



Review

Chemotherapy, chemoresistance and the changing treatment landscape for NSCLC

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ABSTRACT

Management of patients with lung cancer continues to pose a considerable challenge to today's oncologist. While treatment may be curative in the early stages of the disease, the majority of patients are not diagnosed until the tumor has progressed beyond the primary site. Most patients face an intensive and invasive treatment regimen comprising surgery, radiotherapy, or chemotherapy, or combinations thereof depending on disease stage/performance status. Most will require chemotherapy even if their initial surgery is potentially curative; for those with advanced disease, chemotherapy may be their only treatment option. Moreover, the majority of patients will require multiple lines of therapy as their cancer cells acquire resistance to the chemotherapeutic agents to which they are exposed. Resistance to current chemotherapeutics available for the management of non-small cell lung cancer (NSCLC) represents one of the most significant barriers to improving long-term outcomes for this vulnerable patient group. Future management may lie in individualizing therapy through careful selection of appropriate agents based on the likelihood of response and the development of resistance. A number of biomarkers are emerging that predict response to current therapeutics; work is ongoing to develop appropriate algorithms based on such markers to guide treatment selection. In addition, novel chemotherapeutics are in development including new platinum analogs such as picoplatin (a cisplatin analog), ABT-751 (a sulfonamide) and tubulin binding agents (TBAs) such as the epothilones, providing hope for the future.

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1. Introduction

Lung cancer is one of the most common cancers and cause of cancer-related death among adults, with a 5-year survival rate from the time of diagnosis of around 15% in the USA [1,2]. In 2002, there were >1.3 million individuals living with lung cancer globally, with an almost equivalent number of deaths at ~1.2 million [3]. The most recent estimates for the USA suggest that >222,000 individuals will be diagnosed with cancer of the lung and bronchus, with >157,000 individuals dying prematurely as a result of the disease in 2010 [2]. New cases and estimated deaths are anticipated to remain higher among men than women (new cases 116,750 and 105,770, respectively; deaths 86,200 and 71,080, respectively) [2]. However, data from the USA suggests that mortality due to lung cancer appears to have declined among men and stabilized among women in recent years, a situation that may be linked to the continued high relative rate of tobacco use among women [2,4]. Improved and earlier diagnosis, the availability of novel effective chemotherapeutic agents, advancing surgical techniques, and declining smoking rates may all be contributing factors to the gradually improving outcomes for this disease. However, the rate of decline in the mortality rate

associated with non-small cell lung cancer (NSCLC) still lags considerably behind that observed for other prevalent cancers such as those of the breast, prostate, colon and rectum [5]. The predominant form of the disease, NSCLC, accounts for >80% of all cases of lung cancer and is the focus of this review.

The management of NSCLC requires a multidisciplinary approach. Patients will generally require a combination of surgery, radiotherapy and/or chemotherapy, depending on their stage, resectability and overall performance status [6]. Chemotherapy is now recognized as an important component of treatment for all stages of the disease, including patients with completely resected, early stage disease, who benefit with improved survival rates when adjuvant platinum-based chemotherapy is given [7–9]. However, the majority of patients (77%) are not diagnosed until the disease has spread beyond the primary site; approximately 55% of patients have metastatic (stage IV) disease at diagnosis [1]. For these patients, chemotherapy forms the foundation of their treatment and is critical in determining their survival and quality of life. Platinum-based therapy is the mainstay of chemotherapy for NSCLC and is usually given in combination with a tubulin binding agent (TBA; including the taxanes [paclitaxel, docetaxel] and vinca alkaloids [vinorelbine, vincristine]), a camptothecin analog (irinotecan, topotecan), gemcitabine, or pemetrexed.

Despite an expanding panel of chemotherapy agents and emerging data regarding the most effective ways to deploy such agents,

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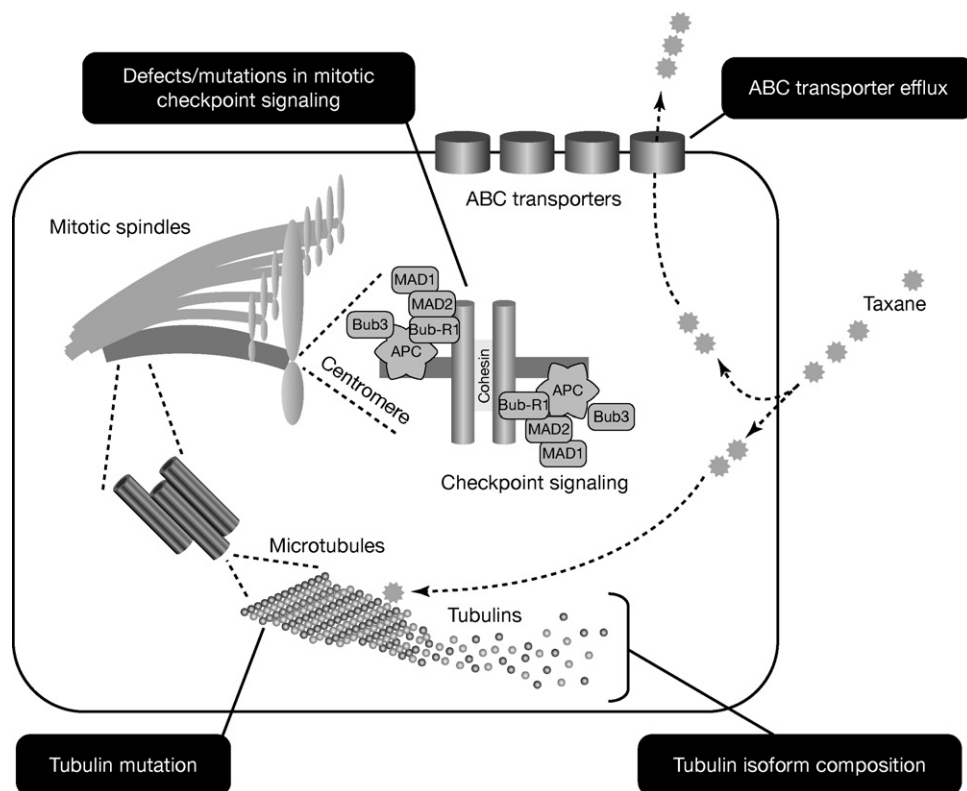


Fig. 1. Potential mechanisms of taxane resistance: (1) change of tubulin isoform composition, (2) tubulin mutation, (3) defects/mutations in mitotic checkpoints signaling, (4) ABC transporter efflux of taxane.

tumor cell resistance to chemotherapy agents (chemoresistance) continues to pose a significant challenge in the management of human neoplasms. This paper will review the molecular mechanisms that drive chemoresistance to the agents most commonly used in the treatment of NSCLC, and will examine ways in which such mechanisms can be avoided or overcome.

2. Mechanisms of resistance to chemotherapeutics

2.1. Chemoresistance is a problem for all oncologists and their patients

Chemoresistance may be innate or acquired and may apply to a single agent or to a class of agents with the same/similar antineoplastic mechanisms of action. Chemoresistance is a multifaceted problem with diverse clinical manifestations that requires an understanding of both the basic mechanisms and the evolution of such resistance as the cancer progresses.

A range of cellular mechanisms that give rise to chemoresistance to taxanes have been identified as depicted in Fig. 1 [10–12]. These established mechanisms of chemoresistance include active efflux of the chemotherapeutic agent from the tumor cell; modification of drug targets; changes or mutation in mitotic checkpoint signals; drug sequestration; de-toxification of cytotoxic agents; and enhanced DNA repair mechanisms. One or more such mechanisms of resistance also apply to other agents. In addition, an individual tumor may express more than one cellular mechanism giving rise to resistance to more than one chemotherapeutic agent or class.

2.2. Chemoresistance mechanisms have been reported for all current chemotherapeutic agents

Active efflux of chemotherapeutic agents is achieved via ABC transporters (including P-glycoprotein [P-gp] and multi-drug resis-

tance proteins [MDRs]). This mechanism contributes to resistance to anthracyclines, taxanes, platinum agents, vinca alkaloids, and topoisomerase inhibitors. Modification of the drug target including mutation of the binding site or, in the case of TBAs, a change in the relative proportion of the various tubulin isoforms, mediates resistance to taxanes, antifolates, and topoisomerase inhibitors. Changes or mutations in mitotic checkpoint signals allowing tumor cells to bypass or overcome agents that exert an antimetabolic/proapoptotic effect mediates resistance to platinum agents, taxanes and anthracyclines. Drug sequestration within defined cellular compartments mediates resistance to platinum agents, antifolates and anthracyclines. De-toxification of cytotoxic agents via enzymes such as glutathione-S transferase mediates resistance to nucleoside analogs, anthracyclines and vinca alkaloids, and enhanced DNA repair mechanisms mediate resistance to platinum agents. Thus, innate and/or acquired resistance mechanisms have been described for all the chemotherapeutic agents currently used in the treatment of this disease.

3. Chemoresistance in NSCLC

Chemoresistance is common in NSCLC. In one study of 3042 NSCLC patient tumor cultures, extreme or intermediate resistance to carboplatin was documented in 68% (1056/1565) of samples and cisplatin resistance was documented in 63% (1409/2227) of samples [13]. Resistance to doxorubicin, etoposide, gemcitabine, vinorelbine, paclitaxel, docetaxel, and topotecan was reported in 75, 63, 72, 42, 40, 52 and 31% of samples, respectively [13]. All NSCLC patients will eventually develop resistance to the chemotherapeutic agents to which they are exposed, even with a good initial response, and most will receive two or three lines of therapy. The challenge in managing patients with NSCLC is the need to establish a long-term view, taking into account the likelihood of developing chemoresistance and the effect of using a given agent at each line of

therapy. Currently, there are no reliable strategies available to guide treatment selection from first-line through all subsequent lines of therapy, to avoid the development of chemoresistance, although active clinical investigations are on going.

3.1. Resistance to platinum agents

Platinum agents exert their antineoplastic effect through direct binding to DNA resulting in cross-linking and ultimately apoptotic cell death. Several mechanisms have been proposed to account for resistance to platinum agents in NSCLC tumor cells including detoxification, intracellular accumulation, and increased DNA repair capacity.

Enzymic inactivation of platinum agents has been associated with resistance in lung cancer cells. Metallothioneins (MTs) and glutathione-related metabolism enzymes, such as glutathione transferase (GST), have been implicated in this type of resistance. One study has demonstrated increased MT expression among NSCLC patients previously treated with cisplatin, indicating that this type of resistance may be inducible [14]. Expression of the π -isoform of GST has been linked to innate resistance of lung cancer cell lines to cisplatin [15].

Expression of the DNA repair enzymes excision repair cross-complementation group 1 (ERCC1) has been shown to have predictive value for response to adjuvant platinum-based chemotherapy, following complete resection among patients with locally advanced NSCLC [16–19]. Olausson et al. [18] found that in the International Adjuvant Lung Trial (IALT), patients with completely resected NSCLC that were ERCC1-negative benefited from four cycles of adjuvant cisplatin therapy. Their retrospective analysis of data from this trial found that patients with ERCC1-negative tumors who received chemotherapy experienced significantly longer overall survival (OS) ($P=0.002$), median survival (14 months longer), and disease-free survival ($P=0.001$) compared with surgery alone [18,19]. However, there was no difference in outcomes among patients with ERCC1 positive tumors when adjuvant chemotherapy was given in addition to surgery, compared with surgery alone, leading the authors to conclude that ERCC1 expression levels can be used as a predictor for the efficacy of cisplatin-based adjuvant chemotherapy in this setting. Similarly, a retrospective analysis by Azuma et al. [17] found that low ERCC1 expression levels were predictive for response and progression-free survival (PFS) among patients with locally advanced NSCLC treated with cisplatin/docetaxel and thoracic irradiation. In a separate study, they found that ERCC1-negative patients had significantly longer median PFS and OS than ERCC1 positive patients who received paclitaxel and carboplatin against recurrent NSCLC after curative resection [16]. Li et al. [20] investigated the prognostic value of a range of biomarkers including ERCC1 in advanced NSCLC patients treated with cisplatin-based chemotherapy. Data showed that ERCC1 was predictive for OS ($P=0.002$), and furthermore, Cox proportional hazards multivariable analysis showed that ERCC1 was an independent prognostic factor ($P=0.026$) [20]. Vilmar et al. [21] conducted a larger, multi-center Phase III trial comparing two cisplatin containing regimens in patients with advanced NSCLC. In 59.5% patients with available tumor samples, they demonstrated that immunohistochemical evaluation of ERCC1 status predicts cisplatin sensitivity, and that this prediction is clinically applicable. Data showed that ERCC1-negative patients had better OS than ERCC1 positive patients, especially in adenocarcinomas with a median OS of 15.2 months as compared to 9.8 months for ERCC1 positive patients [hazard ratio of 0.64]. Their study demonstrated that ERCC1 expression is of importance for predicting treatment outcome of platinum-based chemotherapy in adenocarcinoma patients [21]. However, many studies of ERCC1 status and platinum-based chemotherapy treatment of advanced

NSCLC patients have used a small, heterogeneous population [22]. Some of the results casted doubt over the role of ERCC1 as a predictive biomarker. In contrast to the studies mentioned above, one such Phase II trial of randomized NSCLC patients undergoing platinum-based chemotherapy demonstrated that there was no statistically significant relationship between ERCC1 mRNA expression and response to chemotherapy ($P=0.794$) or hematological toxicity [23]. Due to a lack of prospective randomized trials, no firm conclusions can currently be made regarding the role of ERCC1 as a predictive biomarker for the responses of NSCLC patients to platinum-based therapy although the majority of reports were in favor of ERCC1 being predictive for platinum-based therapy [22].

The ribonucleotide reductase regulatory subunit M1 (RRM1) may also contribute to cisplatin resistance when delivered in combination with gemcitabine [24,25]. However, although preclinical data suggests increased RRM1 expression levels are associated with resistance to cisplatin *in vitro*, the effect is less pronounced than that observed for gemcitabine [24]. It may be that in this doublet combination the relationship to gemcitabine resistance is more clinically relevant in determining outcomes for patients. Both markers may also have a more general prognostic value in NSCLC [26]. Polymorphisms in other DNA repair enzymes have also been implicated in determining outcomes following platinum-based chemoradiation for patients with stage 3 NSCLC, including ERCC2 R1562 [27].

The breast cancer 1 (BRCA1) gene has also been implicated as a potential prognostic marker in resected NSCLC [28] and may predict response to cisplatin-based chemotherapy. A recent study in which chemotherapy was customized according to BRCA1 expressions levels demonstrated encouraging response and survival rates [29]. Patients with high BRCA1 levels received docetaxel alone, patients with intermediate BRCA1 expression levels received cisplatin plus docetaxel, and patients with low BRCA1 expression levels received cisplatin plus gemcitabine. Patients with high BRCA1 expression levels, facing the poorest prognosis, achieved comparable response, 1-year survival rates, and median survival durations compared with those with intermediate or low BRCA1 expression levels. However, no patient with high BRCA1 expression group survived to 2 years.

Other mechanisms of resistance to platinum agents have also been reported. These include decreased influx and increased efflux in ovarian cancer cell lines [30,31], and the requirement for an intact homologous repair mechanism, mediating the synergistic effect of combined cisplatin and gemcitabine (a doublet widely used in the treatment of NSCLC), in a Chinese hamster ovary cell model [32]. However, the contribution of these types of resistance to NSCLC is unclear.

3.2. Resistance to tubulin binding agents

There is a strong rationale for targeting tubulins in rapidly dividing cancer cells. Tubulin is the core component of microtubules which are critical for the maintenance of cell shape, transport of cellular vesicles and organelles around the cell, cell signalling, and cell division and mitosis. Microtubules exist in a state of dynamic equilibrium with the basic building blocks – α and β tubulin heterodimers – being added and removed as required. Thus, TBAs target one of the fundamental structural components of eukaryotic cells. They exert their cytotoxic effect by binding directly to defined regions of tubulin molecules and disrupting microtubule dynamics, preventing them from supporting the normal cell cycle. This results in mitotic arrest and ultimately triggers innate cell death mechanisms [33].

As noted for the platinum agents, several mechanisms have been implicated in resistance of NSCLC tumor cells to TBAs, including alterations in microtubule structure, efflux via MDRs, and alterations in the mitotic checkpoints that govern apoptosis induction. Preclinical and clinical studies suggest that alterations in tubulin

structure are an important form of chemoresistance to TBAs in NSCLC. In particular, β III-tubulin isoform levels have been shown to have prognostic and predictive value in NSCLC patients [34]. Preclinical studies have shown that high β III-tubulin levels are associated with increased taxane resistance in NSCLC cell lines [35,36]. Reducing or silencing β III-tubulin expression or translation can increase the sensitivity of NSCLC cell lines to taxanes and other cytotoxic agents including platinum agents [37–39]. Clinical studies have also demonstrated a correlation between β III-tubulin levels in NSCLC tumor cells and outcomes such as time to progression [17,40–42]. β III-tubulin levels may be predictive of response to paclitaxel, as well as progression-free survival and OS [43]. There may also be differences in response to chemotherapy depending on how advanced the disease is. For example, in an adjuvant study (JBR-10) in patients with early stage NSCLC, greater benefit from chemotherapy was seen in those with high β III-tubulin expression. This is in contrast to findings in more advanced disease [44–46].

MDR mechanisms may also contribute to TBA resistance in NSCLC. However, P-gp-mediated mechanisms appear to play only a minor role in this type of resistance in NSCLC. Instead, both preclinical and early clinical data suggests that other proteins including non-P-glycoprotein MDR (MRP) and lung-resistance-related protein (LRP) may be more important in mediating this type of resistance in NSCLC tumor cells [44].

TBAs may also act to induce apoptosis via their effects on the mitotic cycle, although quite how this is achieved remains unclear. Certain TBAs, including the taxanes paclitaxel and docetaxel, have been shown to exert direct effects on a number of proteins known to be involved in the regulation of apoptosis including Bcl-2 [44]. However, the contribution of this type of resistance in NSCLC tumor cells remains unclear.

3.3. Resistance to camptothecin analogs

Camptothecin analogs, such as irinotecan, act by inhibiting the intranuclear enzyme topoisomerase I (Topo I), an enzyme involved in DNA replication. Resistance mechanisms to camptothecin analogs in the context of NSCLC are poorly defined and data is limited to preclinical evaluations. Potential mechanisms include active efflux (via P-gp, MDR and the breast cancer resistance protein [BCRP]) [44], decreased Topo I levels (the intracellular target) [47,48], Topo I mutations [49,50], altered DNA repair mechanisms [50], and activation of nuclear factor kappa B (NF- κ B) [44].

3.4. Resistance to gemcitabine

Gemcitabine acts by inhibiting DNA synthesis through blocking DNA strand elongation. As for the camptothecin analogs, mechanisms of resistance to gemcitabine in the context of NSCLC remain poorly defined. In this case, the mechanisms may include insufficient intracellular concentrations of the active moiety via reduced levels of the activating enzyme, inefficient cellular uptake of the parent molecule, changes in the ribonucleotide reductase enzyme (the intracellular target), and alterations in associated apoptosis inducing mechanisms [44].

Preclinical studies have indicated that high RRM1 expression levels are a predictor for response of NSCLC tumor cells to gemcitabine/platinum chemotherapy [24]. This early observation has since been confirmed and translated into reduced response rates with high RRM1 expression levels in chemotherapy-naïve patients treated with gemcitabine plus docetaxel as part of a Phase III study [51], in the neoadjuvant setting among patients treated with gemcitabine plus cisplatin [52], and in a retrospective analysis of patients with advanced NSCLC enrolled in clinical trials at a single center [53]. Recent work suggests that the RRM1 gene promoter allotype may also predict response to gemcitabine-based chemotherapy,

although additional studies are required before RRM1 gene promoter sequencing can be recommended as a predictive biomarker in this setting [54].

4. Overcoming chemoresistance in NSCLC

Chemoresistance represents a considerable barrier to improving outcomes for patients with all stages of NSCLC; from those with early resectable disease eligible for adjuvant chemotherapy to those with advanced metastatic disease whose only treatment is chemotherapy. A variety of strategies have been evaluated and continue to be explored to optimize the efficacy of chemotherapeutics in the management of human NSCLC, including treatment holidays and rechallenge, switching to a different agent within the same class and switching to a different class.

4.1. Agents that can overcome resistance to existing agents

Canfosamide (TLK-286) may restore the activity of current chemotherapeutics, notably platinum agents, for whom resistance is mediated via GST π [55]. A Phase III study (ASSIST-3) is currently underway with this agent for the treatment of NSCLC versus gefitinib as the third-line therapy in locally advanced or metastatic NSCLC. Several Phase II studies are also underway with this agent as part of first-line therapy in locally advanced or metastatic NSCLC, and in combination with carboplatin and paclitaxel, cisplatin or docetaxel in platinum-resistant NSCLC.

4.2. Biomarkers and treatment selection

It is the aim of clinical trials to identify those patients, or subgroups of patients, who are likely to gain the most clinical benefit from a particular chemotherapy regimen. This approach has proved highly successful for targeted agents, for example trastuzumab in HER2-positive breast and gastric cancers, and gefitinib and erlotinib in patients with NSCLC and epidermal growth factor receptor (EGFR) mutations. But does this approach work with chemotherapy?

Treatment selection for patients with NSCLC is, at present, largely empiric although a number of biomarkers are emerging as valuable predictors of response to the current panel of chemotherapy agents available for the treatment of patients with NSCLC (Table 1). High β III-tubulin levels may be predictive for poor response to TBAs, notably the taxanes, as well as platinum agents [35,37,39]. In contrast, high β III-tubulin levels are predictive of a good response to the epothilones [56]. High expression levels of ERCC1 are predictive of a poor clinical response to platinum-based therapy [16,17,25], while mutations in the Kirsten-Rous sarcoma virus (K-ras) proto-oncogene have been shown to be predictive for a lack of benefit from the platinum-vinorelbine doublet [57]. Finally, high RRM1 expression is predictive for poor response to

Table 1
Potential biomarkers for treatment selection in NSCLC.

Biomarker	Predictive of good response to:	Predictive of poor response to:
High β III-tubulin	Epothilones	Taxanes Platinum agents
High ERCC1 levels	–	Platinum-based therapy
K-ras mutations	–	Platinum-vinorelbine doublet
High RRM1 levels	–	Gemcitabine
Reduced Tau levels ^a	–	Paclitaxel

NSCLC, non-small cell lung cancer; ERCC1, expression of the DNA repair enzymes excision repair cross-complementation group 1; RRM1, ribonucleotide reductase regulatory subunit M1.

^a Data for breast, ovarian and gastric cancers only.

Table 2
Novel agents in development for the management of advanced NSCLC.

Agent	Class/MoA	Phase of development	References/study identifiers
<i>Platinum analogs</i>			
Picoplatin	Cisplatin analog	II	Eckardt et al. [64]; NCT00021008
Satraplatin	Platinum analog	II	NCT00370383, NCT00268970, and NCT00093132
<i>TBAs</i>			
ABT-751	Sulfonamide	II	Mauer et al. [66]
Ixabepilone	Epothilone	II	Vansteenkiste et al. [67]; NCT00219297, NCT00683904, NCT00723957, and NCT00832117
Patupilone	Epothilone	II	Sanchez et al. [68]; NCT00219297
Sagopilone	Epothilone	II	NCT00160069

NSCLC, non-small cell lung cancer; TBAs, tubulin binding agents.

gemcitabine-based therapy [24,52]. Emerging evidence suggests that reduced expression of the tau family of microtubule stabilizing proteins may predict response to paclitaxel in other tumor types, including breast, gastric, and ovarian cancers [58–60]. It remains to be determined whether tau expression is also a predictive marker for response to paclitaxel in NSCLC.

The use of predictive biomarkers such as those described above should begin to ensure that individual patients receive the regimen most likely to prove effective in improving their long-term prognosis. Studies have begun to evaluate the feasibility of such customization of treatment selection based on tumor biomarkers. For example, in the study reported by Rosell et al. [29], described above, patients were assigned to treatment based on expression of the BRCA1 gene. Those with high BRCA1 expression did not receive cisplatin as part of their chemotherapy; instead they received docetaxel monotherapy and achieved comparable response, 1-year survival rates, and median survival durations compared with those with intermediate or low BRCA1 expression levels who did receive cisplatin-based therapy. Another protein, the receptor-associated protein 80 (RAP-80), may modulate this effect further. RAP-80 acts upstream of BRCA1 and mediates the ability of the BRCA1 protein to exert its action in terms of DNA repair. Among patients with low BRCA1 expression, a high RAP-80 expression level was associated with shorter median survival durations and PFS compared with a low RAP-80 expression level; conversely, among patients with high BRCA1 expression levels, a high RAP-80 expression level was associated with longer median survival and PFS compared with a low RAP-80 expression level. A similar pattern was observed when patients were stratified according to Abraxas (another protein involved in mediating BRCA1-driven DNA repair) expression levels. A Phase III study conducted by the Spanish Lung Cancer Group is now underway to prospectively evaluate the value of customizing treatment according to both BRCA1 and RAP-80 tumor expression levels. In a separate prospective Phase II study, Simon et al. [61] assigned 60 patients with chemotherapy-naïve stage 3B/4 NSCLC to treatment with or without gemcitabine based on RRM1 expression levels, and with or without carboplatin based on ERCC1 expression levels. Patients with high RRM1 expression levels received chemotherapy without gemcitabine while those with low RRM1 expression levels received gemcitabine-based chemotherapy. This approach yielded a response rate of 44% and a 1-year survival rate of 59%, with a median OS of 13.3 months. Results were comparable with a previous Phase II trial performed by this group, in a similar patient population [62]. Larger Phase III studies are now required to further evaluate the potential for this type of treatment customization.

Even with the routine use of predictive biomarkers, a significant proportion of patients with NSCLC exhibit and acquire resistance to the current panel of chemotherapeutics. As such, novel chemotherapeutics are also required that are not susceptible to the key resistance mechanisms described above. Genome-wide scanning

technology is now being applied to identify additional genetic polymorphisms that may be predictive for treatment response [63].

5. New chemotherapeutics for NSCLC

A variety of agents are currently in development for the treatment of advanced NSCLC (Table 2) [64–67]. A number of platinum analogs are in early clinical development including picoplatin (a cisplatin analog). This agent has demonstrated clinical activity among patients with platinum-refractory small cell lung cancer in a Phase II trial [64] and in a Phase III trial as a second-line treatment following platinum therapy [65]. A Phase II study is ongoing among patients with NSCLC whose disease has progressed following platinum-based chemotherapy (NCT00021008). Studies are also ongoing for the platinum analog satraplatin (NCT00370383, NCT00268970, and NCT00093132).

ABT-751 (a sulfonamide) is a novel investigational TBA that binds to the colchicine site of β -tubulin and inhibits the polymerization of microtubules. Early clinical data for this agent is encouraging even in heavily pretreated patients, including those previously treated with platinum-based regimens [66].

The epothilones, including ixabepilone, sagopilone and patupilone, are a novel class of TBAs. The epothilones bind tubulin at the same site as the taxanes. However, the molecular nature of epothilone binding is fundamentally different, involving alternative amino acids moieties [69–71]. Importantly, the epothilones retain activity in cell lines resistant to taxanes and several mechanisms have emerged that might explain this. The epothilones appear to decrease β III-tubulin expression and restore sensitivity of tumor cells to other chemotherapy agents including cisplatin and the taxanes [72]. In breast cancer cells, ixabepilone appears to be less sensitive to mutational changes within the β III-tubulin gene that impair taxane binding [73], and better at suppressing the overall dynamicity of β III-tubulin compared with paclitaxel in taxane-resistant breast cancer cells [56]. In addition, *in vitro* data suggests that ixabepilone may overcome taxane resistance in breast cancer cell lines through preferential binding to the β III-tubulin isoform [74].

Among the epothilones, ixabepilone is the most advanced in terms of clinical development. Ixabepilone has been approved for the treatment of locally advanced or MBC in combination with capecitabine after failure of an anthracycline and a taxane, and as monotherapy after failure of an anthracycline, a taxane, and capecitabine. Data from a Phase II study of ixabepilone as second-line therapy in NSCLC patients, who relapsed following platinum-based chemotherapy, has provided promising results [66]. Two dosing schedules were evaluated, a single 32 mg/m² 3-h infusion every 3 weeks and a 6 mg/m² 1-h infusion given once daily for 5 days also every 3 weeks. Outcomes were generally comparable between the two regimens with objective response rates of 14.3 and 11.6%, a median duration of response of 8.7 and 9.6 months,

Table 3
Recently approved first-line chemotherapy agents for NSCLC.

Chemotherapy agent	Year of FDA approval	Indication	Further indications
Bevacizumab	2006	Unresectable, locally advanced, recurrent or metastatic nonsquamous NSCLC	Approved for first-line treatment of unresectable advanced, metastatic or recurrent NSCLC other than predominantly squamous cell histology in addition to platinum-based chemotherapy in the EU. Approved in the US for first-line treatment of nonsquamous NSCLC in combination with carboplatin and paclitaxel.
Pemetrexed	2004	Second-line treatment for locally advanced or metastatic NSCLC	Approved for first-line treatment of advanced NSCLC by the EMEA in February 2008. Approved in combination with cisplatin therapy for the initial treatment of patients with locally advanced or metastatic nonsquamous NSCLC in the US.

FDA, Food and Drugs Administration; EMEA, European Medicines Agency.

Table 4
Global and Asian studies of treatments for NSCLC patients.

Trial name	Chemotherapy agent	Patients population	Outcomes
IPASS	Gefitinib monotherapy	Chemotherapy-naïve patients from Asian countries with advanced adenocarcinoma and never or light smoker	Improved PFS (hazard ratio [HR] 0.74; 95% confidence interval [CI] 0.65–0.85; $P < 0.0001$) and overall response rates (ORR; 43% vs. 32.2%) compared with a carboplatin/paclitaxel combination regimen. PFS (first 6 months) initially favored the combination regimen, most likely due to differences in outcomes for patients with tumors with and without EGFR mutations.
INTEREST	Gefitinib in comparison to docetaxel as second-line therapy	Locally advanced or metastatic NSCLC previously treated with a platinum-based regimen	Gefitinib proved non-inferior to docetaxel in terms of OS (HR 1.020; 96% CI 0.905–1.150; median survival 7.6 months vs. 8.0 months, respectively) supporting its use as a second-line alternative to docetaxel chemotherapy.
ISEL	Gefitinib monotherapy	1692 Patients with locally advanced or metastatic refractory NSCLC or who were intolerant to their latest chemotherapy regimen	Longer survival among Asian patients compared with placebo (HR 0.66; 95% CI 0.48–0.91; $P = 0.01$; median survival 9.5 months vs. 5.5 months).

and median survival rates of 8.3 and 7.3 months, respectively. The 1-year survival rate was 38% in both arms and responses were achieved for patients with both taxane- and platinum-pretreated tumors. Myelosuppression and neuropathy (primarily sensory) was apparent with both dosing regimens and was generally manageable.

Evaluation of the epothilones in the treatment of NSCLC is ongoing. Four Phase I/II studies are currently underway with ixabepilone in this setting (NCT00219297, NCT00683904, NCT00723957, NCT00832117). A Phase II study with patupilone yielded a response rate of 11% among 47 patients with NSCLC [63] and a Phase II study among NSCLC patients with brain metastases is ongoing (NCT00219297). The epothilones may offer an alternative for patients with innate taxane resistance including those with high pre-treatment β III-tubulin levels. Epothilones may also play a role as part of a rational sequential regimen given either after taxane-based therapy has failed, or before taxane-based therapy, to prolong disease control and improve outcomes for patients who develop taxane-resistance during therapy.

5.1. Discussion: the changing treatment landscape

The chemotherapy landscape is changing in NSCLC. In addition to the availability of novel agents that may retain activity in tumors that exhibit or have acquired chemoresistance, a number of agents originally approved in later lines of therapy, are now approved as first-line options. For example, pemetrexed, bevacizumab; and erlotinib have been approved by the FDA in the last decade (Table 3).

All three agents are now being used as part of first-line regimens with different patient selection criteria. In addition, maintenance therapy with pemetrexed in patients with advanced NSCLC who had not progressed after 4–6 cycles of first-line chemotherapy, sig-

nificantly prolonged PFS and OS as compared to best supporting care [75]. Erlotinib has also been shown to improve PFS and OS in a similar patient population when compared to placebo [76].

In Asia-Pacific countries gefitinib is often used in first- to third-line regimens based on data from both global and Asian studies including the IPASS [77], INTEREST [78], and ISEL [79] trials (Table 4). Gefitinib was recently approved by the European Medicines Agency for use in patients with EGFR mutation positive NSCLC, regardless of which line of therapy.

This expanding panel of agents for earlier lines of therapy adds another layer of complexity to the choices faced by oncologists caring for patients with this devastating disease. For the future, efforts should be made to develop individualized treatment algorithms that guide selection of the most appropriate agents and combination regimens for each line of therapy and disease stage. Such algorithms should take into account the potential for the acquisition of cross-resistance between therapeutic classes and the impact treatment selection for one line of therapy may have on subsequent treatment options. The ultimate goal should be to maximize the potential for disease control at each stage as patients move through the various lines of therapy in response to disease progression and acquisition of resistance.

6. Conclusions

Chemoresistance against the panel of agents currently available for the treatment of NSCLC is a formidable barrier to improving outcomes for this group of patients. Many of the traditional resistance mechanisms described in other solid tumors have also been reported in preclinical and clinical NSCLC studies. In order to better manage patients with NSCLC, biomarkers predictive for

response to treatment have been identified with the aim of ensuring patients receive the most active treatment regimens and, importantly, physicians can avoid prescribing regimens unlikely to yield a relevant clinical response for individual patients to spare the toxicity from chemotherapy.

In addition to more effectively deploying the current panel of chemotherapeutics, agents that restore activity as well as novel agents that retain activity in patients with innate or acquired chemoresistance are emerging. Agents which disrupt tubulin dynamics remain a rationale target in NSCLC and novel TBAs such as the epothilones are in development. The epothilones may provide some hope for attaining both of these aspirations in the future.

Conflicts of interest statement

Professor Chang has received research funding from pharmaceutical companies Bristol-Myers Squibb, AstraZeneca, Esai and Pfizer, and served on the advisory boards of Pfizer, AstraZeneca and Eli Lilly in the past 12 months.

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