Novel Cytotoxic Agents in Chemotherapy-resistant Metastatic Breast Cancer – The Epothilones

a report by

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Antimicrotubule agents have become key compounds in curative and palliative chemotherapy regimens in numerous cancers, particularly in the treatment of breast cancer. The epothilones constitute a new class of non-taxane tubulin polymerisation agents and are obtained by natural fermentation of the myxobacterium *Sorangium cellulosum*. In 1994, the National Cancer Institute (NCI) pointed out the potent cytotoxic activity of these epothilones, which is linked to the stabilisation of microtubules and results in mitotic arrest at the G2/M transition.[1]

Nevertheless, if epothilones have some similarities to taxanes in targeting and stabilising microtubules, they also have important differences.[2,3] These two families target at or near the same binding site on the β-tubulin subunit, but the structural characteristics of epothilones with a 16-member ring and a flexible structure allow binding in a manner that is unique and different from the taxanes. This characteristic provides a low susceptibility to major well-known mechanisms of tumour resistance to taxanes, including changes in tubulin isotypes and/or tubulin mutations. Moreover, epothilones can be distinguished from taxanes with their ability to overcome a resistance linked to cell membrane transporters mediated by P-glycoprotein (P-gp) and multidrug-resistance protein (MRP).[2–4]

Numerous natural epothilones variants have been identified and these are classified as epothilones A, B, C, D, E and F based on the epoxide or olefin group in the C-12 to -13 position on the macrolide ring (see *Figure 1*). Patupilone (EPO960; Novartis) is the first natural epothilone B under clinical development. In the past decade new-generation derivatives of epothilones have been synthesised with enhanced cytotoxic activity and greater stability. Numerous synthetic derivatives are under clinical development, including ixabepilone (Ixempra®, BMS-247550, Bristol Myers Squibb), ZK-EPO (ZK-219477, Schering AG), 20-desmethyl-20methylsulfanyl epothilone B (ABJ879, Novartis), KOS-862 and KOS-1584 (Kosan Biosciences).[5]

Chemotherapy-resistant tumours represent a major obstacle,[6] and the development of an agent that avoids the principal mechanisms of resistance is a welcome advance. Epothilone family agents have demonstrated consistent pre-clinical activity in chemotherapy-resistant cell lines and xenograft models, and clinical development is ongoing. Antimicrotubule agents such as docetaxel and paclitaxel have marked a significance advance in the treatment of breast cancer. Pre-clinical data have motivated the development of some epothilone agents in breast cancer to overcome a tumoral resistance to these agents. Currently, ixabepilone has completed randomised studies in metastatic disease; it is registered by the US Food and Drug Administration (FDA) and is submitting an application for the authorisation of clinical use to the European authorities.

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Epothilones in Pre-clinical Models

Epothilones have demonstrated potent cytotoxicity across a panel of cancer cell lines and has often exhibited greater potency than paclitaxel. Patupilone retained full activity against cancer cells over-expressing the P-gp, the MRP or cancer cells harbouring tubulin mutations. Similarly, the synthetic derivatives of epothilones demonstrated greater cytotoxicity than paclitaxel and remained active in cancer cells over-expressing the P-gp or harbouring tubulin mutations.

Patupilone has shown activity in paclitaxel-sensitive and -resistant tumour models and in a wide range of human cancer xenograft models. Unfortunately, the drug exhibited substantial toxicity in animal models, which limited its therapeutic efficacy. Synthetic derivatives of epothilones B such ABJ-879, ZK-EPO, BMS-310705, KOS-862 and ixabepilone demonstrated activity in paclitaxel-sensitive and -resistant tumour models, with less toxicity in animals. Synthetic derivative compounds offer several advantages, such as responses in breast cancer models of brain metastasis with EPO-ZK and improved efficacy and safety in intra-peritonal administration models with KOS-862 and BMS-30705.

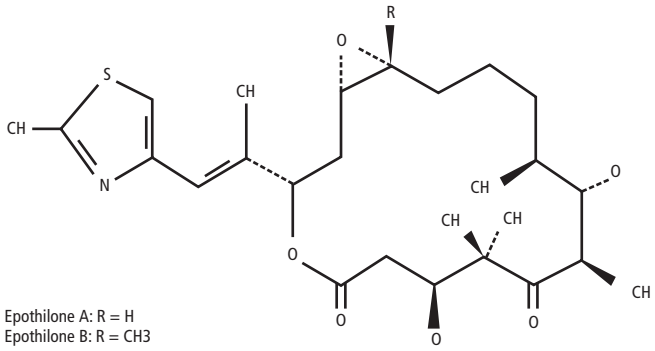
A series of toxicity studies evaluated the single- and repeat-dose intravenous toxicity of ixabepilone in laboratory animals.[7] Of interest, ixabepilone- or paclitaxel-related histopathological changes of the sciatic nerve in rats were observed, consisting of Wallerian-like axonal degeneration with axonal swelling, fragmentation and loss. The severity of the sciatic nerve changes was equivalent for both agents. A study was conducted to evaluate the neurotoxicity of ixabepilone in rats following either a single bolus administration (high peak concentration) or a single three-hour infusion (low peak concentration). Toxicity was assessed using light microscopic severity scores coupled with clinical autopsy observations. The single three-hour infusion was associated with less nerve damage than the single bolus infusion. This finding had an impact on the clinical development of ixabepilone in humans.

Table 1: Epothilones and Breast Cancer Trials

Epothilone Monotherapy Studies in Metastatic Breast Cancer		
Perez et al.[12]	Ixabepilone in patients with advanced breast cancer who are resistant to an anthracycline, a taxane and capecitabine	Phase II
Thomas et al.[11]	Ixabepilone in patients with taxane-resistant metastatic breast cancer	Phase II
Roche et al.[10]	Ixabepilone in patients with metastatic breast cancer previously treated with an anthracycline	Phase II
Denduluri et al.[21]	In patients with no prior exposure to taxanes	Phase II
Overmoyer et al.[8]	KOS-862 in patients with previous exposure to taxanes and anthracyclines	Phase II
Epothilone Combination Therapy Studies in Metastatic Breast Cancer		
Bunnel et al.[15]	Ixabepilone plus capecitabine	Phase I/II
Thomas et al.[14]	Ixabepilone plus capecitabine versus capecitabine alone in patients previously treated with an anthracycline and a taxane	Phase III
Accrual completed[18]	Ixabepilone plus capecitabine versus capecitabine alone in cancer resistant to anthracycline and taxane	Phase II
Ongoing	Weekly ixabepilone plus bevacizumab versus weekly paclitaxel plus bevacizumab in first line treatment	Randomised phase II
Accrual completed	Trastuzumab in combination with ixabepilone in women with metastatic breast cancer	Phase II
Ongoing	Trastuzumab plus weekly ixabepilone and carboplatin in patients with HER2-positive metastatic breast cancer	Phase II
Ongoing	Trastuzumab plus KOS-862 in patients with HER2-positive metastatic breast cancer	Phase II
Epothilone Studies in Early Stage Breast Cancer		
Baselga et al.[20]	Genomics study in patients receiving ixabepilone as neoadjuvant treatment for breast cancer	Phase II
Ongoing	Adjuvant trial comparing an anthracycline containing regimen followed by paclitaxel or ixabepilone	Phase III

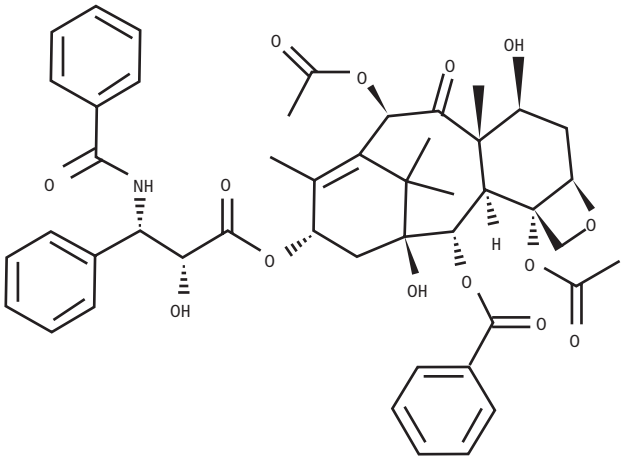
Figure 1: Taxanes and Epothilones Are Structurally Unrelated Microtubules Stabilising Agents

A. General chemical structure of epothilone group



Epothilone A: R = H
Epothilone B: R = CH3

B. Chemical structure of the taxane paclitaxel



Epothilones in Clinical Development

The epothilones and their synthetic derivatives are at various stages of clinical development. Ixabepilone is the most advamced in terms of clinical development.[3] All of the other epothilone agents are either under evaluation in phase I trials or are currently starting early phase II or III studies. For those epothilones in earlier stages of development, responses were observed in numerous tumours among these early trials, but the development of these epothilones in a specific disease is not currently defined. Their toxicity profiles are variable according to dose, schedule and possible association with other agents.

Phase I trials assessed three schedules of administration of patupilone: a weekly administration with three weeks on/one week off and six weeks on/three weeks off were studied. Diarrhoea was the dose-limiting toxicity at a maximal tolerated dose (MTD) of 2.5mg/m^2. A thrice-weekly schedule was studied in two trials: the first study established the MDT at 8mg/m^2, with diarrhoea as main toxicity; the second study escalated the dose to 11.5mg/m^2, with fatigue, diarrhoea and neuropathy as limiting toxicities. Based on those findings, a dose of 10mg/m^2 every three weeks has been selected for phase II trials. Several phase II studies with patupilone have been conducted in colorectal cancer, gastric cancer, neuroendocrine neoplasia, ovarian cancer, renal cancer and non-small-cell lung carcinoma.

BMS-310705, a water-soluble semi-synthetic analogue of epothilone B, has been administered as every-three-weeks and weekly schedules in phase I studies. Neutropenia, diarrhoea and neuropathy were the most common toxicities reported. ZK-EPO was given as an every-three-weeks schedule in a first phase I trial. The main toxicity appeared to be sensory neuropathy.

KOS-1584 has been investigated in two ongoing phase I trials with a three-weeks schedule.[8] KOS-862 is at a more advanced stage of development, with results available from four phase I trials with various schedules: every-three-weeks administration, daily treatment for three consecutive days every three weeks, a weekly schedule with three weeks-on/one week-off, a continuous intravenous infusion (24- and 72-hour) administered every two weeks. Neuropathy appeared to be the limiting toxicity in all these trials. An ongoing phase II study with KOS-862 at 100mg/m^2 for three weeks on/one week off with anthracycline and taxane pre-treated metastatic patients has been reported. Peripheral sensory neuropathy, diarrhoea and fatigue have emerged as the more frequent side effects. The objective response rate among the first 12 assessable patients was 20% and warranted further development. A second trial assessing the combination of KOS-862 and trastuzumab is ongoing. A phase II study with KOS-862 was also performed in non-small-cell lung carcinoma.[9]

Ixabepilone in Clinical Development

As previously mentioned, among the epothilones family, ixabepilone is the most advanced in clinical development.[3] The clinical results observed with ixabepilone have demonstrated clinical activity in breast cancer. Ixabepilone is also currently under evaluation in a wide variety of other cancer types.

Table 2: Grade 3–4 Toxicities in Metastatic Breast Cancer Patients Treated with Ixabepilone

	Ixabepilone Monotherapy		Ixabepilone + Capecitabine
Dose	40mg/m^2 every 3 weeks, n=240 (%)[10–12]	6mg/m^2 during 5 days every 3 weeks, n=37 (%)[16]	40mg/m^2 every 3 weeks, n=369 (%)[15]
Leukopenia	49–58	13	15
Anemia	4–8	0	3
Neutropenia	53–58	22	18
Febrile neutropenia	6	0	1
Peripheral sensory neuropathy	12–20	4	23
Fatigue/asthenia	6–14	13	9
Myalgia/arthralgia	5–10	4	8
Thrombocytopenia	0–8	4	2
Stomatitis/mucositis	4–6	0	3
Nausea/vomiting	2–6	4	4
Nail disorders	0 –2	0	1
Diarrhoea	1–4	4	6
Palmar-plantar syndrome	0–2	0	18
Dysgeusia	0–1	0	0
Hypersensitivity	0–1	0	0

Breast Cancer Trials with Ixabepilone

In metastatic breast cancer, three phase II monotherapy studies have assessed the activity of ixabepilone in several subsets of patients selected according to their tumour resistance or sensitivity to taxanes, anthracyclines and capecitabine.[10–12] *Table 1* summarises the clinical development of epothilones in patients with breast cancer. In 65 patients with taxane-sensitive metastatic breast cancer, ixabepilone showed an objective response rates (odds ratio [OR]) of 41.5% (95% confidence interval [CI] 29.4–54.4%).[10] In 49 patients with taxane-resistant metastatic breast cancer, the OR was 12% (95% CI 4.7–26.5%) in the cohort treated at a dose of 40mg/m^2 over three hours.[11] The third study assessed the activity in tumours resistant to anthracycline, taxane and capecitabine. In this resistant subset of patients, the OR among all 126 treated patients was 18.3% (95% CI 11.9–26.1%). This study appears to be the first that has demonstrated an activity in this unmet situation.[12]

Toxicity Profile of Ixabepilone

The toxicity profile of ixabepilone is summarised in *Table 2*.[7] Neuropathy appears to be the major toxicity of ixabepilone. The neuropathy is dose-dependent and may also be schedule-related. This neuropathy is cumulative in nature and gradually lessens following the discontinuation of therapy.[13] The neuropathy related to ixabepilone is typically described as sensory manifestations (i.e. dysesthesias and paresthesias involving hands and feet), and neuropathic pain has also been reported. With ixabepilone given at 40mg/m^2 as a three-hour infusion, severe peripheral neuropathy generally developed after a median of four treatment cycles (range one to 11).[10–12,14] Peripheral neuropathy (≥ grade 3) was documented as having been resolved with a median time to resolution of 5.4 weeks (95% CI 3.3–11.4 weeks).[12] Motor and autonomic neuropathy has only been reported as a related grade 3/4 event in <1% of all patients, and they were not described in the absence of sensory or painful neuropathy.[10–12,14,15] Interestingly, a phase II trial assessed the administration of ixabepilone at 6mg/m^2/day on days one to five every three weeks in patients previously exposed to taxane-containing therapy. An objective response rate of 22% was reported with a different toxicity profile, including a lower rate of neuropathy.[16] This finding warranted future studies to compare the various schedules.

Combination Studies with Ixabepilone

Combining ixabepilone and capecitabine is based on the complementary mechanisms of action and primarily non-overlapping toxicity profiles of these two agents.[15] The recommended doses resulting from the phase I/II study are ixabepilone 40mg/m^2 (three-hour infusion on day one every 21 days) and capecitabine 2,000mg/m^2 (twice daily divided doses on days one to 14 every 21 days). This association is being studied in two ongoing randomised phase III trials comparing capecitabine alone versus ixabepilone and capecitabine in patients with metastatic breast cancer. Rare studies had demonstrated a survival benefit in patients with metastatic breast cancer with therapy given after the first-line treatment.[17] One can consider that outstanding activity is required to demonstrate a survival benefit with a treatment beyond previous exposure to taxanes and anthracycline. The 752 patients in the first phase III trial, CA163-046, were heavily pre-treated and had widespread metastatic disease (84% had significant baseline visceral disease involving the liver and/or lung).[14] Eighty-five per cent of patients had progressed on prior taxane therapy for MBC, and 93% had experienced ≥1 prior regimen for metastatic disease. Treatment with ixabepilone plus capecitabine resulted in a median progression-free survival (PFS) of 5.8 months compared with 4.2 months for capecitabine monotherapy, reflecting a 25% reduction in estimated risk of disease progression (hazard ratio [HR] 0.75; p=0.0003). Response rates were also greater in the combination arm (35 versus 14%; p<0.0001).[14]

Rare studies had demonstrated a survival benefit in patients with metastatic breast cancer with therapy given after the first-line treatment.

The second study, CA163-048, has enrolled 1,221 patients who were either pre-treated with or resistant to an anthracycline and a taxane. Baseline characteristics were essentially similar between these trials, except that half of the patients enrolled in CA163-046 were resistant to prior therapy, while nearly all of the patients in CA163-048 were resistant.[18]

The median PFS in CA163-048 was 6.2 months for ixabepilone plus capecitabine and 4.4 months for capecitabine alone (p=0.0005) and the hazard ratio was 0.79 (p=0.0005), indicating a 21% reduction in the risk of progression in favour of the combination arm. The OR rates were 43 versus 29%, respectively. Median overall survival for the ixabepilone plus capecitabine arm versus capecitabine alone in the CA163-046 study was 12.9 versus 11.1 months, respectively (HR 0.90, 95% CI 0.77–1.05; p=0.1936), and in the CA163-048 study it was 16.4 versus 15.6 months, respectively (HR 0.90 95% CI 0.78–1.03; p=0.1162).[14,18] A pooled

analysis from the two phase II studies of 1,337 patients with taxane-resistant MBC showed that OR rates (39 versus 22%) and PFS (5.1 versus 3.7 months) favoured the ixabepilone plus capecitabine arm.[19]

Compared with the monotherapy arm, the combination regimen in CA163-046 resulted in higher levels of grade 3/4 treatment-related sensory neuropathy (20.8 versus 0%), fatigue (9 versus 3.3%) and neutropenia (68 versus 11%). Peripheral sensory neuropathy was generally reversible. Capecitabine-related toxicities were similar in both treatment groups.[14] In CA163-048, grade 3/4 neuropathy was 24% for the combination regimen versus 1% for capecitabine alone.[18]

One phase II study is currently evaluating the combination of ixabepilone and trastuzumab in patients with metastatic HER-2-positive tumours. A second trial is assessing the association between carboplatin plus ixabepilone and trastuzumab in the same population. Ixabepilone is being assessed in combination with bevacizumab versus paclitaxel plus bevacizumab in a randomised phase II study of first-line treatment for metastatic disease. In this trial, two schedules of ixabepilone administration were studied: the standard 40mg/m^2 in a three-hour infusion every three weeks versus 16mg/m^2 in a one-hour infusion given on three consecutive weeks followed by a week of rest. Combinations of ixabepilone with liposomal doxorubicin, doxorubicine and epirubicin are also under clinical evaluation.

A phase II trial is evaluating ixabepilone in a neo-adjuvant setting. Following four cycles of ixabepilone 40mg/m^2 given every three weeks, patients underwent surgical resection and an adjuvant anthracycline containing regimen. Among the 164 patients included, preliminary results were available in 96 patients.[20] A complete pathological response was observed in 19% of cases, of whom 11% also had a complete pathological response in the axillary. Interestingly, a subanalysis pointed out an encouraging level of activity in the subgroup of patients with oestrogen-receptor-negative status and HER-2-negative status tumours. Among those patients, 26 and 19% achieved a complete pathological response in the breast and in the axillary, respectively. This result suggests it would be a good idea to plan a subset analysis in the large randomised phase III studies among this subset. This final subset analysis will be available within a few months.

A randomised trial comparing two adjuvant treatments is ongoing: an anthracycline-containing regimen followed by paclitaxel or ixabepilone in patients with negative oestrogen receptors, negative progesterone receptors, negative HER-2 status tumours or in patients with positive oestrogen receptors, negative progesterone receptors, negative HER-2 status and axillary nodal involvement. *Table 1* lists the completed and ongoing clinical trials with ixabepilone in breast cancer.

It seems highly probable that in the near future ixabepilone will demonstrate a benefit in various tumour types and clinical settings.

Conclusion

Chemotherapy resistance represents a major obstacle and the development of an agent that avoids the principal mechanisms of resistance is a welcome advance. Consistent with pre-clinical data, epothilones demonstrate promising antitumour activity in a broad spectrum of taxane-sensitive and -resistant tumours. Epothilones show a difference in toxicity profile for all compound doses, and schedules related to acceptable side effects have been identified. Ixabepilone is at an advanced stage of clinical development, and results may allow a first registration in metastatic breast cancer. Nevertheless, the routine use of this compound requires a stringent follow-up of neurological toxicity to avoid irreversible damage. It seems highly probable that in the near future ixabepilone will demonstrate a benefit in various tumour types and clinical settings. ■

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