REVIEW ARTICLE

A Comprehensive Review on Treatment Modalities of Malignant Melanoma

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Abstract

Melanoma is an aggressive tumor that exhibits an increased occurrence and poor prognosis if metastasized. Early diagnosis and effective management are the mainstays for melanoma management. Effective management includes the selection of an effective treatment plan, supportive care, and training of patients for early diagnosis; various diagnostic options are available *i.e.* biopsy assessment, imaging procedures, and different biomarkers like liquid biopsy, various proteins, and polymorphism. As for the biomarkers, assessment of circulating tumor cells, microRNAs (miRNAs), and cell-free DNAs and RNAs are effectively used in the diagnosis of melanoma cases; the dysfunction of these molecules gives rise to the pathogenesis of melanoma. Many treatment modalities could be used potentially for melanoma management *i.e.* radiation therapy, surgery, systemic therapy, immunotherapy, and antibodies therapy. Proper care and training for melanoma patients contribute to enhance monitoring of the cases, in response to different treatment modalities. In this review, we have summarized details about malignant melanoma, keeping in view its epidemiology, classification, diagnosis, and various treatment modalities.

Keywords: *malignant melanoma, review on malignant melanoma, biopsy in malignant melanoma, malignant melanoma diagnosis.*

INTRODUCTION

Cancer is regarded as among the principal causes of mortality globally as per WHO report [1]. It is expected that the number of persons diagnosed with cancer will be doubled in the next two decades [2]. Cancer mortality rate can be minimized if it is detected earlier and managed rationally [3]. Research advancements to develop early diagnostic approaches are the principal concern of the investigators [4]. Due to increased occurrence and poor prognosis, much attention has been given to determining melanoma's etiology and pathogenesis. The environmental and genetic causes seem to play a significant part in cancer development *i.e.* sun exposure, presence of atypical nevi, phenotype of skin coloration, and family and individuals' histories of cancer [5].

Several researchers have established that the molecular paths are operational in melanoma growth. It was revealed that genomic alterations in DNA copy quantity along with an examination of specific somatic variations might be used for the discernment of discrete subcategories of melanoma having 70% validity [6]. Such statistics provide novel prognostic and diagnostic potentials and emphasize that different melanomas would progress due to the dysfunction of diverse genetic pathways. The enhanced knowledge will offer better staging and subtype categorization, and an improved management plan for melanoma treatment [6].

Common techniques of gene sequencing together with comparative genomic hybridization and mutation investigation have recognized numerous significant cell-signaling pathways in melanoma. Genetic modifications in CDKN2A are found in approximately 50% of all cutaneous melanomas; CDKN2A is a locus on chromosome 9p21 expressing two tumor-suppressor proteins *i.e.* p16 (CDK4 suppressor) and p14ARF [6]. There are mutations seen in BRAF in about 60% and NRAS in about 20% of melanomas. The BRAF and NRAS proteins are implicated in the mitogen-activated protein kinase (MAPK) pathway modifying genes transcription and controlling cell proliferation and existence. Furthermore, upregulation of the phosphatidylinositol-4,5-bisphosphate 3-kinase (PI3K) pathway gives rise to resistance of apoptosis in melanoma [6].

METHODOLOGY

The literature was electronically searched on PubMed. MeSH keywords applied were: malignant melanoma review, epidemiology of malignant melanoma, classification of malignant melanoma, immune checkpoint inhibitors in malignant melanoma, and current therapies in malignant melanoma. The review articles available in the English language and

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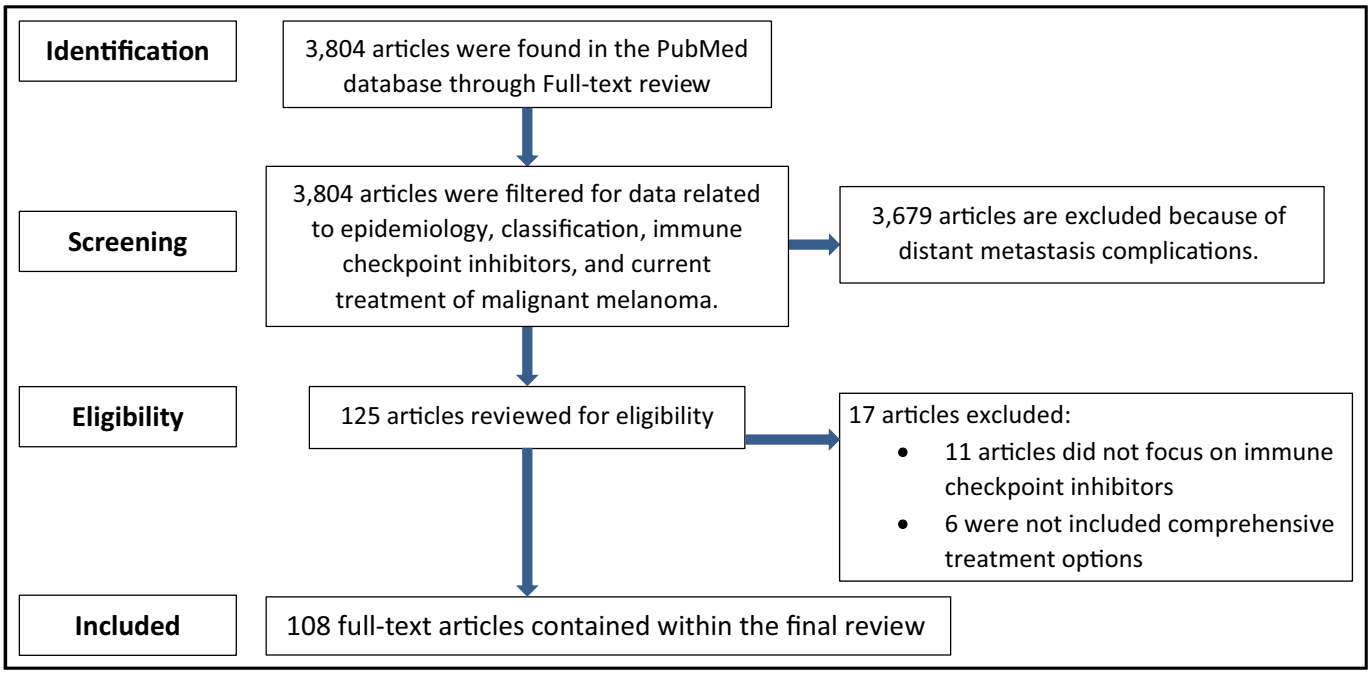

Fig. (1): Flow diagram for article selection.

presenting a generalized overview of the ailment were carefully chosen. Those articles were excluded which encompassed distant metastasis complications. All references were downloaded to EndNote X8 with the removal of duplicates

RESULTS

By the literature search on PubMed, 125 articles were studied out of which 100 are incorporated in the review article (**Fig. 1**).

Epidemiology

Melanoma ascends when there is a carcinogenic modification of melanocytes; the melanocytes are the cells that produce melanin and melanin is a photo-protective pigment [7]. Melanoma can arise from melanocytes in various body parts but most frequently ascends in the skin in case of UV damage [8]. Melanoma is considered the 5th most common malignancy type in grown-ups and is the most lethal type of skin malignancy [9].

It is very alarming that the younger population is affected by malignant melanoma; the median age at identification is just 57 years for cases of cutaneous melanoma as compared to other malignancies *i.e.* the median age for colon cancer is 68 years, for lung cancer it is 70 years and 71 years for prostate cancer [10-12]. Several socioeconomic factors must be considered regarding melanoma oncogenesis in the younger population. The research estimated the overall cost of melanoma treatment to be more than 44,000 Australian dollars for each malignancy; one more study revealed that the diagnosis of melanoma was related to substantial modifications in lifestyle and problems in dealing with the diagnosis. It was seen that women of age group 20-24 years were more expected to be detected with cancerous melanoma than men (4:10), yet, with an increase in age (more than 65 years), men are more susceptible to get melanoma (17:10) [13, 14]. Males diagnosed with cutaneous melanoma exhibit inferior clinical results as compared to females, with greater disease relapse, progression, and death. In general, this may be due to the possibility that males are less probably self-examine for doubtful wounds, more probably defer presentation, and eventually are diagnosed with the disease at more advanced stages [15, 16].

There has been a surge in the occurrence of melanoma worldwide [17, 18], with an estimated 57,220 males and 39,260 females (total of 96,480 adults) detected with melanoma in 2019 in the US; these account for 5.5% of overall incidence, resulting in 7,230 demises *i.e.* 1.2% of all malignancy mortalities [7]. The annual diagnosis is 132,000 cases [19]. A US report by The American Cancer Organization in 2019 estimated that 192,310 individuals were identified with melanoma [4].

Melanoma has been gradually rising like other cancers for more than 3 decades. As compared to the US and Australian people, there has been evidence of a lower incidence of cutaneous melanoma in Europeans though, there are contradictions amid states for melanoma occurrence. The geographical position of southern European states like Greece and Cyprus favors 10%

less incidence of disease in comparison to Denmark, Switzerland, and Norway. According to Forsea *et al.*, as latitude rises through mainland Europe, the occurrence of cutaneous melanoma also increases. There is the highest occurrence of melanoma in equatorial areas and it declines further north or south of the earth's equator, apparently associated with the time of solar radiation in said areas [15, 20]. It is to be noted that with an earlier diagnosis of melanoma, a small surgery might increase recovery chances [21].

Classification of Malignant Melanoma

TNM staging is used to describe melanoma cases: stage I–II is a local ailment, stage III is a node-positive ailment and stage IV is a metastatic illness as per the 8th Edition of American Joint Commission on Cancer (AJCC) [22]. The main clinicopathological characteristics for allocating a phase and/or measuring relapse chances include tumor width (Breslow depth), mitotic rate, presence or absence of ulceration, microsatellites and in-transit wounds, extent of lymph node involvement and existence or lack of distant metastasis. Cutaneous melanomas are mostly localized when identified initially; they are surgically excised with sufficient margins effectively [23].

The malignant cells can also be categorized as benign, pre-malignant, and malignant, based on the severity. Skin lesions like tags or moles are benign and are not cancerous while premalignant are lesions that contain malignant cells and malignant lesions are the ones requiring speedy management due to the high quantity of malignant cells [24].

There are conventional diagnostic techniques available that are costly and also depend on the expertise of a dermatologist and a well-equipped setting. Improved accuracy and efficacy can be achieved through recent computerized systems. To detect and classify melanoma accurately, several measures have to be considered *i.e.* feature extraction and selection, abnormality, contrast distending and origin change. Normally, digital dermoscopy includes 4 important image-processing phases: pre-processing, segmentation, feature extraction and reduction, and wound categorization [25]. Improvement has been reported in the detection (50%) and absolute accuracy from 75% to 84% through dermoscopic imaging [22]. To reduce both inter and intra-observer variability, an alternative can be a computer-based classification of benign and malignant melanoma [24].

Melanoma classification, employing different datasets has been presented by several investigators [24]. Two algorithms for melanoma segmentation were proposed by Azawi *et al.* over dermoscopic images. The adaptive automatic thresholding method is one of the applied algorithms that attained advanced outcomes and warrants aid in the segmentation of skin wounds [26]. A multi-parameterized synthetic neural setup reliant on reachable individual health information was generated and endorsed by David Rofman *et al.* for timely diagnosis of NMSC (Nonmelanoma skin cancer) with greater sensitivity and specificity, even in absenteeism of family history and identified contact of UV radiation [27].

A computer-aided diagnosis (CAD) method was proposed by Zakeri *et al.* for improvement of the diagnostic capacity of the conventionally used ABCD (asymmetry, border irregularity, color, and diameter) assessment. PDT (photodynamic therapy) features stood promising as per experimental readings which when combined with the conventionally used ABCD features, are fit for refining the classification performance of the colored skin wounds [28]. A system initiating with pre-processing methods, then segmenting the wound region by fusing of new even distribution segmentation with active contour method was suggested by Muhammad Nasir *et al.* The outcomes were presented on diverse features combined with varied feature selection techniques and classifiers; results demonstrated that the SVM (Support Vector Machine) assists in greater results on entropy-built features selection [29]. For identifying the cancer of the skin, Victor *et al.* suggested the arrangement of the regulated watershed and active contour model. The feature extraction was applied by use of elementary statistical techniques and the Grey Level Co-Occurrence Matrix (GLCM) with the Support Vector Machine (SVM) for categorization [30]. For the division of melanoma over dermoscopic data of 200 cases, Eltayef *et al.* recommended the combination of fuzzy c-means and Markov random field [31]. For early identification of skin cancers like melanoma, Rundo *et al.* suggested using the neural network [32].

Malignant Melanoma Variations

Malignant melanoma cases comprise cytomorphological structures, architectural arrangements, and stromal modifications. Melanomas may imitate sarcomas, carcinomas, lymphomas, benign stromal growths, plasmacytomas, and germ-cell neoplasms. Melanomas may comprise sizable pleomorphic cells, minor cells, and spindle cells; they may encompass clear, signet-ring, rhabdoid, pseudolipoblastic, plasmacytoid, or balloon cells. Cytoplasm may have different existences and phagocytosed substances. Biro multi-nucleation, inclusions, grooving lobation, and angulation can be seen in the nuclei. Architectural arrangements may

display various patterns, *i.e.* fasciculation, nesting, pseudoglandular / pseudopapillary /pseudofollicular, pseudorosetting, *etc.* The stromal blood vessels display a haemangio-pericytomatous appearance, glomeruloid blood vessel proliferation, and perivascular hyalinization. The melanomas are S100 protein, NKIC3, HMB-45, Melan-A, and tyrosinase positive typically but some may display an unusual immune phenotype and express cytokeratins, desmin, smooth muscle actin, KP1 (CD68), CEA, EMA and VS38 [33].

Diagnosis

Melanoma displays a poor prognosis and is usually diagnosed in the advanced and metastatic stages. Patients with melanoma hence exhibit a tough response to the existing therapeutic modalities [34, 35]. Therefore, melanoma diagnosis in the initial stages is of prime importance. A variety of methodologies like imaging techniques and biomarkers (such as serum proteins, cfDNAs, cfRNAs, miRNAs, CTCs, exosomes, and polymorphism) may perhaps be employed for identifying and monitoring melanoma cases [6].

Imaging Procedures

MelaFind and SIA scope (Spectrophotometric Intracutaneous Analysis) is used to envisage wounds and provide material for clinicians to decide on the need for a biopsy. These devices utilize visible and near-infrared light (~400 nm to ~1000 nm). A completely automatic diagnostic system is MelaFind, established in 2010, which utilizes light to envisage skin wounds ~2.5 mm deep. It offers data on cells' morphologic disarrangement in a wound thereby helping clinicians to rule out the need for lesion biopsy for melanoma [36]. Various types of research support the usage of MelaFind results in making biopsy decisions more accurately [37]. Research in 2017 was performed with MelaFind in which 160 expert dermatologists examined 25 melanomas and 25 benign nevi skin wound examinations. The investigators instituted analyses with MelaFind augmented biopsy sensitivity after clinical assessment only (from 76% to 92%); augmented specificity (from 52% to 79%) and biopsy precision also (from 64% to 86%) in general. Yet, patients' determent from using MelaFind is observed as they have to bear its expenses because several insurance corporations do not pay back for MelaFind use as they think it is an experimental procedure. The multi-spectral imaging technology may be used in routine clinical practice as it turns out to be more advanced and provides cost-effectiveness through more focused biopsy practice [38].

SIA scope (developed in 2002) can measure melanin, collagen, and blood. This device demonstrates whether melanin is limited to the epidermal layer and envisages the wound's vascular network and colorant composition [39]. Earlier forms of this device had some issues: sensitivity of 82.7% and specificity of 80.1%, which is like the sensitivity and specificity of dermatoscopy executed by dermato-pathologists. Dermatoscopy is the optical investigation of a colored wound with a handheld magnifier. Though certain clinicians questioned whether SIA scope offers sufficient benefit to permit its users to detect and diagnose melanoma yet, it may be beneficial to improve the detection of pigmented wounds by those clinicians who practice in rural environments lacking specialized dermatology care [38].

Tumor Markers

Dermoscopy is among the most prevalent imaging procedures. It amplifies the skin wound surface and its structure becomes further detectable for assessment to the dermatologist. This procedure is based entirely on the physician's optical perception and experience, thereby encouraging the researchers to establish new procedures to visualize and diagnose melanoma. The CAD system helps to diagnose malignant melanoma and offers a convenient setting for inexperienced dermatologists [1]. The use of CAD diagnostic tools can be another option to diagnose malignant melanoma [40].

Many biomarkers have been used to diagnose and manage melanoma. A range of tumor markers (Melan-A, CSPG4, HMB-45, and S-100) might be employed as diagnostic biomarkers to detect and monitor melanoma cases. To detect and classify various melanoma tumors, one of the significant diagnostic methods is the application of immune-histochemical staining for tumor markers [41].

Melan-A is a key tumor marker identified by T-cells-1 (MArT-1); it is also a melanocyte differentiation antigen that could be expressed in the melanoma cells' cytoplasm and retinal pigmented epithelium [6, 42]. This is a membrane protein found in the endoplasmic reticulum, melanosomes, and the trans-Golgi network. In research by Suchak *et al.* [43], Melan-A was investigated as a diagnostic biomarker to detect 120 melanoma cases. Immunohistochemical investigation of Melan-A exhibited that this protein may be employed for the timely detection of lentigo maligna cases [43, 44].

HMB-45 (PMel, Pmel17, gp100, or SILV) is a glycoprotein and is a melanoma tumor marker. It exists as a 100 kDa type I transmembrane, noticed in the pigment cells surface of eyes and skin. It has been established that this gene's mutation may be linked with many kinds of melanoma [45].

Chondroitin sulfate proteoglycan 4 (CSPG4) is a tumor marker with a substantial molecular weight melanoma-related antigen (HMw–MAA). Melanoma chondroitin sulfate proteoglycan is recognized as a membrane-bound proteoglycan, expressed in melanocytes, pericytes, and endothelial cells. It is observed that CSPG4 may prompt cell adhesion, growth, and motility in numerous cells. Therefore, CSPG4 shows a vital role in invasion and metastasis [6, 46].

The use of neoplastic tissues is the gold standard to diagnose melanoma. Lately, it was seen that cancer-derived substances, in blood circulation, may perhaps be employed as a useful alternative to overcome certain drawbacks linked with biopsy assessment. A liquid biopsy is executed for the diagnosis of cancer-derived substances taken in blood samples. Several bases of tumor substance can be examined by liquid biopsy together with cell-free or complexed nucleic acids like circulating cell-free DNA (cfDNA), circulating tumor DNA (ctDNA), cell-free RNA (cfRNA), circulating miRNAs and circulating tumor cells [47-49]. cfDNAs are short DNA fragments and are derivatives of apoptotic and necrotic tumor cells and even regular cells can discharge them in the circulating blood [50]. cfDNAs may perhaps be spotted in further body fluids like urine, saliva, and cerebrospinal fluids. The cfDNAs were investigated in 2018 for melanoma cases by Valpione *et al.* as prognostic biomarkers and the results revealed that cfDNA levels (cut-off value = 89 pg/ml), were very much related to the death liability and OS. The cases with cfDNA (89 pg/ml) had petite overall survival (OS) than the others. An association was proposed based on these outcomes amid cfDNA levels and therapy-related amendments in tumor burden [51]. The CTCs (circulating tumour cells) are viable non-hematological cells with malignancy characteristics. They are important constituents of liquid biopsies which can be separated from blood. CTCs are released as single cells or clusters into the bloodstream during metastasis. Therefore, CTCs might be considered as effective biomarkers in melanoma management [6].

Malignant cells have been reported to release many cfRNAs alone or by exosomes into the circulating blood *i.e.* small nucleolar RNAs, microRNAs, PIWI-interacting RNAs, and long noncoding RNAs [50]. Generally, there is much evidence to confirm the employment of liquid biopsies as a potent source for the recognition of different stages of melanoma [52]. Novel biomarkers like microRNAs (miRNAs) have been used potentially to diagnose and monitor cases of several diseases including cancer [53-56]. MiRNAs are tiny and noncoding RNAs, that act as epigenetic regulators [57-59]. These RNAs can control several biological processes *e.g.* angiogenesis, growth, and differentiation [60]. Hence, different miRNAs could be employed as diagnostic, prognostic, and therapeutic biomarkers through their expression levels for several malignancies, for example, melanoma [61, 62]. There is evidence that many miRNAs like miR-148, miR-155, miR-182, miR-200c, miR-211, miR-214, miR-221, and miR-222 could influence the origination and growth of melanoma by targeting different melanoma-related genes (*i.e.* NRAS, microphthalmia-associated transcription factor, receptor tyrosine kinase c-KIT or AP-2 transcription factor). So, these can be employed for diagnosis, prognosis and as therapeutic biomarkers to monitor melanoma cases [63].

With the use of Dermoscopy, a certified dermatologist can accomplish an accuracy: 65% to 75% on an average [4]. Besides, the accuracy can be enhanced by the use of a camera having distinctively high-resolution and a magnifying lens for capturing dermoscopic images for optical examination of suspicious cases. An estimated 50% accuracy is enhanced with this technical provision to diagnose skin malignancy [1, 64]. Automated detection of melanoma can support clinicians routinely through rapid and cost-effective approaches to life-saving diagnoses [65].

The above-mentioned problems and complications accentuate the machine learning populace to focus principally on melanoma categorization [66]. Machine learning gears statistical algorithms for learning that initially sequence the data and then test it [67]. Earlier than 2016, the main focus was on the typical machine learning workflow which involved pre-processing, division, extraction of characteristics, and classification [68]; certain expertise was essentially necessary for the extraction of characteristics from oncogenic pictures. A bad division can reduce the classification accuracy due to poor characteristics selection [64]. A transition occurred in 2016 for skin lesion classification procedures, as displayed at the International Symposium on Biomedical Imaging (ISBI). The researchers employed a procedure of deep learning: convolution neural networks (CNNs) instead of applying traditional machine learning algorithms [69].

Some procedures for skin malignancy detection by the use of images were reviewed by Zilong *et al.* [70]. This research not only supported melanoma identification about it also offered indications about various malignancies using images for their detection. Furthermore, Sultana *et al.* [71] provided a review of a small number of deep-learning approaches and discussed certain standard datasets only. Brinker *et al.* conducted

a systematic literature study on skin lesion classification through CNNs. They gave a summary of various deep learning procedures; the main drawback was that the literature lacked dataset information [72].

Recent Therapies for Malignant Melanoma

The primary phase melanoma is commonly treated with surgery but advanced stages of melanoma are very difficult to treat due to the ineffectiveness of conventional certain malignancy treatments like chemotherapy. In recent times, new immune therapy and targeted treatment have been developed that display more favorable outcomes in the treatment of advanced melanoma [73]. Identification of melanoma susceptibility genes has led to new therapeutic approaches with the development of numerous small molecule inhibitors targeting specific proteins implicated in melanoma pathogenesis. Several Phase-3 clinical trials supported that vemurafenib (BRAF inhibitor) embraces V600E mutation and could extend the survival of advanced melanoma patients [74, 75].

Trametinib is downstream of BRAF in the MAP kinase pathway; it is a tiny molecule inhibitor of MEK1 and MEK2. In a Phase-3 clinical trial, Trametinib showed more significant results in comparison to vemurafenib in the extension of progression-free survival and OS rates [76-79]. Nanotechnology-based methods have also strikingly emerged in melanoma treatment *i.e.* human albumin, dendrimers, liposomes, polymersomes, and carbon-based nanoparticles [6, 7, 79].

Surgical Resection

The main therapy for localized melanoma is the elimination of the neoplasm and adjacent healthy tissue by surgery. Patients with tumors size > 0.8 mm thick or thinner but with ulceration (stage pT1b or more) undergo sentinel lymph node biopsy [80]. If the sentinel lymph nodes contain melanoma cells, the residual lymph nodes in the region are then eliminated occasionally. Metastatic neoplasms can be eliminated surgically in some cases, but surgery is not meant for cure in such cases and will necessitate other therapeutic modalities too [38, 77].

Chemotherapy

Surgery alone is not curative in metastatic cases and drug treatments must be included. In recent times, chemotherapy is the only therapeutic choice for metastatic melanoma cases. FDA-approved dacarbazine is the only chemotherapeutic agent for melanoma treatment [81-83]. Dacarbazine response is limited at its best, offering median survival from 5 to 11 months and a 27% survival rate for a year. Yet, it stays as the standard of care for metastatic melanoma [38].

Targeted Treatments for Melanoma

Numerous targeted treatments have been established against molecular imprecisions existent in melanoma *i.e.* the BRAF inhibitors, vemurafenib, and dabrafenib; in 2011 and 2013, these were official therapy for metastatic and unresectable BRAF-mutated melanomas respectively. Although these agents are greatly beneficial for some partial cases with BRAF mutated melanomas, the affected individuals mostly progress towards secondary resistance in quite a short period. Certain mechanisms responsible for this secondary resistance have been identified [84, 85]. Chemotherapy is not effective in malignant melanoma and existing therapy for tumors with the most common BRAFV600E mutation comprises BRAF inhibitors either alone or combined with MAPK pathway inhibitors. Various chemotherapeutic agents impose DNA damage in cells of malignant melanoma, which is revamped well, related to activation of the ATM-dependent DDR (DNA damage response) equipment. Pharmacologic inhibition of BRAF damages ATM (Ataxia telangiectasia mutated) and DDR activation in such cells, resulting in constant DNA impairment. Combination therapies with DNA-damaging agents and BRAF inhibitors escalate neoplastic cell death *in vivo* and *in vitro* and obstruct malignant melanoma regrowth after therapy termination [86].

Immunotherapies and Immune Response in Melanoma

Certain melanoma cases are not detected till the late stages, thereby indicating the need for the development of novel treatments for melanoma. Great advancements in immunotherapy management for metastatic melanoma have been observed in the past 3 decades [38, 87, 88].

Melanoma has the utmost mutation rates among all malignancies due to which tumor antigens are developed extensively. These are extremely immunogenic as identified by the immune system. Tumor antigens are classified as (i) tumor-associated antigens (TAA) and (ii) tumor-specific antigens (TSA). TAA is frequently located in neoplastic cells however they are expressed in regular cells too, whereas TSA is expressed in tumor cells only [89]. Melanoma neoplasms have a class of antigens specific to melanoma cells as well called melanoma differentiation antigens (MDA), which are responsible for melanoma differentiation. TAAs, TSAs, and MDAs are the proteins present on MHC (major histocompatibility complex) class I proteins on surfaces of melanoma cells; these alarm the immune system about the unhealthy cells [90].

T-cell receptors (TCRs) are located on CD8+ cytotoxic T lymphocytes (CTL) and can detect antigens exhibited on the MHC class I proteins. TCRs can destroy cells that are non-self-diseased or diseased self-cells. The antigen-presenting cells (APCs) like macrophages and dendritic cells have MHC class II proteins on their surface. APCs draw extracellular proteins from the surroundings and present them to CD4 + T helper (Th) cells, due to which an immune response is activated to specific antigens [91].

When the immune system is alert to the existence of non-self or diseased self-cells, lymphocytes occupy the tissue and destroy the abnormal cells. Such lymphocytes are termed tumor-infiltrating lymphocytes (TILs). TILs are envisaged in the microscope histologically to assess a neoplasm and the quantity of TILs in a neoplasm is a prognostic indicator in certain neoplasms, together with melanoma. If TILs are absent in a neoplasm, it indicates that the neoplasm has positively eluded the immune system, correlating with a poorer prognosis. The encounter of the immune system and cancerous neoplasms then comes down to a neoplasm's capacity to develop processes to evade immune recognition before the killing of the neoplasm by the immune system. This mechanism is called immune editing [38, 92]. Immune editing comprises 3 stages:

a) Elimination: when the natural killer (NK), dendritic cells (DC), CTLs, and B cells move in the neoplasm microenvironment or peripheral tissue and start destroying malignant cells quicker than they are developed.
b) Equilibrium: when the malignant cells are continuously developing but are restrained by the immune cells, thereby, counterbalancing the development and spread of malignant cells.
c) Evasion: lastly, if neoplasms move in this phase, they may defuse the immune response and grow unhindered [93].

Manipulation of immune checkpoints is a general way through which the melanoma cells escape immune recognition. The programmed cell death protein 1 (PD-1) is present on the surface of T-cells. When an immune cell detects the PD-1 ligand (PD-L1/2) on the somatic cell's surface, the immune cells are further communicated that the cell is a self-cell; the immune system is interdicted with the promotion of self-tolerance and autoimmunity prevention. The PD-1 immune checkpoint usually controls the immune system through the induction of apoptosis of growing T-cells that detect self-antigens in the lymph nodes. PD-1 averts apoptosis of regulatory T-cells also; regulatory T-cells are anti-inflammatory repressing the immune response to self-cells. This process generally guards tissue impairment in the course of anti-microbial immune responses, yet, PD-L1/2 is normally overexpressed in malignancies like melanoma, facilitating neoplastic cells to successfully seize the immune response and escape immune eradication [94].

Due to the availability of ample information about the response of the immune system to carcinoma and the processes by which malignant cells escape the immune system, novel treatments are established to attempt to relay the immune system thereby stimulating an anti-neoplastic response. Three key immune therapies for carcinomas include vaccines, adoptive cell therapies, and immunomodulatory approaches [38]. Interleukein-2 (IL2) therapy is among the initially developed immune therapies for metastatic melanoma. It is a pro-proliferative cytokine, which endorses melanoma-specific T-cells expansion. IL-2 therapies produced responses in a few cases, with 6% of patients exhibiting a complete response. This therapy was evidenced as highly toxic, and then other new therapies exhibited less toxic and more beneficial responses [95, 96].

Immune checkpoint inhibitors are the most beneficial therapies for metastatic melanoma to date, initially accepted in 2011 for clinical use [95]. To overcome melanoma manipulation of immune checkpoints, management with antibodies to counter PD1, PD-L1/2, and CTLA-4 is effective. Cytotoxic T-lymphocyte-associated protein 4 (CTLA-4) is one more immune checkpoint receptor that is constitutively expressed on T-reg cells and identifies the B7-1/2 receptor on APCs. It acts in a manner like PD-1. CTLA-4 contests with CD28 for binding to B7-1/2 on T-cells. When CD28 binds B7-1/2, there is the activation of the immune response whereas CTLA-4 binding suppresses the response. Management with antibodies to PD-1, PD-L1/2, and CTLA-4 successfully inhibits binding to the corresponding ligands and the respective signal that leads to tolerance, prompting an immune response. Three immune checkpoint inhibitor agents are officially permitted in melanoma therapy; ipilimumab (anti-CTLA-4 antibody) and 2 anti-PD-1 antibodies namely pembrolizumab and nivolumab [97, 98]. Ipilimumab therapy exhibited existence for up to 10 years in 20% of melanoma patients, in comparison to the median survival rate of <1 year in melanoma cases (stage IV). The case-control clinical trials on ipilimumab in late-stage melanoma cases have revealed a dose-dependent response to the antibody, with collective analysis steadily revealing better survival in metastatic disease cases [99]. The OS rates for ipilimumab were considerably

enhanced when compared to the vaccine alone, due to blockage of this key immune escape process, whether alone or in combination with a glycoprotein 100 peptide (GP-100) vaccine. Ipilimumab is a complete humanized IgG1 antibody approved by FDA in 2011 and was the first anti-CTLA-4 therapy [100]. There is ~37–38% response rate of Pembrolizumab in metastatic melanoma cases with a total 74% survival in a year. Nivolumab therapy displayed a ~40% response rate with a total of 73% survival rate in a year in comparison to 43% of cases having dacarbazine therapy. The combined therapy of Ipilimumab and nivolumab has brought about ~57% response rate and progression-free survival of 11.5 months in patients [101, 102].

Although checkpoint inhibitors are favorable therapies yet, complications exist in obstructing the processes that stimulate the tolerance of self-cells. In general, the adverse effects include immune-linked inflammatory disorders of the skin, GI tract, and endocrine parts. It is important to identify and manage the side effects of these therapies; the toxicity of these therapies can be counterbalanced by corticosteroid treatment in some patients, yet, some of them cannot bear the adverse effects and hence the therapy should be stopped [103].

Despite immune checkpoint inhibitors being a revolutionary breakthrough in treating metastatic melanoma, a substantial subcategory of cases still does not react to them, and if several patients are responding, they may advance to secondary resistance. Research is ongoing to find the reasons for this; great interest has been developed in the discovery of biomarkers that can foresee whether a patient responds to therapy or not, due to the cost and severe side effects of the therapies [95, 96].

Current Developments: Oncolytic Viruses used as Immunotherapies (beyond Antibody Engineering)

Oncolytic viruses (OVs) are established as cancer immunotherapies with approval of the FDA; the modified herpes simplex virus type 1 talimogene laherparepvec (T-VEC, or OncoVEXGM−CSF) being used for malignant melanoma [104], while the ClinicalTrials. gov database at present listing 22 trials for metastatic melanoma, analyzing oncolytic herpes simplex virus after the success of T-VEC [105]. Several neoplasms cannot sufficiently safeguard themselves against viral infections, this makes the advancement of oncolytic agents a striking option that may infect neoplastic cells selectively. The OVs can enhance the immune response to neoplasms, and also increase checkpoint block in combination with checkpoint inhibitors. OV treatment can trigger the release of tumor-linked antigens by lysing tumor cells, *via* dying cells and adequate cross-appearance of antigens to DCs (dendritic cells), thereby improving anti-tumor response [100].

OVs are developed as cancer-specific in clinical practice by attenuating the virus so that preferential infection of tumor cells occurs [106]. Additionally, OVs' cytokine expression may enhance the anti-cancer characteristics of these treatments escaping systemic toxicity, owing to the special action of OVs on neoplastic cells. A valuable cancer immune response modulator is IL-12 and oncolytic herpes viruses (oHSVs) expressing IL-12 have exhibited effectiveness on glioblastoma multiforme in preclinical investigations [107]. specific cytokine expression of these OVs can be the solution to achieve anti-neoplastic ability. Clinically-licensed T-VEC is equipped with GM-CSF which is an antigen-presenting cell (APC activator) [108]. A murine study reported that in comparison to its unarmed counterpart, an IL-12 with oHSVs prolonged tumor proliferation and decreased tumor growth much more effectively [106].

CONCLUSION

Melanoma is a devastating illness and displays good response in the advanced phases to limited existing therapeutic methodologies. The elementary step for melanoma treatment is its effective diagnosis. Various therapeutic modalities are available in various stages for melanoma *i.e.* surgery, radiotherapy, immunotherapy, and antibodies therapy. There is ongoing research into how and why these treatment modalities do well or fail. Biomarker discovery is important with the anticipation that sooner or later clinicians might be developing more tailored managements built on mutational and biomarker profiles. This will not only advance the prognosis but will also be cost-effective and decrease the adverse effects of the therapy.

CONSENT FOR PUBLICATION

Written informed consent was taken from the participants.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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Declared none.

AUTHOR'S CONTRIBUTION

All the authors contributed equally to the publication of this article.

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