second line. No previous treatment for melanoma had shown improved OS before. In the pivotal clinical trial, almost 700 patients were included and randomly assigned, in a 3:1:1 ratio, to receive ipilimumab (3 mg/kg) plus a glycoprotein 100 (gp 100) peptide vaccine, ipilimumab (3 mg/kg) alone or gp 100 alone. The median OS was 10.0 months among patients receiving ipilimumab plus gp 100 and 10.1 months for patients receiving ipilimumab alone which was significantly longer compared to 6.4 months for those receiving only gp 100. In total, 10-15% of the patients developed grade 3 or 4 IRAEs and 14 patients died related to the study drug (2.1%), 7 deaths were immune-related [82].

In another phase III study, ipilimumab was evaluated as a first-line treatment for patients with metastatic CMM. Approximately 500 patients were randomized in a 1:1 ratio to receive either ipilimumab (10 mg/kg) plus dacarbazine or dacarbazine plus placebo. The median OS was 11.2 months in the ipilimumab plus dacarbazine group, compared to 9.1 months in the dacarbazine plus placebo group. Long term survival rates were also significantly higher in the ipilimumab-dacarbazine group compared to the dacarbazine-placebo group (1 year: 47.3% vs. 36.3%, 2 years: 28.5% vs. 17.9%, 3 years: 20.8% vs. 12.2%). Grade 3 or 4 adverse events occurred in 56.3% of patients receiving ipilimumab-dacarbazine, as compared to 27.5% in the dacarbazine-placebo group. Notably, no drug-related deaths were reported in the ipilimumab-dacarbazine group [83]. Additionally, a pooled analysis of multiple phase II and III trials of ipilimumab in stage III or IV CMM showed long-term survival for up to 10 years in some patients [84].

Nowadays, ipilimumab is primarily used in combination with PD-1/PD-L1 inhibitors and is approved for treatment of various cancers including CMM, renal cell carcinoma (RCC), non-small cell lung cancer (NSCLC), malignant mesothelioma, specific subtypes of colorectal cancer and esophageal cancer. More recently, another CTLA-4 inhibitor, tremelimumab, has become available for the treatment of unresectable hepatocellular carcinoma (HCC) and NSCLC. It is used in combination with durvalumab, a PD-L1 inhibitor, for hepatocellular carcinoma and with the addition of platinum-based chemotherapy for NSCLC [85, 86].

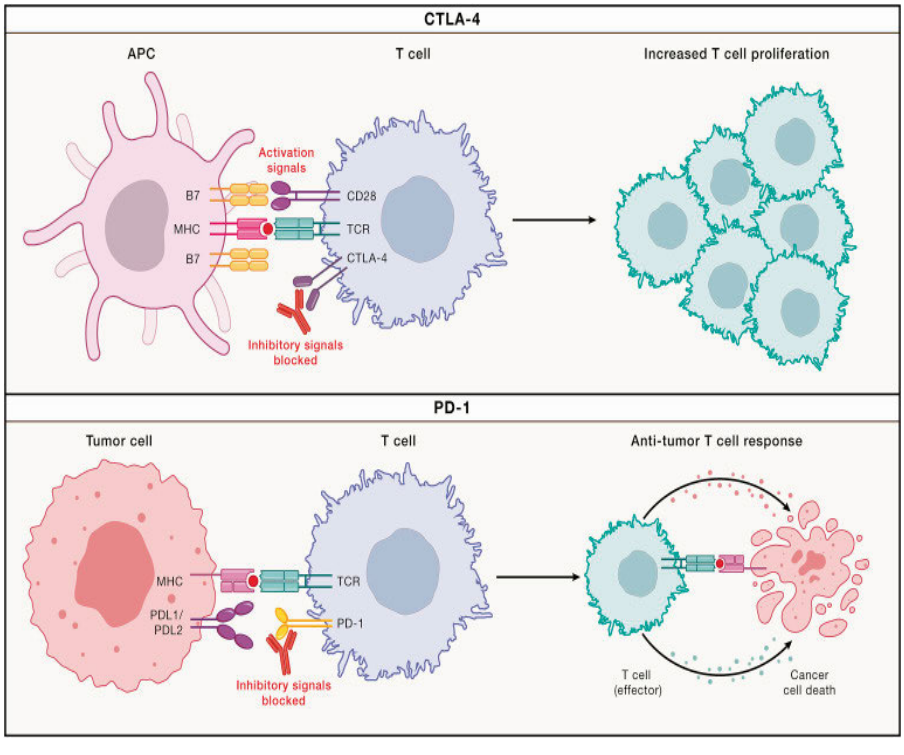

Figure 4. Schematic demonstrating the difference in mechanisms of action of CTLA-4 and PD-1. CTLA-4 modifies APC-induced T cell responses by competing with CD28 for B7 binding, thereby limiting the uncontrolled expansion of activated T cells. PD-1 engagement with PD-L1 inhibits TCR signaling, regulating the effector phase of T cell responses. Blocking CTLA-4 or PD-1 using anti-CTLA-4 or anti-PD-1 antibodies disrupts these inhibitory signals in T cells, enhancing their ability to eliminate tumor cells. Abbreviations: APC, antigen-presenting cell; CTLA-4, cytotoxic T lymphocyte-associated protein 4; MHC, major histocompatibility complex; PD-1, programmed death 1; PD-L1, programmed death ligand 1; TCR, T cell receptor. Reprinted from the article "Immune checkpoint theory – current perspectives and future directions" 2023, Vol 186/Issue 8, p. 1652-1669, with permission from Elsevier [81].

PD-1/PD-L1 inhibition, mechanism of action

PD-1 is expressed on the surface of various immune cells, including CD4+ and CD8+ T-cells, Tregs, B cells and natural killer (NK) cells. It interacts with two ligands, PD-L1 and PD-L2, with PD-L2 exhibiting a higher affinity for PD-1 but having a more restricted expression compared to PD-L1 [87]. PD-L1 is broadly expressed on various hematopoietic cells (such as T cells, B cells, macrophages, DCs) as well as non-hematopoietic cells (including epithelial, stromal and endothelial cells). In contrast, PD-L2 is predominantly found on hematopoietic cells (such as Th2 cells, B cells, DCs, macrophages), but is also expressed on certain epithelial cells, particularly in the lungs. Apart from binding to PD-1, PD-L1 also inhibits T-cell responses by binding to B7-1/CD80 [87]. PD-L1 is often overexpressed on tumor cells, where its interaction with PD-1 on T-cells suppresses antigen specific T-cell immune responses, enabling tumors to evade immune detection, as demonstrated in Figure 4 [81]. The expression of PD-L1 is regulated by various factors including cytokines, interferons (IFNs), vascular endothelial growth factor (VEGF) and tumor necrosis factor (TNF), which can enhance PD-L1 expression and contribute to immune escape mechanisms in cancer [87].

PD-1/PD-L1 inhibitors

In 2014, the FDA approved the first PD-1 inhibitors, nivolumab and pembrolizumab, for the treatment of advanced CMM. Pembrolizumab, a humanized IgG4 immune CPI antibody, was evaluated in the phase II study Keynote 002. The study included over 500 patients with metastatic CMM who had progressed during treatment with ipilimumab, and if presence of BRAF-mutation, had been treated with a BRAF or MEK inhibitor or both. Patients were randomized in a 1:1:1 ratio to treatment with pembrolizumab 2 mg/kg, pembrolizumab 10 mg/kg, every 3 weeks, or investigator's choice of chemotherapy. The study demonstrated a significant improvement in both PFS, and overall response rate, for patients receiving pembrolizumab, compared to those treated with chemotherapy [88]. In the phase III Keynote-006 study, over 800 patients with metastatic CMM were enrolled. Patients could have received one previous systemic treatment, provided it was not an anti-CTLA-4 or anti-PD-1/L1 agent. They were randomly assigned to receive pembrolizumab 10 mg/kg every 2 weeks or every 3 weeks, or four doses of ipilimumab. In the final study report, the median OS in the ipilimumab group was 16 months, while in the pembrolizumab groups, the median OS had not yet been reached (HR 0.68 95% CI: 0.53-0.87 for pembrolizumab every 2 weeks vs ipilimumab p=0.0009 and 0.68 0.53-0.86 for pembrolizumab every 3 weeks vs ipilimumab p=0.0008). The median follow-up time was 23

months, and the two-year OS was significantly improved with 55% in both pembrolizumab groups compared to 43% in the ipilimumab group. Additionally, pembrolizumab was associated with a lower frequency of serious treatment related adverse events [89]. Long-term follow-up data (Keynote 587) demonstrate median OS of 32.7 months (95% CI: 24.5-41.6) for pembrolizumab and 15.9 months (95% CI: 13.3-22.0) for ipilimumab (HR 0.71 95% CI 0.6-0.85) and an OS rate of 34% in the combined pembrolizumab arm and 24% in the ipilimumab arm, at 10 years [90].

Nivolumab, a human IgG4 PD-1 immune CPI antibody, was evaluated in two different phase III trials, and the results from these studies were of great importance for the drug's approval. In CheckMate 066, more than 400 previously untreated patients with metastatic CMM and BRAF wild-type status, were randomly assigned to receive eighter nivolumab (3 mg/kg every 3 weeks) or dacarbazine. The median PFS was significantly improved with 5.1 months in the nivolumab group compared to 2.2 months in the dacarbazine group, while the overall response rate (ORR) also was significantly enhanced with 40% versus 14% respectively. At one year, the OS rate was 73% (95% CI: 65.5-78.9) in the nivolumab group, compared to 42% (95% CI: 33.0-50.9) in the dacarbazine group (HR 0.42: 99.79% CI: 0.25-0.73; $p<0.001$) [91]. In the three-year follow-up, published in 2019, the survival rates at tree years, were 51% for patients treated with nivolumab and 22% for those treated with dacarbazine, with a median OS of 37.5 months (95% CI: 25.5 months - not reached) in the nivolumab group and 11.2 months (95% CI: 9.6-13) in the dacarbazine group (HR 0.46; 95% CI, 0.36-0.59; $P< .001$). Complete responses were reported in 19% of patients, and partial responses in 24% in the nivolumab group, compared to 1.4% and 13% respectively, in the dacarbazine group [92]. In the phase III trial CheckMate 037, the safety and efficacy of nivolumab were compared with the investigator's choice of chemotherapy (either dacarbazine or paclitaxel combined with carboplatin), in patients with advanced CMM, after progression on treatment with ipilimumab and, if BRAF mutant, a BRAF inhibitor. In total, over 400 patients were enrolled. An interim analysis, published in 2015 reported an ORR of 32% in the nivolumab arm compared to 10% in the chemotherapy arm [93]. Updated results from the cohort, published in 2018, reveal an ORR of 27% vs 10% and a median duration of response of 32 months versus 13 months in the nivolumab and chemotherapy arms, respectively. No significant differences in OS were observed between the groups. The authors suggested that this could be due to a higher proportion of patients with poor prognostic factors in the nivolumab group and a considerable proportion of patients in the chemotherapy arm receiving PD-1 inhibitors as crossover therapy [94].

In 2015 both pembrolizumab and nivolumab were approved for the treatment of NSCLC. In the same year, the FDA also granted approval for nivolumab as a second-line treatment for metastatic RCC, following antiangiogenetic therapy [95]. Since then, the FDA has approved ten monoclonal antibodies targeting PD-1 (cemiplimab, toripalimab, nivolumab, pembrolizumab, tislelizumab and dostarlimab) or PD-L1 (atezolizumab, avelumab, cosibelimab and durvalumab) for treatment of different malignancies in various therapeutic settings [96]. The spectrum of cancers for which PD-1/PD-L1 inhibitors have been approved has expanded significantly and now includes genitourinary tract cancers, such as bladder and kidney cancer, gastrointestinal cancers like biliary tract cancer, colorectal cancer, esophageal cancer, gastric cancer and liver cancer as well as breast cancer, gynecological cancers such as cervical cancer and endometrial carcinoma, head-and neck cancer, lymphoma, lung cancer, and various skin cancers, i.e. CMM, squamous cell carcinoma, basal cell carcinoma and Merkel cell carcinoma. Additionally, these immune CPIs have been incorporated into treatment approaches for certain sarcomas, further expanding their role in modern oncology. The drugs also target biomarker-based indications and are approved for the treatment of microsatellite instability high (MSI-H) or defect mismatch repair (dMMR) tumors.

Other immune checkpoints

Lymphocyte activation gene-3 (LAG-3) is a cell-surface molecule expressed on various immune cells, including T cells, where it plays a regulatory role by suppressing T-cell proliferation and effector function. In multiple tumor types, including CMM, LAG-3 is often upregulated. In 2022, the FDA approved relatlimab in combination with nivolumab for the treatment of patients with advanced CMM. Relatlimab is a first-in class human IgG4 LAG-3 blocking antibody. The approval was based on the phase II/III RELATIVITY-047 study, which enrolled over 700 patients with previously untreated, unresectable CMM. Participants were randomized to receive either relatlimab plus nivolumab or nivolumab alone. The study demonstrated a significant improvement in median PFS, with the relatlimab-nivolumab combination achieving 10.1 months compared to 4.6 months in the nivolumab monotherapy arm, at a median follow-up time of 13 months. In patients with PD-L1 expression of less than 1%, the median PFS was 6.4 months, as compared to 2.9 months in the relatlimab-nivolumab versus nivolumab arm. For patients with PD-L1 expression of 1% and above, PFS was similar in both groups. Based on these results, the EMA approved relatlimab-nivolumab for patients with PD-L1 expression below 1%. Sweden has adopted the same subgroup

approval criterium. Grade 3 or 4 adverse events were reported to be 18.9% versus 9.7% in the relatlimab-nivolumab versus nivolumab group [97].

Potential drugs targeting several other immune checkpoints are currently under investigation, including T cell immunoglobin and mucin-domain containing-3 (TIM-3), T cell immunoglobin and immunoreceptor-tyrosine-based-inhibitory-motif domain (TIGIT), V-domain Ig suppressor of T cell activation (VISTA) [81].

Toxicity and efficacy of checkpoint inhibitors

Immune CPIs may cause IRAEs, which can affect nearly any organ in the body (as illustrated in Figure 5) and can arise at any time during the treatment or after its discontinuation [98]. These toxicities result from dysregulated immune activation causing inflammation in various tissues. Mechanisms underlying IRAEs include enhanced T-cell activity against host tissues, T-cell cross-reactivity due to antigens shared by both tumors and healthy tissue, activation of B-cells resulting in production of autoantibodies and increased levels of preexisting autoantibodies, and elevated pro-inflammatory cytokine levels. These processes reflect a breakdown of immune homeostasis induced by CPI [99].

The development of IRAEs indicates activation of the immune system [98, 99]. A positive correlation between the occurrence of IRAEs and treatment response has been observed across multiple tumor types, including CMM, NSCLC, urothelial cancer, RCC, head and neck cancers and gastrointestinal malignancies [100-104]. The correlation between development of multiple IRAEs and treatment efficacy is less studied and more extensively described in Paper II.

Incidence of IRAEs due to type of CPI

The patterns and severity of IRAEs vary among different CPIs, with CTLA-4 inhibitors generally associated with higher frequency of IRAEs compared to PD-1/L1- inhibitors. Most IRAEs are mild and reversible with appropriate treatment, but some can be long-lasting and, in rare cases, life-threatening [105, 106]. Toxicity-related fatality rates have been reported as being 0.36% for PD-1 inhibitors, 0.38% for PD-L1 inhibitor, 1.08% for CTLA-4 inhibitors and 1.23% for PD-1/PD-L1 plus CTLA-4 inhibitor, in a meta-analysis of 112 trials involving over 19 000 patients [106].

A systematic review and meta-analysis reported an overall incidence of IRAEs of 72% in patients treated with anti-CTLA-4 antibodies, with high grade (≥grade 3) IRAEs occurring in 24% of cases [107]. In the CheckMate 067 study, treatment-related grade 3 or 4 adverse events were observed in 16%

of patients receiving nivolumab (3 mg/kg) alone, 55% of those treated with nivolumab (1 mg/kg) in combination with ipilimumab (3 mg/kg), and 27% of patients receiving ipilimumab (3 mg/kg) alone [53]. These findings demonstrate that IRAEs are more frequent and severe when CTLA-4 inhibition is used, particularly in combination therapies.

Large meta-analyses of clinical trials involving patients with various cancers, treated with PD-1/PD-L1 inhibitors, report the incidence of IRAEs of any grade ranging from 17% to 27%, with grade ≥ 3 IRAEs occurring in 4%-6% of patients [108-110]. However, another meta-analysis, which also included patients from clinical trials, reported a higher frequency of IRAEs, with any grade AEs occurring in 66% of patients, and AEs grade ≥ 3 in 14% [111]. This discrepancy may be due to differences in the classification of adverse events, with focus on treatment related adverse events in general rather than specifically on IRAEs. Moreover, real-world studies have reported an even higher incidence of IRAEs. In a large study of over 3000 lung cancer patients, the cumulative incidence of any-grade IRAEs at 12 months was 52.2% [112], while a smaller cohort of almost 90 patients with various malignancies reported an overall incidence of 73% [113]. The discrepancy between real-world data and clinical trial cohorts may be due to broader inclusion criteria of in real-world settings, which encompass patients who might be excluded from clinical trials due to comorbidities or other factors. Additionally, differences in the detection and reporting of IRAEs between clinical trials and routine clinical practice could also contribute to the variation in reported frequencies.

No randomized head-to-head studies have directly compared the effects of PD-1 and PD-L1 inhibitors. However, several meta-analyses based on clinical trial data have evaluated the incidence of IRAEs between these two classes of inhibitors, showing a higher frequency of IRAEs with PD-1 inhibitors [109-111, 114]. This topic is further explored in Paper III.

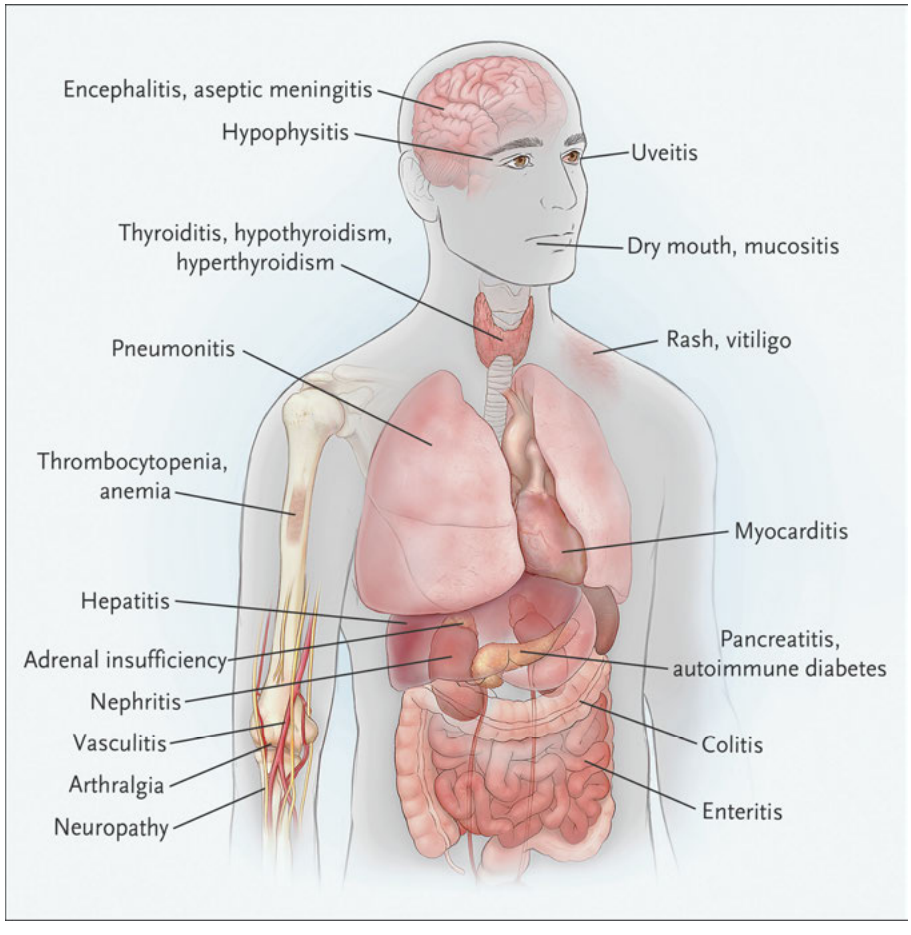

Figure 5. Organs affected by immune checkpoint blockade. The figure illustrates the most common immune-related adverse events. Reproduced with permission from Postow MA, Sidlow R, Hellmann MD. Immune-Related Adverse Events Associated with Immune Checkpoint Blockade. N Engl J Med. 2018;378:158–168. Copyright Massachusetts Medical Society.

Incidence of IRAEs in relation to dose

For patients treated with ipilimumab IRAEs appear to be dose dependent. In the CheckMate 511 study, patients with advanced CMM were randomized to receive either nivolumab 3 mg/kg plus ipilimumab 1 mg/kg or nivolumab 1 mg/kg + ipilimumab 3 mg/kg. The incidence of grade 3 to 5 adverse events was significantly lower in the group receiving nivolumab 3 mg/kg plus ipilimumab 1 mg/kg (34% 95% CI, 27.0% to 41.3%) compared to those receiving nivolumab 1mg/kg plus ipilimumab 3 mg/kg (48% 95% CI, 40.8% to 55.9%). Despite this difference in toxicity, PFS, ORR and OS were similar between

the groups [115, 116]. A similar result was observed in a study evaluating pembrolizumab with two different doses of ipilimumab, where a lower ipilimumab dose was associated with reduced grade 3-5 adverse events without compromising efficacy [117].

The development of IRAEs has not been shown to be dose-dependent for PD-1/PD-L1 inhibitors [108].

Organ specific IRAEs in relation to type of CPI

Mice deficient in the CTLA-4 gene experience fatal lymphoproliferation, whereas PD-1 knockout mice exhibit a more limited and variable spectrum of autoimmune manifestations, including arthritis and cardiomyopathy [98]. Similarly, patients treated with CTLA-4 inhibitors tend to develop IRAEs that differ in organ involvement from those seen with PD-1 or PD-L1 inhibitors.

Gastrointestinal toxicities and hepatitis are generally more common with CTLA-4 inhibition, while PD-1/PD-L1 inhibitors are associated with a lower frequency of these events [118, 119]. Among endocrine toxicities, hypophysitis and thyroid dysfunction are the most frequently reported. Hypophysitis occurs more often with CTLA-4 inhibitors, potentially due to direct targeting of pituitary cells that express CTLA-4, while thyroid dysfunction is more frequently observed with PD-1/PD-L1 inhibitors [118, 119]. Dermatologic toxicity is the most common IRAE across CPIs and symptoms are often described as rash, pruritus or vitiligo. Rash and pruritus are more commonly associated with CTLA-4 inhibitors, whereas vitiligo is observed more frequently in patients treated with PD-1/PD-L1 inhibitors [118]. Pulmonary toxicity also differs between these drug classes. PD-1/L1 inhibitors are linked to a higher incidence and severity of pneumonitis, whereas pneumonitis is less commonly reported with CTLA-4 inhibitors [118, 119]. Neurologic IRAEs can be classified as either inflammatory (encephalitis, myelitis, vasculitis and meningitis) or peripheral neuromuscular autoimmune disorders (such as myasthenia gravis and Guillian-Barré syndrome). Myasthenia gravis, non-infectious encephalitis and myelitis appear more frequently with PD-1/PD-L1 inhibitors, while Guillian-Barré syndrome and non-infectious meningitis have been more commonly associated with CTLA-4 inhibitor therapy [119]. Arthralgia and myalgia are reported more frequently in patients receiving PD-1/PD-L1 inhibitors compared to those treated with CTLA-4 inhibitors [118]. Additionally, ocular toxicities, while less common overall, tend to be more frequent and severe with CTLA-4 inhibitors than with PD-1/PD-L1 inhibitors [119].

A systematic review and meta-analysis reported a total of 613 fatal events associated with CPI therapy. The cause of fatality varied depending on the

type of treatment. For CTLA-4-inhibitors, most deaths (70%) were attributed to colitis. In contrast, fatalities related to PD-1/PD-L1-inhibitors were most commonly due to pneumonitis (35%), hepatitis (22%), neurologic toxicity (15%) and myocarditis (8%). Among patients receiving combined CTLA-4 and PD-1 inhibition, the leading causes of death were colitis (37%) and myocarditis (25%). Notably, myocarditis carried the highest fatality rate. Fatal IRAEs typically occurred early during treatment. The median time to death was 35 days after initiation of combined therapy and 43 days for patients receiving PD-1-inhibition or ipilimumab [106].

Organ specific IRAEs in relation to tumor type

Comparative studies of IRAEs across different tumor types have revealed distinct patterns linked to cancer type [118, 120, 121]. A systematic review of 48 trials involving nearly 7000 patients with various cancers treated with CPIs identified tumor-specific IRAE profiles. Melanoma patients had a higher incidence of gastrointestinal and dermatologic IRAEs, but a lower incidence of pneumonitis, compared to those with NSCLC. Additionally, CMM patients experienced more frequent occurrences of arthritis and hypothyroidism than those with RCC, whereas RCC patients were more prone to develop pulmonary IRAEs [118]. These findings are supported by another study showing a similar association between skin IRAEs and CMM, as well as respiratory IRAEs and NSCLC [120]. In a retrospective analysis of over 14 000 patients, Kalinich et al. reported that patients with CMM (OR: 0.70, p=0.01), RCC (OR: 0.71, p=0.03) and other cancers (OR: 0.50, p<0.001) had significantly lower odds of developing IRAEs requiring hospitalization compared to patients with NSCLC [121].

In one study investigating IRAE patterns, a strong association was observed between thymic cancer and the development of myotoxicities. Compared to patients without thymic cancer, those with thymic malignancies had significantly higher odds of developing myocarditis (OR=32; 95%CI; 19-51), myositis (OR=28 95% CI; 16-45), and myasthenia-gravis (OR=32 95%CI; 31-81) [122].

Furthermore, certain types of IRAEs have been shown to correlate more strongly with treatment efficacy, and these associations varies by cancer type. For instance, the development of vitiligo is associated with improved treatment response in CMM patients, while thyroid dysfunction has been linked to better outcomes in patients with NSCLC and RCC [123-127].

Proposed mechanisms underlying cancer-type variation in IRAEs include differences in the TME, immune infiltrate composition, adaptive immune

responses, neo-antigen burden and the presence of shared antigens between tumors and normal tissues. In addition, variations in comorbidities and prior treatments may influence the risk of IRAEs. For example, chronic obstructive pulmonary disease and prior lung irradiation may contribute to a higher incidence of pneumonitis in patients with NSCLC [99, 118].

Timeline of IRAE onset

The timing of onset and resolution of IRAEs vary depending on the affected organ system and the type of CPI administered [128]. A pooled analysis of 23 clinical trials involving nearly 8500 patients with various cancer types investigated the pooled median time to onset (PMT-O) and resolution of IRAEs. The PMT-O for all-grade IRAEs ranged from a median of 2.2 to 14.8 weeks. The earliest-onset IRAEs included dermatologic, gastrointestinal, neurologic and hypersensitivity/infusion reactions, while renal IRAEs had the longest time to onset. Severe IRAEs (grade ≥ 3) tended to occur later than low-grade events. For example, with PD-1/PD-L1 inhibitors, the median PMT-O for grade ≥ 3 was significantly longer than for all-grade IRAEs (27.5 weeks vs. 8.4 weeks, $p<0.001$). Similarly, with combination treatment using ipilimumab and nivolumab, severe IRAEs appeared later compared to all-grade events (7.9 weeks vs. 6 weeks $p< 0.0001$). Overall, IRAEs emerged earlier with ipilimumab/nivolumab combination therapy than with nivolumab monotherapy [128]. These patterns are visually summarized in Figure 6. Similar findings were reported in a cohort of 15551 CMM patients, where IRAEs occurred earlier with combination therapy than with monotherapy [129].

Further insight into IRAE timing was provided by Gougis et al., who analyzed data from VigiBase, the international pharmacovigilance database. They classified IRAEs into 25 distinctive types based on reports from patients treated with CTLA-4 inhibitor, PD-1 or PD-L1 inhibitor, combination therapy with CTLA-4 and PD-1/L1 inhibitor or anti-Lag 3 agents. Median time-to onset (TTO) ranged from 1 to 9 months. The shortest TTOs were observed for myotoxicities, with median onset times of 31 days for myositis, 33 days for myocarditis, and 31 days for myasthenia gravis. Severe cutaneous adverse reaction also had early onset (median 40 days). Conversely, more delayed IRAEs included arthritis (104 days), diabetes (114 days), pancreatitis (121 days), esogastritis (126 days), sarcoidosis (141 days) and vitiligo (179 days). Bullous autoimmune skin reactions showed the most delayed onset, with a median TTO of 273 days [122].

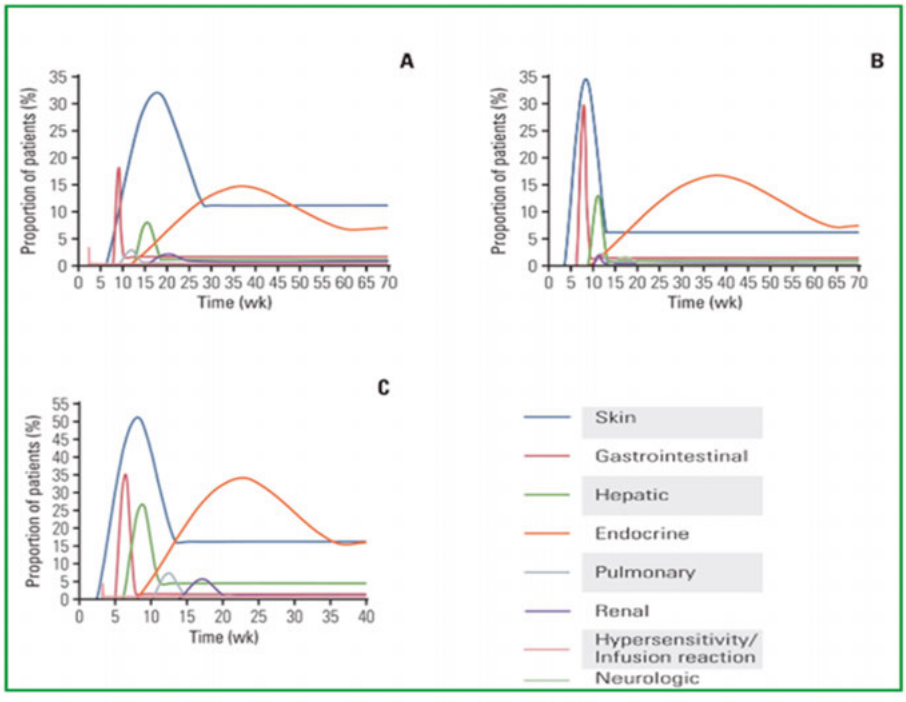

Figure 6. Kinetics of the onset and resolution of all-grade IRAEs caused by nivolumab (A), ipilimumab (B) and nivolumab plus ipilimumab (C). The figure is reprinted from Tang et al. (2021) "The pattern of time to onset and resolution of immune-related adverse events caused by checkpoint inhibitors in cancer: a pooled analysis of 23 clinical trials and 8,436 patients", Cancer Research & Treatment, 52(2):339-354 [128]. Licensed under CC BY-NC 4.0, adapted (statistical values removed).

Treatment of IRAEs and impact on efficacy

As previously mentioned, numerous studies report an association between the development of IRAEs and treatment efficacy with CPIs [100-104]. When a low-grade IRAE develops, treatment can often continue with intensified monitoring, and patients may be treated with topical steroids (for cutaneous IRAEs), hormone replacement (for endocrine IRAEs) or loperamide (for gastrointestinal IRAEs). For grade ≥ 3, and in some cases of grade 2 IRAEs, treatment must be temporarily or permanently withheld, and high-dose corticosteroids (0.5-2 mg/kg) are recommended [130]. If symptoms do not improve, various second line immunosuppressive treatments are recommended including mycophenolate-mofetil, TNF inhibition and intravenous immunoglobulins [130]. Several retrospective and non-randomized studies have explored the effects of glucocorticoids on the outcomes of patients treated with CPIs and found no negative association when steroids were used to treat IRAEs [131-

133], even though conflicting results have been reported [134]. Cook et al. observed an improved OS in patients who developed IRAEs, even in cases of severe IRAEs requiring hospitalization [135]. In a systematic review and meta-analysis, patients receiving steroids were compared to non-steroid users. A total of over 4000 patients were included. Patients taking steroids for supportive care or brain metastasis experienced reduced PFS and OS, compared to non-steroid users. However, the use of steroids for the treatment of IRAEs did not negatively affect OS [132].

Retrospective studies of patients with CMM suggest an association between a high corticosteroid peak-dose and poorer PFS and OS [136, 137]. In a recent study by Verhejden et al., the associations between peak and cumulative corticosteroid doses, as well as second line immunosuppressive treatment, with PFS and OS were investigated. A post hoc analysis was performed on patient data from six clinical trials involving patients with various malignancies treated with a combination of PD-1 and CTLA-4 inhibitors. A total of 1959 patients received treatment, of whom 834 (43%) required immunosuppressive treatment for IRAEs. High corticosteroid peak doses were associated with worse PFS and OS. The adjusted hazard ratio (HRadj) for PFS was 1.15 (95% CI 1.02-1.29) for 1 versus 0.5 mg/kg prednisolone and HRadj 1.43 (95% CI, 1.05-1.96) for 2 versus 0.5 mg/kg. The corresponding results for OS were HRadj 1.21 (95% CI 1.06-1.39) and HRadj 1.66 (95% CI 1.17-2.37) for 1 and 2 versus 0.5 mg/kg, respectively. No significant association was observed between cumulative corticosteroid dose and PFS or OS. The number of patients receiving second line immunosuppressive treatment was too small to draw any conclusions regarding its association with survival. The authors state that, based on the results, clinicians should consider starting with a lower corticosteroid dose whenever possible [138].

Impact of antibiotic use on efficacy of checkpoint inhibitors

Not all patients respond to treatment with CPIs and the response varies depending on the tumor type. Response rates have been reported to range from 15 to 60% influenced by the type of cancer [139]. In recent years, extensive research has highlighted the role of the gut microbiome in modulating the immune system. Studies suggest that gut bacteria can be associated with response, as well as lack of response, in cancer patients treated with CPIs [140]. Notably, fecal microbiota transplantation (FMT) from CPI responders to germ-free or antibiotic-treated mice has been shown to improve the response in these mice models [140]. Additionally, FMT together with PD-1 inhibitor

has been tested in patients with PD-1-refractory CMM. Clinical benefit was reported in 6 of 15 patients [141].

The use of antibiotics is known to alter the gut microbiome, disrupting the balance between various bacterial groups. This disruption can persist for weeks, months or even years following antibiotic use [142]. Several meta-analyses have shown a significant correlation between antibiotic use and reduced treatment efficacy with CPIs [143-145]. The first meta-analysis, published in 2019, included 2740 cancer patients and found that antibiotic use was negatively associated with both OS (HR=2.37; 95%CI=2.05-2.75; P<0.001) and PFS (HR=1.84; 95% CI = 1.49-2.26; P< 0.001). The findings were consistent across subgroup analyses stratified by timing of antibiotic use and cancer type [144]. Another meta-analysis, which included almost 13 000 patients demonstrated that antibiotic use was associated with poorer OS regardless of if antibiotics were used <1 month from CPI initiation (HR=1.83, 95%CI 1.38-2.43) or >1 month from CPI initiation (HR=2.02, 95% CI 1.51-2.7). In contrast, the association between antibiotic use and disease progression was significant only when antibiotics were taken more than 1 month after of starting CPI treatment (OR 2.15, 95% CI 1.17-3.95) [145]. In 2023, Crespin et al. published a systematic review and meta-analysis that included over 40 000 patients with various cancers, with 28% having received antibiotics around the time of CPI initiation. The pooled HRs for OS and PFS were 1.69 (95% CI 1.48-1.76) and 1.45 (95% CI 1.32-1.6) confirming that antibiotic use was significantly associated with shorter survival. The negative impact on survival was observed across all cancer types for OS, while it varied by cancer type for PFS. The loss of survival was particularly pronounced when antibiotics were administered shortly before or after CPI initiation [143].

Predictive markers for treatment response

Several predictive markers for treatment response to CPIs have been identified. MSI-H and dMMR have been linked to improved treatment efficacy across various cancer types, leading to the approval of pembrolizumab, as well as dostarlimab, for various cancers [96]. Tumor mutational burden (TMB) has also been correlated to favorable treatment outcomes, resulting in a tumor-agnostic approval for pembrolizumab in TMB-high malignancies [96]. However, while high TMB can indicate a better treatment response, it does not guarantee treatment success, and conversely, low TMB does not exclude the possibility of a response, reflecting the limitation of this marker. PD-L1 expression is strongly associated with treatment response in certain tumor types, with some requiring a specific threshold of expression to be eligible for

treatment. Nevertheless, responses can still occur even in patients with low PD-L1 expression [81]. Additionally, several concerns surround the assessment of PD-L1 status, including variability between analyses performed on primary tumors versus metastatic sites, multiple scoring algorithms, inter-assay heterogeneity, potential alterations in expression following prior chemotherapy, and interobserver variability among pathologists interpreting the results [146, 147]. Research is ongoing to identify additional and reliable markers for treatment response.

Predictive markers of IRAEs

Extensive research has been conducted to identify risk factors for development of IRAEs. Identifying a predictive marker prior to drug administration would be of great value in identifying high-risk patients, enabling optimized treatment decisions and follow-up strategies, to prevent development of severe IRAEs. Several patient-specific factors have been explored in this context.

High age has been considered as a potential factor linked to an increased risk of IRAEs, but no significant association was found in pivotal randomized trials [148] or in real-world evidence studies [121, 149-151], even if conflicting results exists [152]. Treatment with CPIs appears to be safe and effective even in patients with poor PS although response rates and OS are notably lower in this group compared to patients with PS 0-1 [149, 153, 154]. This suggests that PS alone does not predict the development of IRAEs. Further, patients with a high-BMI have been observed to have an increased risk of IRAEs [155].

Gender differences in IRAEs have been investigated with conflicting results. Two large meta-analyses found no statistically significant IRAE risk difference between males and females [156, 157]. One retrospective study including almost 4000 patients found an increased risk of all-grade IRAEs in females [158]. A systematic review found that men tend to have slightly lower frequencies of thyroid IRAEs but a higher frequency of hypophysitis, neurologic and vascular IRAEs, as well as Sicca/Sjogren's syndrome, while no significant differences in hematologic or gastrointestinal IRAEs were observed between genders [159]. Additionally, men have been reported to be at a higher risk of immune mediated cardiac events [160].

Patients with pre-existing autoimmune disease have typically been excluded from clinical trials. A systematic review and meta-analysis of observational studies and uncontrolled trials included nearly 24, 000 patients from 95 studies, with autoimmune diseases treated with CPIs. The analysis revealed that patients with autoimmune disease were more likely to develop IRAEs

compared to those without autoimmune disease, with a relative risk of 1.3 (95% CI 1.0-1.6). The pooled occurrence rate of any IRAEs was 61% and that of flares 36% and 32% of the patients with IRAEs required hospitalization. No statistically significant difference in treatment response was observed between patients with or without autoimmune disease. The authors concluded that CPIs can be used in patients with pre-existing autoimmune disease, but close monitoring is essential [161]. Another review and meta-analysis of 52 articles found that patients with autoimmune disease had a higher risk of experiencing IRAEs of any grade (RR: 1.23, 95% CI: 1.12-1.35) and were more likely to discontinue treatment due to IRAEs (1.4, 95% CI: 1.11-1.78). However, no statistically significant differences were observed in the rates of grade ≥ 3 IRAEs, ORR, disease control rate, OS or PFS between the two groups. Like the previous analysis, the authors of this study conclude that CPIs are effective in patients with autoimmune disease and emphasized rigorous monitoring [162].

Numerous biomarkers have been explored as potential predictors of IRAEs including circulating blood cell counts and ratios, cytokines, T-cell-expansion and diversification, autoantibodies, serum proteins, HLA genotypes, microRNA, gene expression profiling and the composition of the gastrointestinal microbiome [163]. However, the findings across studies have been difficult to generalize due to limitations such as retrospective design, short follow-up periods, and a focus on specific cancer types, IRAEs or CPIs. Two recent review articles underscore the need for well-designed prospective studies to more accurately evaluate the predictive value of these candidate biomarkers [163, 164].

Aims of the thesis

The overall aim of this doctoral project is to improve healthcare for melanoma patients by investigating IRAEs associated with CPI therapy and evaluate the impact of adding whole-body imaging to the follow-up after radical surgery for high-risk CMM on patient outcomes.

The specific aims of the studies included:

- To investigate whether a simplified frailty score, made up of parameters easily assessable in everyday clinical life, can be used as a predictive tool for patients treated with CPIs (Paper I).
- To investigate the association between development of multiple IRAEs and treatment outcomes in patients treated with CPIs (Paper II).
- To compare treatment efficacy and the occurrence of IRAEs between PD-1 and PD-L1 antibody therapies in a real-world cohort (Paper III).
- To assess whether the addition of imaging to the follow-up program for patients with high-risk CMM improves survival through earlier detection of recurrence (Paper IV).

Patients and methods

Paper I, II and III

Paper I evaluates a frailty score composed of parameters readily available in clinical practice among patients treated with CPIs. Paper II investigates the patterns and clinical impact of multiple IRAEs on CPI effectiveness. Paper III evaluates differences in the occurrence of IRAEs between patients treated with PD-1 and PD-L1 inhibitors in a real-world setting.

All three studies are multicenter retrospective cohort studies. Paper I and II include patients from Södermland, Örebro and Uppsala counties in Sweden who received CPIs, either PD-1 or PD-L1 inhibitors as monotherapy or PD-1 inhibitor in combination with anti-CTLA-4 therapy, for advanced malignancies between January 1st 2017 and December 31st 2021. Paper III includes patients treated with PD-1 or PD-L1 inhibitor monotherapy from the same cohort and from Stockholm County between June 2016 and August 2022.

Data was extracted from electronic medical records (EMR) by researchers using pre-specified parameters. Collected variables included age at diagnosis, gender, comorbidities expressed as Charlson Comorbidity Index (CCI), type of cancer, primary treatment at diagnosis, age at diagnosis of advanced cancer, metastatic sites, CPI initiation date, type of CPI, performance status (PS; WHO classification) at CPI initiation, number of previous lines of treatment, best response to CPI therapy, date of disease progression, IRAEs (date, type, grade, outcome), date of death and cause of death.

IRAEs were graded according to the CTCAE grading system, version 5.0. If the grade was not explicitly stated in the EMRs, grades were approximated based on clinical descriptions and laboratory findings, when possible. Multiple IRAEs were defined as IRAEs affecting more than one organ system, either concurrently or sequentially. To mitigate the risk of immortal time bias (ITB) in Paper II, a time-depending Cox-regression model was applied.

In Paper I, the frailty score was calculated based on age, PS and CCI as follows:

- Age: ≤ 75 years = 0 points; 76-80 years = point; > 80 years = 2 points
- PS: 0 = 0 points; 1 = 1 point; 2 = 2 points
- CCI; ≤ 1 = 0 points; CCI > 1 = 1 point

Patients with a sum of score ≥ 2 were considered frail, and those with 0-1 as non-frail.

Paper IV

Paper IV reports interim results from TRIM (NCT03116412) a nationwide prospective randomized phase III trial. After radical surgery of stage IIB-C and III CMM, patients were randomly assigned 1:1 (stratified for tumor stage) to follow-up with physical examinations and regular ultrasound examinations for sentinel node positive participants for 3 years, or with physical examinations plus whole-body imaging with CT or FDG-PET-CT at baseline, 6, 12, 24 and 36 months.

The first patient was enrolled in June 2017 and interim analysis was performed after inclusion of 1000 patients in August 2023. The target sample size of 1300 patients has recently been reached. The primary aim of the interim analysis was to determine whether the study should continue to include patients according to protocol. The primary endpoint of the trial is OS at 5 years, and secondary endpoints include health-related quality of life. For the present interim analysis, endpoints included RFS, DMFS, locoregional relapse-free survival (LRFS), and OS, with the primary focus on evaluating relative differences between study arms across the entire follow-up period.

Key inclusion criteria were age ≥ 18 years, signed and dated informed consent, radical surgery for stage IIB-C and III CMM, and sufficient renal function for iv contrast-enhanced CT. Key exclusion criteria included life-expectancy < 2 years due to comorbid disease, inability to comply with the follow-up program, or assessment as unfit to receive treatment in case of recurrence. Following an amendment in February 2022, it was also possible to include participants after radical surgery for Merkel cell carcinoma.

Results

Paper I and II

Study cohort characteristics

A total of 596 patients were included in the study cohort for Paper I and 600 patients in Paper II, with a median age of 66 years. The most common underlying malignancy was NSCLC (34%), followed by CMM (33%) and RCC (15%). The most frequently administered CPI was nivolumab monotherapy (47%) followed by pembrolizumab monotherapy (35%), while 7% of patients received combination therapy with nivolumab and ipilimumab. Median follow-up time in cohort II was 15 months (range: 1 to 95 months; IQR: 6 to 28).

Paper I

Frequency, outcomes and prediction of IRAEs

Using the simplified frailty-score, 361 patients (60.6%) were classified as non-frail and 235 (39.4%) as frail. In multivariate analyses, age, CCI and PS alone were not independently associated with IRAE occurrence or treatment discontinuation due to IRAEs. When applying the simplified frailty score, frail patients had a significantly higher risk of developing any-grade IRAEs (OR: 1.58; 95% CI: 1.09-2.28) and multiple IRAEs (OR: 1.62; 95% CI: 1.00-2.64), though not high-grade IRAEs or treatment discontinuation. No significant differences were observed between frail and non-frail patients regarding IRAE resolution or worsening.

Paper II

Characteristics of multiple IRAEs and impact on efficacy

Out of 600 patients, 97 (16.2%) developed multiple IRAEs, while 197 (32.8%) experienced a single IRAE. Patients with multiple IRAEs were significantly more likely to develop grade ≥ 2 (93.8% v s 65.0% $p < 0.001$), grade ≥ 3 IRAEs (43.3% v s 31.5% p=0.046) and to discontinue treatment due to IRAE

(44.3% v s 28.9% p=0.009), compared to patients with a single IRAE. Patients with a PS ≥ 2 were less likely to develop multiple IRAEs (OR: 0.42 95% CI: 0.24-0.75) whereas combined CPI therapy was strongly associated with an increased risk (OR: 4.64 95% CI: 1.83-11.9). No specific risk factors were identified that distinguished patients with multiple IRAEs from those with a single IRAE.

Patients with multiple IRAEs showed significantly improved PFS (HR=0.78 95% CI: 0.57-0.98) and OS (HR=0.65 95% CI: 0.44-0.95) compared to patients with single IRAE or no IRAE (HR=0.46 95% CI:0.34-0.62 for PFS vs HR=0.41 95% CI: 0.28-0.60 for OS) in the overall cohort, as demonstrated in Figure 7 and 8.

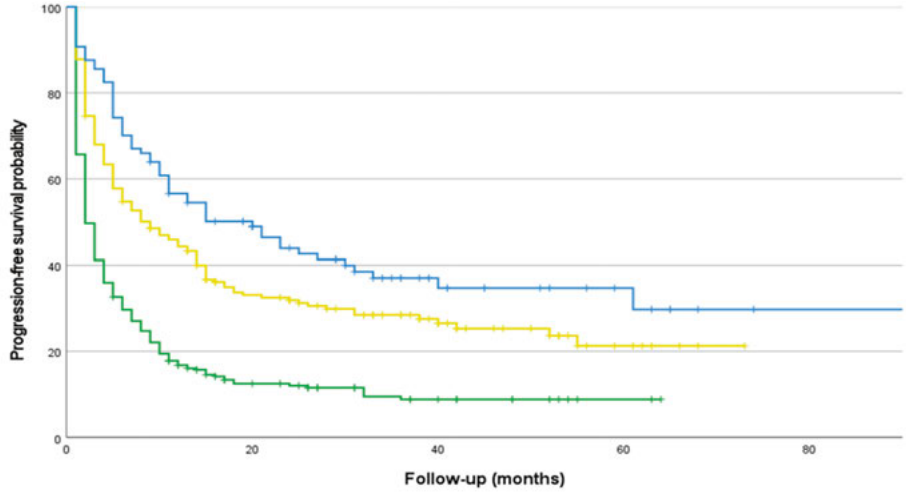

Figure 7. Progression-free survival in a cohort of cancer patients with advanced disease treated with checkpoint inhibitors (n=600) based on the occurrence of immune-related adverse events. Blue line for multiple IRAEs; yellow line for single IRAE; green line for no IRAE.

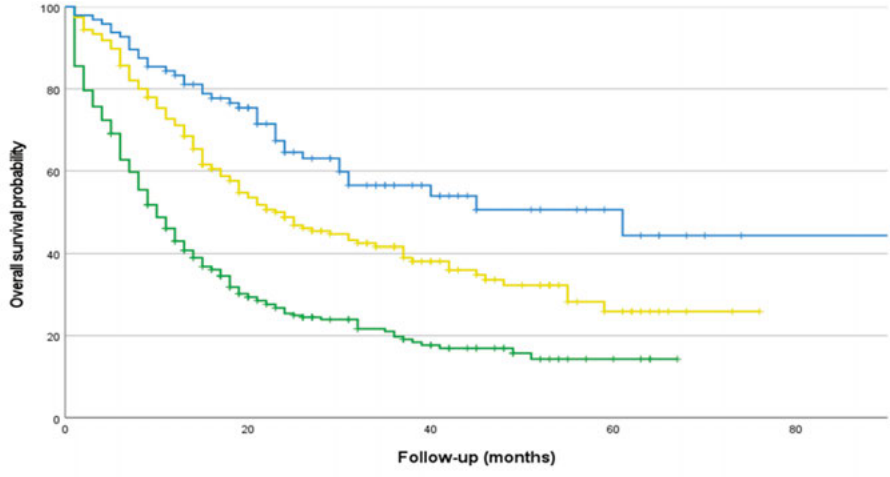

Figure 8. Overall survival in a cohort of cancer patients with advanced disease treated with checkpoint inhibitors (n=600) based on the occurrence of immune-related adverse events. Blue line for multiple IRAEs; yellow line for single IRAE; green line for no IRAE.

Paper III

Characteristics of the study cohort

A total of 605 patients were included, with a median age of 67 years (range: 20-87). The most common malignancy was NSCLC (41.5%), followed by CMM (28.6%), RCC (11.7%), urothelial carcinoma (5.8%), head and neck squamous cell carcinoma (3.8%) and other malignancies (8.6%). Most patients were treated with PD-1 inhibitors (82.3%), while the remaining patients received PD-L1 inhibitors (17.7%).

Notable differences in cancer type distribution were observed between treatment groups. Among patients treated with PD-1 inhibitors, CMM was the most common cancer type (34.5%), followed by NSCLC (30.1%) and RCC (14.3%). In contrast, nearly all patients treated with PD-L1 inhibitors had NSCLC (94.4%).

Occurrence of IRAEs with PD-1 versus PD-L1 inhibitors

IRAEs grade ≥ 2 occurred significantly more often in patients treated with PD-1 inhibitors compared with those receiving PD-L1 inhibitors (35.7% vs. 22.4% p=0.008). Treatment discontinuation due to toxicity was also more common in the PD-1 inhibitor group (17.3% vs. 7.5% p=0.011). Although not statistically

significant, numerically higher rates of any-grade IRAEs, IRAEs grade ≥ 3 and multiple IRAEs were observed among patients treated with PD-1 inhibitors.

Adjusted models on the impact of CPI type on IRAEs revealed treatment with PD-L1 inhibitors was associated with a significantly lower risk of developing IRAEs grade ≥ 2 (OR: 0.63; 95% CI: 0.35-0.98) and treatment discontinuation due to IRAEs (OR: 0.38; 95% CI: 0.16-0.88) compared with PD-1 inhibitors. When the analyses were restricted to patients with NSCLC (as most of the patients treated with PD-L1 inhibitors had NSCLC), similar trends were observed, although the results did not reach statistical significance (IRAEs grade ≥ 2 OR: 0.63; 95% CI: 0.33-1.19; treatment discontinuation due to IRAEs: OR: 0.46; 95% CI: 0.19-1.12).

Paper IV

A total of 1018 participants were included at the time of data extraction in August 2023. Nine participants were on surveillance after treatment for Merkel cell carcinoma and therefore excluded, and one participant was never randomly assigned to a group, resulting in 1008 participants. Following randomization, 25 participants withdrew their consent, and ten participants were lost to follow up, resulting in 498 followed up with physical examinations and regular ultrasound examinations for sentinel node positive participants (standard arm) and 485 to follow up with physical examinations and whole-body imaging (experimental arm).

A total of 159 relapses were detected in the standard arm and 177 in the experimental arm. Among these, distant relapses (including patients with a synchronous locoregional relapse) numbered 78 and 90, while locoregional relapses totaled 81 and 87 in the standard versus experimental arm. Overall, 53 patients in the standard arm and 54 patients in the experimental arm died during follow-up.

In time-to-event analysis, there were no statistically significant differences in RFS, DMFS, locoregional relapse free survival (LRFS) or OS between the groups at a median follow-up time of 33.6 (IQR 16.3-49.8) months. Three-year rates for RFS were 69.3% (95% CI 64.9 -74.0%) and 67.1% (95% CI 62.5-72.0%), for LRFS 84.3% (95% CI 80.9-87.9%) versus 84.1% (95% CI 80.6-87.8%), for DMFS 81.6% (95% CI 77.9-85.6%) versus 79.3% (95% CI 75.3-83.5%) and for OS 88.2% (95% CI 85.0-91.6%) versus 87.7% (95% CI 84.3-91.3%) in the standard and imaging groups, respectively.

Within the experimental group, no statistically significant differences were observed between the surveillance methods (CT or FDG-PET-CT) regarding LRFS, DMFS, RFS or OS.

Discussion

Over the past decade immunotherapy with CPIs has become a cornerstone in the treatment of several malignancies. Before the introduction of CPIs and targeted therapies, the prognosis for patients with metastatic CMM was poor, with a median OS of less than one year and a 5-year survival rate of approximately 10% [165]. Survival outcomes have since improved significantly with these therapies, although exact figures vary depending on patient and disease characteristics, as well as the treatments given [166]. Although effective, CPIs are associated with IRAEs, which, although often manageable and reversible with timely intervention and adherence to established guidelines, may occasionally be severe, persistent or life-threatening [105, 167].

Identifying patients at increased risk of developing IRAEs is essential for optimizing treatment decisions and follow-up strategies. Against this background, we conducted the study presented in Paper I, aiming to determine whether a simplified frailty score, based on age, PS and CCI, could predict the development of IRAEs. The score was originally developed to define transplant-ineligible patients with newly diagnosed multiple myeloma, and its prognostic significance has been validated [168-170]. We applied it in a retrospective multicentre cohort study of nearly 600 CPI-treated patients with advanced malignancies. Of the total cohort, approximately 60 % were classified as nonfrail and 40 % as frail. Frail patients experienced a higher incidence of any grade and multiple IRAEs, however, no individual component of the score (i.e., age, CCI or PS) were independently associated with an increased risk of IRAEs. Moreover, the frailty score did not identify patients at increased risk of severe (grade 3-4) IRAEs or treatment discontinuation, which represent the most clinically relevant outcomes.

Although age has previously been considered a potential predictive factor for IRAEs [152], neither pivotal randomized trials [148] nor real-world studies have confirmed this association [121, 149-151]. Our results are consistent with these results, demonstrating no significant association between age and either IRAE development or treatment discontinuation. Similarly, although CPIs appear safe and effective even in patients with poor PS, lower response rates and reduced OS have been observed in this group [149, 153, 154]. In our study, PS alone did not predict IRAE development. Comorbidity, assessed by CCI,

showed no independent association with IRAEs, supporting previous evidence that CPIs may be safe even in patients with certain comorbidity, including pre-existing low-risk autoimmune disease, mild to moderate hepatic and renal dysfunction and some chronic viral infections [171-174].

The association between IRAEs and treatment efficacy has been extensively investigated, with multiple studies demonstrating improved outcomes among patients who develop IRAEs [100-104]. However, IRAEs affecting multiple organ systems, referred to as multiple IRAEs, have been less extensively studied. Paper II addresses this gap by evaluating the clinical characteristics and prognostic impact of multiple IRAEs in a cohort of 600 patients with advanced cancer treated with CPIs. Approximately one-sixth of patients developed multiple IRAEs, which were associated with a significantly increased risk of high-grade toxicity and treatment discontinuation. Despite this, the occurrence of multiple IRAEs was strongly associated with improved PFS and OS, exceeding the benefit observed with single IRAEs.

Importantly, this association was evaluated using time dependent Cox regression models to mitigate immortal time bias (ITB), a major methodological limitation in earlier studies. There are only a few previous studies, most of them indicating a better prognosis in patients with multiple IRAEs compared to patients with single IRAEs [175-180], even if conflicting results exists [181]. However, previous reports on multiple IRAEs have often failed to account adequately for ITB, some studies did not account for ITB at all [178, 179], while others used landmark analyses [176, 177, 181], which can still induce bias when compared to the more robust time-dependent Cox model [182]. Moreover, most studies have focused exclusively on patients with NSCLC [175-177, 180, 181]. We only identified two previous studies that appropriately accounted for ITB and reported a positive association between multiple IRAEs and improved outcomes [175, 180], yet both were restricted to NSCLC cohorts. By including a broader cancer population and applying robust statistical methods, our findings provide novel and more generalizable evidence supporting an association between multiple IRAEs and favourable treatment outcomes.

Given the distinct mechanisms of action of PD-1 and PD-L1 inhibitors, differences in toxicity profiles are biologically plausible. Meta-analyses of clinical trials have suggested a higher incidence of IRAEs with PD-1 inhibitors compared with PD-L1 inhibitors [109-111, 114]. However, no previous study investigated this in a real-world population and conflicting results exist [183-185]. Paper III addresses this gap by comparing IRAEs in a cohort of more than 600 patients with advanced solid malignancies treated with PD-1 or PD-L1 inhibitor in monotherapy. Treatment with PD-L1 inhibitors was

associated with a significantly lower risk of IRAEs grade ≥ 2 and treatment discontinuation due to IRAEs. Although numerical differences favouring PD-L1 inhibitors were also observed for other toxicity outcomes, these did not reach statistical significance. These findings suggest a more favourable tolerability profile for PD-L1 inhibitors, which may be clinically relevant when treatment options are otherwise equivalent.

Paper IV presents interim results from the TRIM study, the largest interventional trial of CMM patients ever conducted in Sweden. This randomized, phase III trial evaluates whether the addition of scheduled whole-body imaging to standard follow-up improves survival after radical surgery for stage IIB-C and III CMM. At a median follow-up of 33.6 months, no survival benefit was observed for patients undergoing routine whole-body imaging compared with those followed with clinical examinations and ultrasound for sentinel node positive patients.

Postoperative surveillance strategies for high-risk CMM vary considerably between countries due to limited supporting scientific evidence [186-188]. Historically, routine imaging was considered of limited value because effective systemic therapies were unavailable. Although scheduled imaging has been shown to detect distant metastases earlier in high-risk patients [189-191], it has been uncertain whether earlier diagnosis of relapse translates into improved survival. In line with previous non-randomized studies, our interim analysis found no survival benefit associated with earlier detection of relapses by scheduled whole-body surveillance [192-194].

The cohort studies presented have several limitations. First, the retrospective design of Papers I-III increases the risk of bias, including underreporting and misclassification of IRAEs. Although standardized grading criteria were applied, reliance on EMRs may have led to incomplete capture of low-grade events. In Paper I, an additional limitation is the fact that the simplified frailty score originally was developed in a different disease context, which may limit its ability to capture frailty dimensions specific to patients receiving immunotherapy. In Paper II, ITB represents the primary potential confounder in assessing the association between IRAEs and clinical outcomes. Although a robust time-depending Cox-regression model was applied to address this issue, residual bias cannot be fully excluded. Paper III is limited by imbalance in cancer types between treatment groups, although multivariate adjustment suggests that the main findings are not driven by this heterogeneity.

Conclusions

I. The simplified frailty score, derived from age, CCI and PS predicted occurrence of all grade IRAEs and multiple IRAEs in multivariate analyses, in a cohort of patients with various malignancies treated with CPIs in advanced setting. In contrast, age, CCI or PS did not separately predict IRAEs. However, the difference in IRAEs between frail and nonfrail patients did not reach statistical significance for patients with severe (grade 3-4) toxicity or treatment discontinuation.

II. Patients with various malignancies treated with CPIs in advanced setting demonstrates a stronger association between development of multiple as opposed to single IRAEs and CPI treatment effectiveness (measured as PFS and OS). The findings suggest that multiple IRAEs may constitute a surrogate marker for treatment efficacy. However, further studies with a larger sample size and prospective design to overcome the inherent biases of retrospective studies are needed to further confirm the association.

III. This real-world study of patients with various malignancies treated with anti-PD-1 or anti-PD-L1 monotherapy in advanced disease supports previous results from clinical trials demonstrating a favourable toxicity profile for PD-L1 inhibitors versus PD-1 inhibitors. However, further studies utilizing large datasets from real-world data are needed to validate our findings.

IV. The interim analysis of the TRIM study indicates no survival benefit from the addition of whole-body imaging to the follow-up protocol after radical surgery of stage IIB-C and III CMM. However, the median follow-up time is relatively short, and so far, only a few patients have completed the follow-up time of 5 years. Therefore, the results must be interpreted with caution. The numerical differences between the study groups in RFS and DMFS motivate to continue the study according to protocol.

Future perspectives

The studies in this thesis contribute to a better understanding of IRAEs from treatment with CPIs in patients with various malignancies and give a better scientific ground for follow-up strategies in high-risk CMM patients. However, several questions remain and there is a need for further investigations.

In the first study, the frailty score did not identify patients at risk for high-grade IRAEs or treatment discontinuation, which is the clinically most relevant IRAEs. Though, there is still a need to find a tool that could be used in everyday clinical life to predict toxicity. Finding an immunotherapy-specific model for assessing frailty would be of great value. How to design such a model is yet to find out. Maybe such a model could benefit from incorporating preexisting autoimmune disease, body composition measures and laboratory-based biomarkers.

It is now well known that development of IRAEs is associated with improved treatment outcome. In the second study we observed an association between development of multiple IRAEs and improved efficacy, with better outcomes among patients developing multiple IRAEs as compared to those with a single IRAE. This raises an important clinical question: how can efficacy be maintained while avoiding IRAE? Studies of the underlaying mechanisms, to reveal if there is a difference when a single or multiple IRAEs develop, might bring further insight in identifying predictive markers for toxicity and efficacy. Another approach could be to prophylactically use agents aiming at preventing IRAE development in combination with CPI. Interleukin-6 overexpression is associated with therapeutic resistance to treatment with CPI, as well as development of toxicity, and is therefore of particular interest. Multiple trials are currently ongoing across various malignancies [195]. In melanoma the approach is evaluated in two small phase II trials, combining the IL-6 inhibitor tocilizumab with ipilimumab and nivolumab. Preliminary data suggest that tocilizumab may reduce high-grade IRAEs while maintaining efficacy [196, 197]. The final results from these trials are awaited with great interest.

The results from Paper III suggest clinically relevant differences in toxicity profiles between PD-1 and PD-L1 inhibitors in a real-world setting. As treatment indications expand and therapeutic options increase comparative studies of efficacy and toxicity will become even more important. Large studies could

provide sufficient power to explore differences in toxicity and efficacy across different tumor types.

Finally, the results from the TRIM trial suggest that adding whole-body imaging to the follow-up program after radical surgery for high-risk CMM does not confer a survival benefit. However, these findings should be interpreted with caution, as only a limited number of patients had completed the full five-year follow-up at the time of the analysis. Definitive conclusions must await the final results. If these findings are confirmed, it is likely that many international follow-up programs will adopt their follow-up routine. However, it is possible that the value of early relapse detection will change in the future, with introduction of new effective treatments. It will also be interesting to evaluate whether diagnostic sensitivity differs between patients monitored with CT and those monitored with FDG-PET, as well as whether the rate of false positive findings varies between modalities, although the study may not have sufficient power to fully address these questions.

Liquid biopsy is a promising tool for identifying circulating molecular residual disease in patients with resected melanoma. Circulating tumor DNA (ctDNA) is associated with worse PFS and OS in metastatic melanoma [198]. In a review of eight studies of patients with resected non-metastatic melanoma, ctDNA detection rates ranged from 4-63%, with sensitivity of 11-80% and specificity of 55-100%. Differences in testing methods (e.g., whole genome, exome or non-target approaches) and testing schedule (single vs serial tests) influence sensitivity [198]. In the future, ctDNA surveillance may play a role in follow-up strategies, potentially helping guide the use of imaging.

Populärvetenskaplig sammanfattning på svenska (Summary in Swedish)

Malignt melanom är den form av hudcancer som orsakar flest dödsfall. Antal diagnostiserade melanom har ökat kraftigt i Sverige under de senaste decennierna, en trend som också ses globalt. År 2014 diagnostiserades cirka 3800 maligna melanom i Sverige, jämfört med cirka 6000 år 2024, en ökning med 60% på 10 år. Malignt melanom är därmed den cancerform som ökat snabbast i landet. Majoriteten av maligna melanom som diagnostiseras är tunna (≤ 1mm) och har mycket god prognos.

Behandling av melanom är i första hand kirurgisk. Patienter med hög risk för återfall erbjuds ofta ett års medicinsk tilläggsbehandling efter operation. Uppföljning sker enligt ett nationellt vårdprogram med kliniska kontroller under tre år, där läkarbesök med undersökning av hud och lymfkörtlar ingår. För patienter som haft sjukdomsspridning till lymfkörtlar ingår även ultraljudsundersökning av den angripna lymfkörtelstationen. Rutinmässig helkroppsröntgen ingår inte. Vid misstanke om återfall görs riktad undersökning, ofta med röntgen.

Tidigare fanns mycket begränsade möjligheter att behandla patienter som utvecklade spridd (generaliserad) melanomsjukdom. Den behandling som fanns hade oftast kortvarig effekt och hjälpte endast ett fåtal patienter. Därför fanns det heller inget tydligt värde i att aktivt leta efter återfall. Under de senaste 15 åren har dock behandlingsmöjligheterna förbättrats avsevärt genom införandet av immunterapi och målstyrd behandling (BRAF + MEK-hämmare). Det första immunterapiläkemedlet godkändes just för malignt melanom. Numera är immunterapi godkänt för behandling av många olika cancersjukdomar, men malignt melanom är en av de cancerformer som svarar allra bäst på behandlingen.

Immunterapi verkar genom att aktivera kroppens immunförsvar så att det kan angripa cancerceller. Biverkningar uppstår när immunsystemet angriper frisk vävnad och orsakar inflammation. Dessa inflammationer kan drabba i princip alla organ och liknar autoimmuna sjukdomar. Oftast är biverkningarna behandlingsbara och går tillbaka, men i vissa fall kan de bli kroniska och i sällsynta fall livshotande. Patienter med autoimmuna sjukdomar har en ökad

risk för försämring av sin grundsjukdom och för andra immunrelaterade biverkningar. I övrigt är kunskapen begränsad kring vilka faktorer som ökar risken för sådana biverkningar.

I delarbete I undersöktes om ett skörhetsindex kan användas för att förutsäga risken för immunrelaterade biverkningar. Patienterna klassificerades som sköra eller icke-sköra utifrån allmäntillstånd, samsjuklighet och ålder. Sköra patienter utvecklade fler immunrelaterade biverkningar i allmänhet, samt biverkningar från flera organsystem (multipla). Däremot sågs ingen ökad risk för svåra biverkningar, eller biverkningar som krävde att behandlingen fick avbrytas. Därav bedöms detta skörhetsindex ha begränsat kliniskt värde, då det är särskilt viktigt att kunna identifiera patienter med risk för allvarliga biverkningar.

Det är känt att patienter som utvecklar immunrelaterade biverkningar ofta har bättre effekt av immunterapi. Sambandet mellan biverkningar från flera organsystem hos samma patient (multipla) och behandlingseffekt är dock mindre studerat. I delarbete II undersöktes detta hos patienter med olika cancersjukdomar. Resultaten visade att patienter som utvecklade multipla biverkningar hade signifikant bättre behandlingsresultat, mätt som både längre till sjukdomsförsämring och längre total överlevnad, jämfört med de som inte utvecklade någon biverkan alls eller de som utvecklade en enstaka biverkan.

Immunsystemet har både gas- och bromssystem för att kunna angripa hot utan att skada kroppen. En viktig broms utgörs av proteinet PD-1 på ytan av immunförsvarets T-celler. Många cancerceller uttrycker PD-L1 på sin yta, som genom bindning till PD-1 skickar en stopp-signal till immunförsvaret och därmed undgår attack. PD-1- och PD-L1-hämmare är två närbesläktade typer av immunterapi som blockerar denna signal på olika sätt. Kliniska prövningar tyder på att PD-1-hämmare är förknippade med fler immunrelaterade biverkningar än PD-L1-hämmare men studier som direkt jämför dessa läkemedel saknas. Eftersom patienter i kliniska studier ofta är yngre, friskare och piggare än patienter i klinisk vardag var det viktigt att undersöka om det gäller även i en mer representativ patientpopulation. I delarbete III bekräftades att PD-L1-hämmare är förknippade med lägre risk för immunrelaterade biverkningar även hos ”vanliga” patienter med olika cancersjukdomar.

I takt med förbättrade behandlingsmöjligheter vid spridd melanomsjukdom har flera länder infört rutinmässig helkroppsröntgen i uppföljningsprogrammet efter operation av malignt melanom med hög risk för återfall. Uppföljningsprogrammen varierar dock kraftigt mellan länder, vilket speglar brist på vetenskapligt underlag. Studier har visat att återfall kan hittas tidigare med rutinmässig helkroppsröntgen, men det är oklart om detta leder till bättre överlevnad. Samtidigt kan röntgenundersökningar ibland visa

misstänkta förändringar som senare visar sig vara ofarliga, vilket kan orsaka onödig oro. Undersökningarna innebär också exponering för röntgen-bestrålning och är resurskrävande.

TRIM är en nationell randomiserad studie där patienter opererade för malignt melanom med hög risk för återfall antingen följs enligt det nationella vårdprogrammet (kliniska kontroller) eller med tillägg av helkroppsröntgen vid totalt fem tillfällen under tre år. Det primära målet är att jämföra överlevnaden efter 5 år. I delarbete IV presenteras resultaten från en interim analys efter att 1000 av planerade 1300 patienter inkluderats. Analysen visade ingen skillnad i överlevnad mellan grupperna vid en medianuppföljningstiden på 34,6 månader. Eftersom få patienter ännu nått full femårsuppföljning bör resultaten tolkas med viss försiktighet. Slutgiltiga resultat förväntas 2030. TRIM är en unik och omfattande studie med potential att internationellt påverka framtida uppföljningsprogram för patienter med malignt melanom.

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References

1. Erdmann, F., et al., *International trends in the incidence of malignant melanoma 1953-2008--are recent generations at higher or lower risk?* Int J Cancer, 2013. **132**(2): p. 385-400.
2. Tryggvadóttir, L., et al., *Trends in the survival of patients diagnosed with malignant melanoma of the skin in the Nordic countries 1964-2003 followed up to the end of 2006.* Acta Oncol, 2010. **49**(5): p. 665-72.
3. Nikolaou, V. and A.J. Stratigos, *Emerging trends in the epidemiology of melanoma.* Br J Dermatol, 2014. **170**(1): p. 11-9.
4. Sacchetto, L., et al., *Trends in incidence of thick, thin and in situ melanoma in Europe.* Eur J Cancer, 2018. **92**: p. 108-118.
5. Helgadottir, H., et al., *Multiple Primary Melanoma Incidence Trends Over Five Decades: A Nationwide Population-Based Study.* J Natl Cancer Inst, 2021. **113**(3): p. 318-328.
6. World Health Organisation. *Cancer Today*. 2022; Available from: https://gco.iarc.who.int/today.
7. Arnold, M., et al., *Global Burden of Cutaneous Melanoma in 2020 and Projections to 2040.* JAMA Dermatol, 2022. **158**(5): p. 495-503.
8. Socialstyrelsen. *Statistik om nyupptäckta cancerfall*. 2024 251104; Available from: https://www.socialstyrelsen.se/statistik-och-data/statistik/alla-statistikamnen/cancer/.
9. Registry, N.Q. *Swedish Melanoma Registry (SweMR)* Accessed 2026; Available from: https://statistik.incanet.se/Hudmelanom/.
10. *Svenska Melanomregsitret - SweMR Nationell årsrapport för hudmelanom diagnosår 1990-2024*. 2025; Available from: https://cancercentrum.se/download/18.29f46a199c90154403c01/1760104329410/A%CC%8Arsrapport%20SweMR%201990-2024.pdf.
11. Helgadottir, H., et al., *Melanoma Incidence and Mortality Trends Among Patients Aged 59 Years or Younger in Sweden.* JAMA Dermatol, 2024. **160**(11): p. 1201-1210.
12. El Sharouni, M.A., et al., *Sex matters: men with melanoma have a worse prognosis than women.* J Eur Acad Dermatol Venereol, 2019. **33**(11): p. 2062-2067.
13. Lundberg, F.E., et al., *Survival trends for patients diagnosed with cutaneous malignant melanoma in the Nordic countries 1990-2016: The NORDCAN survival studies.* Eur J Cancer, 2024. **202**: p. 113980.
14. Leonardi, G.C., et al., *Cutaneous melanoma: From pathogenesis to therapy (Review).* Int J Oncol, 2018. **52**(4): p. 1071-1080.
15. Alexandrov, L.B., et al., *Signatures of mutational processes in human cancer.* Nature, 2013. **500**(7463): p. 415-21.
16. Gosman, L.M., D.A. Țăpoi, and M. Costache, *Cutaneous Melanoma: A Review of Multifactorial Pathogenesis, Immunohistochemistry, and Emerging Biomarkers for Early Detection and Management.* Int J Mol Sci, 2023. **24**(21).

17. Strashilov, S. and A. Yordanov, *Aetiology and Pathogenesis of Cutaneous Melanoma: Current Concepts and Advances.* Int J Mol Sci, 2021. **22**(12).
18. Beadling, C., et al., *KIT gene mutations and copy number in melanoma subtypes.* Clin Cancer Res, 2008. **14**(21): p. 6821-8.
19. Sun, J., F. Chen, and G. Wu, *Potential effects of gut microbiota on host cancers: focus on immunity, DNA damage, cellular pathways, and anticancer therapy.* Isme j, 2023. **17**(10): p. 1535-1551.
20. Dunn, G.P., et al., *Cancer immunoediting: from immunosurveillance to tumor escape.* Nat Immunol, 2002. **3**(11): p. 991-8.
21. Zanetti, R., et al., *Comparison of risk patterns in carcinoma and melanoma of the skin in men: a multi-centre case-case-control study.* Br J Cancer, 2006. **94**(5): p. 743-51.
22. Gandini, S., et al., *Meta-analysis of risk factors for cutaneous melanoma: II. Sun exposure.* Eur J Cancer, 2005. **41**(1): p. 45-60.
23. Wu, S., et al., *Long-term ultraviolet flux, other potential risk factors, and skin cancer risk: a cohort study.* Cancer Epidemiol Biomarkers Prev, 2014. **23**(6): p. 1080-9.
24. *The association of use of sunbeds with cutaneous malignant melanoma and other skin cancers: A systematic review.* Int J Cancer, 2007. **120**(5): p. 1116-22.
25. Stanganelli, I., et al., *The association between pesticide use and cutaneous melanoma: a systematic review and meta-analysis.* J Eur Acad Dermatol Venereol, 2020. **34**(4): p. 691-708.
26. Davies, J.R., et al., *Development and validation of a melanoma risk score based on pooled data from 16 case-control studies.* Cancer Epidemiol Biomarkers Prev, 2015. **24**(5): p. 817-24.
27. Gershenwald, J.E., et al., *Melanoma staging: Evidence-based changes in the American Joint Committee on Cancer eighth edition cancer staging manual.* CA Cancer J Clin, 2017. **67**(6): p. 472-492.
28. Gershenwald JE, H.K., et al., *AJCC Cancer Staging Manual 8th edition*. 2017: Springer.
29. Helvind, N.M., et al., *Stage-Specific Risk of Recurrence and Death From Melanoma in Denmark, 2008-2021: A National Observational Cohort Study of 25 720 Patients With Stage IA to IV Melanoma.* JAMA Dermatol, 2023. **159**(11): p. 1213-1222.
30. Nelson, A., et al., *Neoadjuvant immunotherapy for patients with resectable stage III/IV cutaneous melanoma - A Swedish retrospective real-world study (NEO-MEL).* Eur J Cancer, 2025. **224**: p. 115485.
31. *Nationellt vårdprogram melanom, version 8.0*. last updated 2025-06-24 2025-09-16]; Available from: https://kunskapsbanken.cancercentrum.se/diagnoser/melanom/vardprogram/.
32. *MelmarT Melanoma Margins Trial Investigating 1cm v 2cm Wide Excision Margins for Primary Cutaneous Melanoma*. Available from: https://clinicaltrials.gov/ct2/show/NCT02385214.
33. Wennberg, E., et al., *Wise or wide (WoW) study protocol: a national, multicentre, prospective, randomised and controlled, parallel group, non-inferiority study to compare single-staged versus two-staged excisions of thin invasive (≤1.0 mm) melanoma.* BMJ Open, 2025. **15**(4): p. e094544.
34. Morton, D.L., et al., *Final trial report of sentinel-node biopsy versus nodal observation in melanoma.* N Engl J Med, 2014. **370**(7): p. 599-609.
35. Leiter, U., et al., *Final Analysis of DeCOG-SLT Trial: No Survival Benefit for Complete Lymph Node Dissection in Patients With Melanoma With Positive Sentinel Node.* J Clin Oncol, 2019. **37**(32): p. 3000-3008.

36. Broman, K.K., et al., *Active surveillance of patients who have sentinel node positive melanoma: An international, multi-institution evaluation of adoption and early outcomes after the Multicenter Selective Lymphadenectomy Trial II (MSLT-2).* Cancer, 2021. **127**(13): p. 2251-2261.
37. Blank, C.U., et al., *Neoadjuvant versus adjuvant ipilimumab plus nivolumab in macroscopic stage III melanoma.* Nat Med, 2018. **24**(11): p. 1655-1661.
38. Rozeman, E.A., et al., *Identification of the optimal combination dosing schedule of neoadjuvant ipilimumab plus nivolumab in macroscopic stage III melanoma (OpACIN-neo): a multicentre, phase 2, randomised, controlled trial.* Lancet Oncol, 2019. **20**(7): p. 948-960.
39. Versluis, J.M., et al., *Survival update of neoadjuvant ipilimumab plus nivolumab in macroscopic stage III melanoma in the OpACIN and OpACIN-neo trials.* Ann Oncol, 2023. **34**(4): p. 420-430.
40. Patel, S.P., et al., *Neoadjuvant-Adjuvant or Adjuvant-Only Pembrolizumab in Advanced Melanoma.* N Engl J Med, 2023. **388**(9): p. 813-823.
41. Patel, S.P., et al., *1601O 3-year survival with neoadjuvant-adjuvant pembrolizumab from SWOG S1801.* Annals of Oncology, 2025. **36**: p. S882-S883.
42. Blank, C.U., et al., *Neoadjuvant Nivolumab and Ipilimumab in Resectable Stage III Melanoma.* N Engl J Med, 2024. **391**(18): p. 1696-1708.
43. Lucas, M.W., et al., *LBA57 Two-year clinical update and first biomarker analyses of the phase III NADINA trial comparing neoadjuvant nivolumab plus ipilimumab versus adjuvant nivolumab in resectable stage III melanoma.* Annals of Oncology, 2025. **36**: p. S1600-S1601.
44. Weber, J., et al., *Adjuvant Nivolumab versus Ipilimumab in Resected Stage III or IV Melanoma.* N Engl J Med, 2017. **377**(19): p. 1824-1835.
45. Ascierto, P.A., et al., *Nivolumab for Resected Stage III or IV Melanoma at 9 Years.* N Engl J Med, 2025.
46. Eggermont, A.M., et al., *Seven-year analysis of adjuvant pembrolizumab versus placebo in stage III melanoma in the EORTC1325 / KEYNOTE-054 trial.* Eur J Cancer, 2024. **211**: p. 114327.
47. Luke, J.J., et al., *Pembrolizumab Versus Placebo as Adjuvant Therapy in Resected Stage IIB or IIC Melanoma: Final Analysis of Distant Metastasis-Free Survival in the Phase III KEYNOTE-716 Study.* J Clin Oncol, 2024. **42**(14): p. 1619-1624.
48. Kirkwood, J.M., et al., *Adjuvant nivolumab in resected stage IIB/C melanoma: primary results from the randomized, phase 3 CheckMate 76K trial.* Nat Med, 2023. **29**(11): p. 2835-2843.
49. Long, G.V., et al., *1077MO Adjuvant nivolumab v placebo in stage IIB/C melanoma: 3-year results from CheckMate 76K.* Annals of Oncology, 2024. **35**: p. S713-S714.
50. Dummer, R., et al., *Five-Year Analysis of Adjuvant Dabrafenib plus Trametinib in Stage III Melanoma.* N Engl J Med, 2020. **383**(12): p. 1139-1148.
51. Long, G.V., et al., *Final Results for Adjuvant Dabrafenib plus Trametinib in Stage III Melanoma.* N Engl J Med, 2024. **391**(18): p. 1709-1720.
52. Henderson, M.A., et al., *Adjuvant lymph-node field radiotherapy versus observation only in patients with melanoma at high risk of further lymph-node field relapse after lymphadenectomy (ANZMTG 01.02/TROG 02.01): 6-year follow-up of a phase 3, randomised controlled trial.* Lancet Oncol, 2015. **16**(9): p. 1049-1060.
53. Larkin, J., et al., *Combined Nivolumab and Ipilimumab or Monotherapy in Untreated Melanoma.* N Engl J Med, 2015. **373**(1): p. 23-34.

54. Wolchok, J.D., et al., *Final, 10-Year Outcomes with Nivolumab plus Ipilimumab in Advanced Melanoma.* N Engl J Med, 2025. **392**(1): p. 11-22.
55. Chapman, P.B., et al., *Improved survival with vemurafenib in melanoma with BRAF V600E mutation.* N Engl J Med, 2011. **364**(26): p. 2507-16.
56. McArthur, G.A., et al., *Safety and efficacy of vemurafenib in BRAF(V600E) and BRAF(V600K) mutation-positive melanoma (BRIM-3): extended follow-up of a phase 3, randomised, open-label study.* Lancet Oncol, 2014. **15**(3): p. 323-32.
57. Chapman, P.B., et al., *Vemurafenib in patients with BRAFV600 mutation-positive metastatic melanoma: final overall survival results of the randomized BRIM-3 study.* Ann Oncol, 2017. **28**(10): p. 2581-2587.
58. Ascierto, P.A., et al., *5-Year Outcomes with Cobimetinib plus Vemurafenib in BRAFV600 Mutation-Positive Advanced Melanoma: Extended Follow-up of the coBRIM Study.* Clin Cancer Res, 2021. **27**(19): p. 5225-5235.
59. Flaherty, K.T., et al., *Improved survival with MEK inhibition in BRAF-mutated melanoma.* N Engl J Med, 2012. **367**(2): p. 107-14.
60. Hauschild, A., et al., *Dabrafenib in BRAF-mutated metastatic melanoma: a multicentre, open-label, phase 3 randomised controlled trial.* Lancet, 2012. **380**(9839): p. 358-65.
61. Robert, C., et al., *Improved overall survival in melanoma with combined dabrafenib and trametinib.* N Engl J Med, 2015. **372**(1): p. 30-9.
62. Long, G.V., et al., *Dabrafenib plus trametinib versus dabrafenib monotherapy in patients with metastatic BRAF V600E/K-mutant melanoma: long-term survival and safety analysis of a phase 3 study.* Ann Oncol, 2017. **28**(7): p. 1631-1639.
63. Schadendorf, D., et al., *COLUMBUS 7-year update: A randomized, open-label, phase III trial of encorafenib plus binimetinib versus vemurafenib or encorafenib in patients with BRAF V600E/K-mutant melanoma.* Eur J Cancer, 2024. **204**: p. 114073.
64. Patel, P.M., et al., *Extended schedule, escalated dose temozolomide versus dacarbazine in stage IV melanoma: final results of a randomised phase III study (EORTC 18032).* Eur J Cancer, 2011. **47**(10): p. 1476-83.
65. Huncharek, M., J.F. Caubet, and R. McGarry, *Single-agent DTIC versus combination chemotherapy with or without immunotherapy in metastatic melanoma: a meta-analysis of 3273 patients from 20 randomized trials.* Melanoma Res, 2001. **11**(1): p. 75-81.
66. Middleton, M.R., et al., *Randomized phase III study of temozolomide versus dacarbazine in the treatment of patients with advanced metastatic malignant melanoma.* J Clin Oncol, 2000. **18**(1): p. 158-66.
67. Kiebert, G.M., D.L. Jonas, and M.R. Middleton, *Health-related quality of life in patients with advanced metastatic melanoma: results of a randomized phase III study comparing temozolomide with dacarbazine.* Cancer Invest, 2003. **21**(6): p. 821-9.
68. Andtbacka, R.H., et al., *Talimogene Laherparepvec Improves Durable Response Rate in Patients With Advanced Melanoma.* J Clin Oncol, 2015. **33**(25): p. 2780-8.
69. Marty, M., et al., *Electrochemotherapy – An easy, highly effective and safe treatment of cutaneous and subcutaneous metastases: Results of ESOPE (European Standard Operating Procedures of Electrochemotherapy) study.* European Journal of Cancer Supplements, 2006. **4**(11): p. 3-13.
70. Moreno-Ramirez, D., et al., *Isolated limb perfusion for malignant melanoma: systematic review on effectiveness and safety.* Oncologist, 2010. **15**(4): p. 416-27.

71. Lens, M. and J. Schachter, *The Role of Radiotherapy in the Management of Melanoma Brain Metastases: An Overview.* Curr Treat Options Oncol, 2025. **26**(1): p. 36-44.
72. Challis, G.B. and H.J. Stam, *The spontaneous regression of cancer. A review of cases from 1900 to 1987.* Acta Oncol, 1990. **29**(5): p. 545-50.
73. Carlson, R.D., J.C. Flickinger, Jr., and A.E. Snook, *Talkin' Toxins: From Coley's to Modern Cancer Immunotherapy.* Toxins (Basel), 2020. **12**(4).
74. Hanahan, D. and R.A. Weinberg, *The hallmarks of cancer.* Cell, 2000. **100**(1): p. 57-70.
75. Hanahan, D. and R.A. Weinberg, *Hallmarks of cancer: the next generation.* Cell, 2011. **144**(5): p. 646-74.
76. Hanahan, D., *Hallmarks of Cancer: New Dimensions.* Cancer Discov, 2022. **12**(1): p. 31-46.
77. Hanahan, D., *Hallmarks of cancer-Then and now, and beyond.* Cell, 2026.
78. Chen, D.S. and I. Mellman, *Oncology meets immunology: the cancer-immunity cycle.* Immunity, 2013. **39**(1): p. 1-10.
79. Mellman, I., et al., *The cancer-immunity cycle: Indication, genotype, and immunotype.* Immunity, 2023. **56**(10): p. 2188-2205.
80. Liu, C., et al., *Clinical cancer immunotherapy: Current progress and prospects.* Front Immunol, 2022. **13**: p. 961805.
81. Sharma, P., et al., *Immune checkpoint therapy-current perspectives and future directions.* Cell, 2023. **186**(8): p. 1652-1669.
82. Hodi, F.S., et al., *Improved survival with ipilimumab in patients with metastatic melanoma.* N Engl J Med, 2010. **363**(8): p. 711-23.
83. Robert, C., et al., *Ipilimumab plus dacarbazine for previously untreated metastatic melanoma.* N Engl J Med, 2011. **364**(26): p. 2517-26.
84. Schadendorf, D., et al., *Pooled Analysis of Long-Term Survival Data From Phase II and Phase III Trials of Ipilimumab in Unresectable or Metastatic Melanoma.* J Clin Oncol, 2015. **33**(17): p. 1889-94.
85. Abou-Alfa, G.K., et al., *Tremelimumab plus Durvalumab in Unresectable Hepatocellular Carcinoma.* NEJM Evid, 2022. **1**(8): p. EVIDoa2100070.
86. Johnson, M.L., et al., *Durvalumab With or Without Tremelimumab in Combination With Chemotherapy as First-Line Therapy for Metastatic Non-Small-Cell Lung Cancer: The Phase III POSEIDON Study.* J Clin Oncol, 2023. **41**(6): p. 1213-1227.
87. LaFleur, M.W., et al., *Inhibitors of the PD-1 Pathway in Tumor Therapy.* J Immunol, 2018. **200**(2): p. 375-383.
88. Ribas, A., et al., *Pembrolizumab versus investigator-choice chemotherapy for ipilimumab-refractory melanoma (KEYNOTE-002): a randomised, controlled, phase 2 trial.* Lancet Oncol, 2015. **16**(8): p. 908-18.
89. Schachter, J., et al., *Pembrolizumab versus ipilimumab for advanced melanoma: final overall survival results of a multicentre, randomised, open-label phase 3 study (KEYNOTE-006).* Lancet, 2017. **390**(10105): p. 1853-1862.
90. Long, G.V., et al., *Pembrolizumab versus ipilimumab for advanced melanoma: 10-year follow-up of the phase III KEYNOTE-006 study.* Ann Oncol, 2024. **35**(12): p. 1191-1199.
91. Robert, C., et al., *Nivolumab in previously untreated melanoma without BRAF mutation.* N Engl J Med, 2015. **372**(4): p. 320-30.
92. Ascierto, P.A., et al., *Survival Outcomes in Patients With Previously Untreated BRAF Wild-Type Advanced Melanoma Treated With Nivolumab Therapy: Three-Year Follow-up of a Randomized Phase 3 Trial.* JAMA Oncol, 2019. **5**(2): p. 187-194.

93. Weber, J.S., et al., *Nivolumab versus chemotherapy in patients with advanced melanoma who progressed after anti-CTLA-4 treatment (CheckMate 037): a randomised, controlled, open-label, phase 3 trial.* Lancet Oncol, 2015. **16**(4): p. 375-84.
94. Larkin, J., et al., *Overall Survival in Patients With Advanced Melanoma Who Received Nivolumab Versus Investigator's Choice Chemotherapy in CheckMate 037: A Randomized, Controlled, Open-Label Phase III Trial.* J Clin Oncol, 2018. **36**(4): p. 383-390.
95. Hammers, H., *Immunotherapy in kidney cancer: the past, present, and future.* Curr Opin Urol, 2016. **26**(6): p. 543-7.
96. Cancer Resaearch Institute. *Regulatory Approval Timeline of Active immunotherapies*. 2025 20260219]; Available from: https://www.cancerresearch.org/regulatory-approval-timeline-of-active-immunotherapies.
97. Tawbi, H.A., et al., *Relatlimab and Nivolumab versus Nivolumab in Untreated Advanced Melanoma.* N Engl J Med, 2022. **386**(1): p. 24-34.
98. Postow, M.A., R. Sidlow, and M.D. Hellmann, *Immune-Related Adverse Events Associated with Immune Checkpoint Blockade.* N Engl J Med, 2018. **378**(2): p. 158-168.
99. Casagrande, S., et al., *Immune-Related Adverse Events Due to Cancer Immunotherapy: Immune Mechanisms and Clinical Manifestations.* Cancers (Basel), 2024. **16**(7).
100. Hussaini, S., et al., *Association between immune-related side effects and efficacy and benefit of immune checkpoint inhibitors - A systematic review and meta-analysis.* Cancer Treat Rev, 2021. **92**: p. 102134.
101. Petrelli, F., et al., *Immune-related Adverse Events and Survival in Solid Tumors Treated With Immune Checkpoint Inhibitors: A Systematic Review and Meta-Analysis.* J Immunother, 2020. **43**(1): p. 1-7.
102. Eggermont, A.M.M., et al., *Association Between Immune-Related Adverse Events and Recurrence-Free Survival Among Patients With Stage III Melanoma Randomized to Receive Pembrolizumab or Placebo: A Secondary Analysis of a Randomized Clinical Trial.* JAMA Oncol, 2020. **6**(4): p. 519-527.
103. Maher, V.E., et al., *Analysis of the Association Between Adverse Events and Outcome in Patients Receiving a Programmed Death Protein 1 or Programmed Death Ligand 1 Antibody.* J Clin Oncol, 2019. **37**(30): p. 2730-2737.
104. Cortellini, A., et al., *A systematic review on the emerging association between the occurrence of immune-related adverse events and clinical outcomes with checkpoint inhibitors in advanced cancer patients.* Semin Oncol, 2019. **46**(4-5): p. 362-371.
105. Ghisoni, E., et al., *Late-onset and long-lasting immune-related adverse events from immune checkpoint-inhibitors: An overlooked aspect in immunotherapy.* Eur J Cancer, 2021. **149**: p. 153-164.
106. Wang, D.Y., et al., *Fatal Toxic Effects Associated With Immune Checkpoint Inhibitors: A Systematic Review and Meta-analysis.* JAMA Oncol, 2018. **4**(12): p. 1721-1728.
107. Bertrand, A., et al., *Immune related adverse events associated with anti-CTLA-4 antibodies: systematic review and meta-analysis.* BMC Med, 2015. **13**: p. 211.
108. Wang, P.F., et al., *Immune-Related Adverse Events Associated with Anti-PD-1/PD-L1 Treatment for Malignancies: A Meta-Analysis.* Front Pharmacol, 2017. **8**: p. 730.

109. Sun, X., et al., *Immune-related adverse events associated with programmed cell death protein-1 and programmed cell death ligand 1 inhibitors for non-small cell lung cancer: a PRISMA systematic review and meta-analysis.* BMC Cancer, 2019. **19**(1): p. 558.
110. Sonpavde, G.P., et al., *Immune-related adverse events with PD-1 versus PD-L1 inhibitors: a meta-analysis of 8730 patients from clinical trials.* Future Oncol, 2021. **17**(19): p. 2545-2558.
111. Wang, Y., et al., *Treatment-Related Adverse Events of PD-1 and PD-L1 Inhibitors in Clinical Trials: A Systematic Review and Meta-analysis.* JAMA Oncol, 2019. **5**(7): p. 1008-1019.
112. Cathcart-Rake, E.J., et al., *A Population-based Study of Immunotherapy-related Toxicities in Lung Cancer.* Clin Lung Cancer, 2020. **21**(5): p. 421-427.e2.
113. Sabaté Gallego, M., et al., *Incidence and characteristics of adverse drug reactions in a cohort of patients treated with PD-1/PD-L1 inhibitors in real-world practice.* Front Med (Lausanne), 2022. **9**: p. 891179.
114. Miyashita, H., et al., *Incidence and Risk of Colitis With Programmed Death 1 Versus Programmed Death Ligand 1 Inhibitors for the Treatment of Cancer.* J Immunother, 2020. **43**(9): p. 291-298.
115. Lebbé, C., et al., *Evaluation of Two Dosing Regimens for Nivolumab in Combination With Ipilimumab in Patients With Advanced Melanoma: Results From the Phase IIIb/IV CheckMate 511 Trial.* J Clin Oncol, 2019. **37**(11): p. 867-875.
116. Lebbé, C., MeyerN, Mortier L, Marquez-Rodas I, Robert C, Rutkowski P, et al, *Two dosing regimens of nivolumab (NIVO) plus ipilimumab (IPI) for advanced (adv) melanoma: Three-year results of Checkmate 511.* Journal of Clinical Oncology, 2021. **39**.
117. Long, G.V., et al., *Standard-Dose Pembrolizumab Plus Alternate-Dose Ipilimumab in Advanced Melanoma: KEYNOTE-029 Cohort 1C, a Phase 2 Randomized Study of Two Dosing Schedules.* Clin Cancer Res, 2021. **27**(19): p. 5280-5288.
118. Khoja, L., et al., *Tumour- and class-specific patterns of immune-related adverse events of immune checkpoint inhibitors: a systematic review.* Ann Oncol, 2017. **28**(10): p. 2377-2385.
119. Choi, J. and S.Y. Lee, *Clinical Characteristics and Treatment of Immune-Related Adverse Events of Immune Checkpoint Inhibitors.* Immune Netw, 2020. **20**(1): p. e9.
120. Rose, L.M., et al., *Incidence of Skin and Respiratory Immune-Related Adverse Events Correlates With Specific Tumor Types in Patients Treated With Checkpoint Inhibitors.* Front Oncol, 2020. **10**: p. 570752.
121. Kalinich, M., et al., *Prediction of severe immune-related adverse events requiring hospital admission in patients on immune checkpoint inhibitors: study of a population level insurance claims database from the USA.* J Immunother Cancer, 2021. **9**(3).
122. Gougis, P., et al., *Clinical spectrum and evolution of immune-checkpoint inhibitors toxicities over a decade-a worldwide perspective.* EClinicalMedicine, 2024. **70**: p. 102536.
123. Verkhovskaia, S., et al., *Vitiligo-like leukoderma as an indicator of clinical response to immune checkpoint inhibitors in late-stage melanoma patients.* J Cancer Res Clin Oncol, 2022. **148**(9): p. 2529-2538.
124. Teulings, H.E., et al., *Vitiligo-like depigmentation in patients with stage III-IV melanoma receiving immunotherapy and its association with survival: a systematic review and meta-analysis.* J Clin Oncol, 2015. **33**(7): p. 773-81.

125. Thuillier, P., et al., *Thyroid dysfunction induced by immune checkpoint inhibitors is associated with a better progression-free survival and overall survival in non-small cell lung cancer: an original cohort study.* Cancer Immunol Immunother, 2021. **70**(7): p. 2023-2033.
126. Kim, H.I., et al., *Development of thyroid dysfunction is associated with clinical response to PD-1 blockade treatment in patients with advanced non-small cell lung cancer.* Oncoimmunology, 2017. **7**(1): p. e1375642.
127. Martini, D.J., et al., *Immune-Related Adverse Events as Clinical Biomarkers in Patients with Metastatic Renal Cell Carcinoma Treated with Immune Checkpoint Inhibitors.* Oncologist, 2021. **26**(10): p. e1742-e1750.
128. Tang, S.Q., et al., *The Pattern of Time to Onset and Resolution of Immune-Related Adverse Events Caused by Immune Checkpoint Inhibitors in Cancer: A Pooled Analysis of 23 Clinical Trials and 8,436 Patients.* Cancer Res Treat, 2021. **53**(2): p. 339-354.
129. Hassel, J.C., et al., *Combined immune checkpoint blockade (anti-PD-1/anti-CTLA-4): Evaluation and management of adverse drug reactions.* Cancer Treat Rev, 2017. **57**: p. 36-49.
130. Haanen, J., et al., *Management of toxicities from immunotherapy: ESMO Clinical Practice Guideline for diagnosis, treatment and follow-up.* Ann Oncol, 2022. **33**(12): p. 1217-1238.
131. Skribek, M., et al., *Effect of corticosteroids on the outcome of patients with advanced non-small cell lung cancer treated with immune-checkpoint inhibitors.* Eur J Cancer, 2021. **145**: p. 245-254.
132. Petrelli, F., et al., *Association of Steroids use with Survival in Patients Treated with Immune Checkpoint Inhibitors: A Systematic Review and Meta-Analysis.* Cancers (Basel), 2020. **12**(3).
133. Weber, J.S., et al., *Safety Profile of Nivolumab Monotherapy: A Pooled Analysis of Patients With Advanced Melanoma.* J Clin Oncol, 2017. **35**(7): p. 785-792.
134. Bruyère, C.L., et al., *Investigating the Impact of Immune-Related Adverse Events, Glucocorticoid Use and Immunotherapy Interruption on Long-Term Survival Outcomes.* Cancers (Basel), 2021. **13**(10).
135. Cook, S., et al., *Immune-Related Adverse Events and Survival Among Patients With Metastatic NSCLC Treated With Immune Checkpoint Inhibitors.* JAMA Netw Open, 2024. **7**(1): p. e2352302.
136. Bai, X., et al., *Early Use of High-Dose Glucocorticoid for the Management of irAE Is Associated with Poorer Survival in Patients with Advanced Melanoma Treated with Anti-PD-1 Monotherapy.* Clin Cancer Res, 2021. **27**(21): p. 5993-6000.
137. Verheijden, R.J., et al., *Immunosuppression for immune-related adverse events during checkpoint inhibition: an intricate balance.* NPJ Precis Oncol, 2023. **7**(1): p. 41.
138. Verheijden, R.J., et al., *Corticosteroids for Immune-Related Adverse Events and Checkpoint Inhibitor Efficacy: Analysis of Six Clinical Trials.* J Clin Oncol, 2024. **42**(31): p. 3713-3724.
139. Das, S. and D.B. Johnson, *Immune-related adverse events and anti-tumor efficacy of immune checkpoint inhibitors.* J Immunother Cancer, 2019. **7**(1): p. 306.
140. Shui, L., et al., *Gut Microbiome as a Potential Factor for Modulating Resistance to Cancer Immunotherapy.* Front Immunol, 2019. **10**: p. 2989.
141. Davar, D., et al., *Fecal microbiota transplant overcomes resistance to anti-PD-1 therapy in melanoma patients.* Science, 2021. **371**(6529): p. 595-602.

142. Zimmermann, P. and N. Curtis, *The effect of antibiotics on the composition of the intestinal microbiota - a systematic review.* J Infect, 2019. **79**(6): p. 471-489.
143. Crespin, A., et al., *A systematic review and meta-analysis evaluating the impact of antibiotic use on the clinical outcomes of cancer patients treated with immune checkpoint inhibitors.* Front Oncol, 2023. **13**: p. 1075593.
144. Huang, X.Z., et al., *Antibiotic use and the efficacy of immune checkpoint inhibitors in cancer patients: a pooled analysis of 2740 cancer patients.* Oncoimmunology, 2019. **8**(12): p. e1665973.
145. Tsikala-Vafea, M. and D. Farmakiotis, *Corrigendum to 'Worse clinical outcomes in patients with cancer treated with immune checkpoint inhibitors: A systematic review and meta-analysis'.* Int J Infect Dis, 2021. **113**: p. 34-35.
146. Vranic, S. and Z. Gatalica, *PD-L1 testing by immunohistochemistry in immuno-oncology.* Biomol Biomed, 2023. **23**(1): p. 15-25.
147. Robert, M.E., et al., *High Interobserver Variability Among Pathologists Using Combined Positive Score to Evaluate PD-L1 Expression in Gastric, Gastroesophageal Junction, and Esophageal Adenocarcinoma.* Mod Pathol, 2023. **36**(5): p. 100154.
148. Bastiaannet, E., et al., *Immunotherapy and targeted therapies in older patients with advanced melanoma; Young International Society of Geriatric Oncology review paper.* J Geriatr Oncol, 2019. **10**(3): p. 389-397.
149. Ksienski, D., et al., *Association of age with differences in immune related adverse events and survival of patients with advanced nonsmall cell lung cancer receiving pembrolizumab or nivolumab.* J Geriatr Oncol, 2020. **11**(5): p. 807-813.
150. Ksienski, D., et al., *Immune related adverse events and treatment discontinuation in patients older and younger than 75 years with advanced melanoma receiving nivolumab or pembrolizumab.* J Geriatr Oncol, 2022. **13**(2): p. 220-227.
151. Basak, E.A., et al., *Associations between patient and disease characteristics and severe adverse events during immune checkpoint inhibitor treatment: An observational study.* Eur J Cancer, 2022. **174**: p. 113-120.
152. Baldini, C., et al., *Impact of aging on immune-related adverse events generated by anti-programmed death (ligand)PD-(L)1 therapies.* Eur J Cancer, 2020. **129**: p. 71-79.
153. Krishnan, M., et al., *Impact of Performance Status on Response and Survival Among Patients Receiving Checkpoint Inhibitors for Advanced Solid Tumors.* JCO Oncol Pract, 2022. **18**(1): p. e175-e182.
154. Tomasik, B., et al., *Effectiveness and safety of immunotherapy in NSCLC patients with ECOG PS score ≥2 - Systematic review and meta-analysis.* Lung Cancer, 2021. **158**: p. 97-106.
155. Guzman-Prado, Y., J. Ben Shimol, and O. Samson, *Body mass index and immune-related adverse events in patients on immune checkpoint inhibitor therapies: a systematic review and meta-analysis.* Cancer Immunol Immunother, 2021. **70**(1): p. 89-100.
156. Jing, Y., et al., *Association Between Sex and Immune-Related Adverse Events During Immune Checkpoint Inhibitor Therapy.* J Natl Cancer Inst, 2021. **113**(10): p. 1396-1404.
157. Vitale, E., et al., *Sex differences in adverse events among cancer patients receiving immune checkpoint inhibitors: the MOUSEION-07 systematic review and meta-analysis.* Sci Rep, 2024. **14**(1): p. 28309.

158. Fang, Q., et al., *Predictors of severity and onset timing of immune-related adverse events in cancer patients receiving immune checkpoint inhibitors: a retrospective analysis.* Front Immunol, 2025. **16**: p. 1508512.
159. Triggianese, P., et al., *Immune checkpoint inhibitors-induced autoimmunity: The impact of gender.* Autoimmun Rev, 2020. **19**(8): p. 102590.
160. Lal, J.C., et al., *A retrospective analysis of cardiovascular adverse events associated with immune checkpoint inhibitors.* Cardiooncology, 2021. **7**(1): p. 19.
161. Lopez-Olivo, M.A., et al., *A systematic review and meta-analysis of observational studies and uncontrolled trials reporting on the use of checkpoint blockers in patients with cancer and pre-existing autoimmune disease.* Eur J Cancer, 2024. **207**: p. 114148.
162. Le, J., et al., *Immune checkpoint inhibitors in cancer patients with autoimmune disease: Safety and efficacy.* Hum Vaccin Immunother, 2025. **21**(1): p. 2458948.
163. Les, I., et al., *Predictive Biomarkers for Checkpoint Inhibitor Immune-Related Adverse Events.* Cancers (Basel), 2023. **15**(5).
164. Chennamadhavuni, A., et al., *Risk Factors and Biomarkers for Immune-Related Adverse Events: A Practical Guide to Identifying High-Risk Patients and Rechallenging Immune Checkpoint Inhibitors.* Front Immunol, 2022. **13**: p. 779691.
165. McDermott, D., et al., *Durable benefit and the potential for long-term survival with immunotherapy in advanced melanoma.* Cancer Treat Rev, 2014. **40**(9): p. 1056-64.
166. van Not, O.J., et al., *Long-Term Survival in Patients With Advanced Melanoma.* JAMA Netw Open, 2024. **7**(8): p. e2426641.
167. Ruste, V., et al., *The determinants of very severe immune-related adverse events associated with immune checkpoint inhibitors: A prospective study of the French REISAMIC registry.* Eur J Cancer, 2021. **158**: p. 217-224.
168. Facon, T., et al., *A simplified frailty scale predicts outcomes in transplant-ineligible patients with newly diagnosed multiple myeloma treated in the FIRST (MM-020) trial.* Leukemia, 2020. **34**(1): p. 224-233.
169. Stege, C.A.M., et al., *Validation of the FIRST simplified frailty scale using the ECOG performance status instead of patient-reported activities.* Leukemia, 2020. **34**(7): p. 1964-1966.
170. Mian, H.S., et al., *External validation of the FIRST trial's simplified frailty score in a population-based cohort.* Leukemia, 2021. **35**(6): p. 1823-1827.
171. Zeng, X., et al., *Effect of Comorbidity on Outcomes of Patients with Advanced Non-Small Cell Lung Cancer Undergoing Anti-PD1 Immunotherapy.* Med Sci Monit, 2020. **26**: p. e922576.
172. von Itzstein, M.S., et al., *Immunotherapy Use in Patients With Lung Cancer and Comorbidities.* Cancer J, 2020. **26**(6): p. 525-536.
173. Cortellini, A., et al., *Clinical Outcomes of Patients with Advanced Cancer and Pre-Existing Autoimmune Diseases Treated with Anti-Programmed Death-1 Immunotherapy: A Real-World Transverse Study.* Oncologist, 2019. **24**(6): p. e327-e337.
174. Leonardi, G.C., et al., *Safety of Programmed Death-1 Pathway Inhibitors Among Patients With Non-Small-Cell Lung Cancer and Preexisting Autoimmune Disorders.* J Clin Oncol, 2018. **36**(19): p. 1905-1912.
175. Shankar, B., et al., *Multisystem Immune-Related Adverse Events Associated With Immune Checkpoint Inhibitors for Treatment of Non-Small Cell Lung Cancer.* JAMA Oncol, 2020. **6**(12): p. 1952-1956.

176. Cortellini, A., et al., *Immune-related Adverse Events of Pembrolizumab in a Large Real-world Cohort of Patients With NSCLC With a PD-L1 Expression ≥ 50% and Their Relationship With Clinical Outcomes.* Clin Lung Cancer, 2020. **21**(6): p. 498-508.e2.
177. Ricciuti, B., et al., *Impact of immune-related adverse events on survival in patients with advanced non-small cell lung cancer treated with nivolumab: long-term outcomes from a multi-institutional analysis.* J Cancer Res Clin Oncol, 2019. **145**(2): p. 479-485.
178. Shimozaki, K., et al., *Multiple Immune-Related Adverse Events and Anti-Tumor Efficacy: Real-World Data on Various Solid Tumors.* Cancer Manag Res, 2020. **12**: p. 4585-4593.
179. Hata, H., et al., *A Retrospective Cohort Study of Multiple Immune-Related Adverse Events and Clinical Outcomes Among Patients With Cancer Receiving Immune Checkpoint Inhibitors.* Cancer Control, 2022. **29**: p. 10732748221130576.
180. Kichenadasse, G., et al., *Multiorgan Immune-Related Adverse Events During Treatment With Atezolizumab.* J Natl Compr Canc Netw, 2020. **18**(9): p. 1191-1199.
181. Cortellini, A., et al., *Correlations Between the Immune-related Adverse Events Spectrum and Efficacy of Anti-PD1 Immunotherapy in NSCLC Patients.* Clin Lung Cancer, 2019. **20**(4): p. 237-247.e1.
182. Jones, M. and R. Fowler, *Immortal time bias in observational studies of time-to-event outcomes.* J Crit Care, 2016. **36**: p. 195-199.
183. Duan, J., et al., *Use of Immunotherapy With Programmed Cell Death 1 vs Programmed Cell Death Ligand 1 Inhibitors in Patients With Cancer: A Systematic Review and Meta-analysis.* JAMA Oncol, 2020. **6**(3): p. 375-384.
184. Pillai, R.N., et al., *Comparison of the toxicity profile of PD-1 versus PD-L1 inhibitors in non-small cell lung cancer: A systematic analysis of the literature.* Cancer, 2018. **124**(2): p. 271-277.
185. Gu, Y., et al., *Different patterns of treatment-related adverse events of programmed cell death-1 and its ligand-1 inhibitors in different cancer types: A meta-analysis and systemic review of clinical trials.* Asia Pac J Clin Oncol, 2020. **16**(5): p. e160-e178.
186. Network, N.C.C. *NCCN Guidelines Melanoma: Cutaneus, 2024*. Version 3 2024.
187. Swetter, S.M., et al., *Guidelines of care for the management of primary cutaneous melanoma.* J Am Acad Dermatol, 2019. **80**(1): p. 208-250.
188. Garbe, C., et al., *European consensus-based interdisciplinary guideline for melanoma. Part 1: Diagnostics: Update 2022.* Eur J Cancer, 2022. **170**: p. 236-255.
189. Helvind, N.M., et al., *Routine PET-CT scans provide early and accurate recurrence detection in asymptomatic stage IIB-III melanoma patients.* Eur J Surg Oncol, 2021. **47**(12): p. 3020-3027.
190. Lewin, J., et al., *Surveillance imaging with FDG-PET/CT in the post-operative follow-up of stage 3 melanoma.* Ann Oncol, 2018. **29**(7): p. 1569-1574.
191. Lim, K.H.J., et al., *Contemporary outcomes from the use of regular imaging to detect relapse in high-risk cutaneous melanoma.* ESMO Open, 2018. **3**(2): p. e000317.
192. Rueth, N.M., et al., *Is surveillance imaging effective for detecting surgically treatable recurrences in patients with melanoma? A comparative analysis of stage-specific surveillance strategies.* Ann Surg, 2014. **259**(6): p. 1215-22.

193. Koskivuo, I., et al., *Whole body PET/CT in the follow-up of asymptomatic patients with stage IIB-IIIB cutaneous melanoma().* Acta Oncol, 2016. **55**(11): p. 1355-1359.
194. Dieng, M., et al., *The Impact of Surveillance Imaging Frequency on the Detection of Distant Disease for Patients with Resected Stage III Melanoma.* Ann Surg Oncol, 2022. **29**(5): p. 2871-2881.
195. Liang, Y., Y. Shao, and W. Gu, *Role of interleukin-6 in resistance to tumor therapy.* Discov Oncol, 2025. **16**(1): p. 1791.
196. al, A.B.e., *Phase II trial of weekly or bi-weekly tocilizumab with ipilimumab and nivolumab in advanced melanoma: Clinical outcomes and biomarker analysis.*, in *ASCO*. 2024: Chicago, Illinois, USA.
197. Weber, J.S., et al., *Interleukin-6 receptor blockade with tocilizumab to reduce immune-related toxicity with ipilimumab and nivolumab in metastatic melanoma.* Journal of Clinical Oncology, 2024. **42**(16_suppl): p. 9538-9538.
198. De Simoni, E., et al., *Circulating tumor DNA-based assessment of molecular residual disease in non-metastatic melanoma.* Cancer Treat Rev, 2024. **129**: p. 102788.

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