Tucidinostat plus exemestane for postmenopausal patients with advanced, hormone receptor-positive breast cancer (ACE): a randomised, double-blind, placebo-controlled, phase 3 trial

Zefei Jiang, Wei Li, Xichun Hu, Qingyuan Zhang, Tao Sun, Shude Cui, Shusen Wang, Quchang Ouyang, Yongmei Yin, Cuizhi Geng, Zhongsheng Tong, Ying Cheng, Yueyin Pan, Yuping Sun, Hong Wang, Tao Ouyang, Kangsheng Gu, Jifeng Feng, Xiaojia Wang, Shubin Wang, Tianshu Liu, Jinghua Gao, Massimo Cristofanilli, Zhiqiang Ning, Xianping Lu

Lancet Oncol 2019; 20: 806–15

Published Online
April 26, 2019
http://dx.doi.org/10.1016/S1470-2045(19)30164-0

See Comment page 746

The Fifth Medical Centre of Chinese PLA General Hospital, Beijing, China (Prof Z Jiang MD); **The First Hospital of Jilin University, Changchun, China** (Prof W Li MD); **Fudan University Shanghai Cancer Centre, Shanghai, China** (Prof X Hu MD); **Harbin Medical University Cancer Hospital, Harbin, China** (Prof Q Zhang MD); **Liaoning Cancer Hospital & Institute, Shenyang, China** (Prof T Sun MD); **Henan Cancer Hospital, Zhengzhou, China** (Prof S Cui MD); **Sun Yat-Sen University Cancer Centre, Guangzhou, China** (Prof Shus Wang MD); **Hunan Cancer Hospital, Changsha, China** (Prof Q Ouyang MD); **Jiangsu Province Hospital, Nanjing, China** (Prof Y Yin MD); **Tumour Hospital of Hebei Province, Shijiazhuang, China** (Prof C Geng MD); **Tianjin Medical University Cancer Institute and Hospital, Tianjin, China** (Prof Z Tong MD); **Jilin Cancer Hospital, Changchun, China** (Prof Y Cheng MD); **Anhui Provincial Hospital, Hefei, China** (Prof Y Pan MD); **Jinan Central Hospital, Jinan, China** (Y Sun MD); **The Third Hospital of Nanchang, Nanchang, China** (H Wang MD); **Beijing Cancer Hospital, Beijing, China** (Prof T Ouyang MD); **The First Affiliated Hospital of Anui Medical University, Hefei, China** (Prof K Gu MD); **Jiangsu Cancer Hospital, Nanjing, China** (Prof J Feng MD); **Zhejiang Cancer Hospital, Hangzhou, China** (Prof X Wang MD);

Summary

Background Tucidinostat (formerly known as chidamide) is an oral subtype-selective histone deacetylase inhibitor. In an exploratory study, the combination of tucidinostat with exemestane showed preliminary signs of encouraging anti-tumour activity in patients with advanced hormone receptor-positive breast cancer. To build on these findings, we aimed to assess the efficacy and safety of this combination in a randomised trial in a larger population of postmenopausal patients with advanced, hormone receptor-positive breast cancer.

Methods We did the randomised, double-blind, placebo-controlled, phase 3 ACE trial at 22 specialist cancer centres in China. Eligible patients were postmenopausal women (aged ≥60 years or aged <60 years if their serum follicle-stimulating hormone and oestradiol concentrations were within postmenopausal ranges) with hormone receptor-positive, HER2-negative breast cancer, whose disease had relapsed or progressed after at least one endocrine therapy (either in advanced or metastatic or adjuvant setting), and who had at least one measurable lesion, adequate organ function, Eastern Cooperative Oncology Group (ECOG) performance status of 0–1, and adequate haematological and biochemical parameters. Endocrine therapy did not have to be the most recent therapy before randomisation, but recurrence or progression after the most recent therapy was a prerequisite. Patients were randomly assigned (2:1) by a dynamic randomisation scheme via an interactive web-response system to receive 30 mg oral tucidinostat or placebo twice weekly. All patients in both groups also received 25 mg oral exemestane daily. Randomisation was stratified according to the presence of visceral metastases (yes *vs* no). Patients, investigators, study site staff, and the sponsor were masked to treatment assignment. The primary endpoint was investigator-assessed progression-free survival. Efficacy analyses were done in the full analysis set population, comprising all patients who received at least one dose of any study treatment, and safety analyses were done in all patients who received at least one dose of any study treatment and for whom at least one safety case report form was available. This study is registered with ClinicalTrials.gov, number NCT02482753. The study has reached the required number of events for final analysis of the primary endpoint. The trial is no longer enrolling patients, but follow-up for investigation of overall survival is ongoing.

Findings Between July 20, 2015, and June 26, 2017, 365 patients were enrolled and randomly assigned, 244 to the tucidinostat group and 121 to the placebo group. The median duration of follow-up was 13·9 months (IQR 9·8–17·5). Investigator-assessed median progression-free survival was 7·4 months (95% CI 5·5–9·2) in the tucidinostat group and 3·8 months (3·7–5·5) in the placebo group (HR 0·75 [95% CI 0·58–0·98]; p=0·033). The most common grade 3 or 4 adverse events in either group were neutropenia (124 [51%] of 244 patients in the tucidinostat group *vs* three [2%] of 121 patients in the placebo group), thrombocytopenia (67 [27%] *vs* three [2%]), and leucopenia (46 [19%] *vs* three [2%]). Serious adverse events of any cause occurred in 51 (21%) of 244 patients in the tucidinostat group and seven (6%) of 121 patients in the placebo group. No treatment-related deaths were reported.

Interpretation Tucidinostat plus exemestane improved progression-free survival compared with placebo plus exemestane in patients with advanced, hormone receptor-positive, HER2-negative breast cancer that progressed after previous endocrine therapy. Grade 3–4 haematological adverse events were more common in the tucidinostat plus exemestane group than in the placebo plus exemestane group. Tucidinostat plus exemestane could represent a new treatment option for these patients.

Funding Chipscreen Biosciences.

Research in context

Evidence before this study

We searched PubMed with the search terms "HDAC inhibitor" AND "breast" AND ("advanced" or "metastatic") for reports published in English between Jan 1, 2006, and Jan 28, 2019, to identify clinical studies of histone deacetylase (HDAC) inhibitors in advanced breast cancer. We identified one randomised phase 2 trial and six single-arm phase 1 and 2 studies. Although several HDAC inhibitors have been approved for haematological malignancies, their efficacy in solid tumours has not been shown. In an exploratory clinical trial in patients with advanced hormone receptor-positive breast cancer, the combination of tucidinostat and exemestane had encouraging activity and a tolerable safety profile.

Added value of this study

To our knowledge, our trial is the first phase 3 randomised clinical trial of an HDAC inhibitor (tucidinostat) in combination with an endocrine therapy (exemestane) in patients with advanced, hormone receptor-positive breast cancer that has progressed after previous endocrine therapy. Compared with placebo plus exemestane, tucidinostat plus exemestane significantly improved progression-free survival and substantially increased overall response and clinical benefit in this patient population. The combination of tucidinostat plus exemestane was associated with a manageable safety profile.

Implications of all the available evidence

Primary and secondary drug resistance to endocrine therapy remains a major challenge in the treatment of hormone receptor-positive breast cancer. Epigenetic modulation plus endocrine blockade with tucidinostat and exemestane is a feasible and adequately tolerated strategy against drug resistance and relapse in advanced breast cancer. This combination could be a new treatment option for patients with advanced hormone receptor-positive, HER2-negative breast cancer that has progressed after previous endocrine therapy.

Beijing University Shenzhen Hospital, Shenzhen, China (Prof Shub Wang MD); **Fudan University Zhongshan Hospital, Shanghai, China** (Prof T Liu MD); **Cangzhou Central Hospital, Cangzhou, China** (J Gao MD); **Feinberg School of Medicine, Chicago, IL, USA** (Prof M Cristofanilli MD); **and Chipscreen Biosciences, Shenzhen, China** (Z Ning MD, X Lu PhD)

Correspondence to:
Prof Zefei Jiang, Department of Breast Cancer, The Fifth Medical Centre of Chinese PLA General Hospital, Beijing 00071, China
jiangzefei@csco.org.cn

Introduction

Endocrine therapy is the recommended systemic treatment for patients with hormone receptor-positive breast cancer, the most common subtype of this disease. However, in a large subset of patients, both primary and acquired resistance to endocrine therapies can develop. Thus, identification and development of effective therapeutic options that prolong or restore sensitivity to endocrine treatment are important in patients with advanced hormone receptor-positive breast cancer.

Aberrant gene expression due to epigenetic alterations correlates with disease progression and resistance to endocrine therapy in breast cancers.[1,2] Epigenetic alterations can be modulated or reversed with histone deacetylase (HDAC) inhibitors, which modify the status of acetylation on histone and non-histone proteins to induce cell cycle arrest, differentiation, and death in cancer cells, and to change the tumour microenvironment.[3,4] Several HDAC inhibitors have been approved for use in haematological malignancies,[5] but efficacy in solid tumours has not been shown.

Tucidinostat (formerly known as chidamide) is an oral benzamide class of HDAC inhibitor with subtype specificity for inhibition of HDAC1, HDAC2, HDAC3, and HDAC10.[6] It has been approved in China for relapsed or refractory peripheral T-cell lymphoma,[7,8] and is under clinical development globally for various cancers. Previous studies[6,9,10] have shown that the benzamide class of subtype-selective HDAC inhibitors, including tucidinostat and entinostat, enhance tumour immune surveillance via activation of natural killer cell-specific and antigen-specific cytotoxic T-cell-mediated cellular anti-tumour immunity, which differentiates the subtype-selective agent from other non-selective HDAC inhibitors. Tucidinostat downregulates oestrogen-independent growth factor signalling pathways and restores sensitivity to anti-oestrogen agents.[11] An exploratory clinical study[12] showed that the combination of tucidinostat with the aromatase inhibitor exemestane had encouraging activity and a tolerable safety profile in postmenopausal patients with advanced hormone receptor-positive breast cancer. We aimed to assess the efficacy and safety of tucidinostat plus exemestane in postmenopausal patients with hormone receptor-positive, HER2-negative advanced breast cancer that had progressed after previous endocrine therapy.

Methods

Study design and participants

The Chidamide and Exemestane (ACE) study was a randomised, double-blind, placebo-controlled, phase 3 trial done at 22 specialist cancer centres in China (appendix p 3). Eligible patients were postmenopausal women with histologically or cytologically confirmed hormone receptor positive, HER2-negative, inoperable breast cancer, whose disease relapsed or progressed after at least one endocrine therapy (either in the advanced or metastatic or adjuvant setting), and who had at least one measurable lesion according to the Response Evaluation Criteria in Solid Tumours (RECIST; version 1.1);[13] adequate organ function; Eastern Cooperative Oncology Group (ECOG) performance status of 0 or 1; a life expectancy of at least 3 months; and absolute neutrophil count of at least 1500 cells per μL, platelet count of at least 100 000 per μL, haemoglobin concentrations of 9·0 g/dL or higher, total bilirubin concentrations less than or equal to 1·5 times the upper limit of normal (ULN), alanine aminotransferase and aspartate aminotransferase concentrations less than or equal to 2·5 times the ULN (≤5 times the ULN in patients with

See Online for appendix

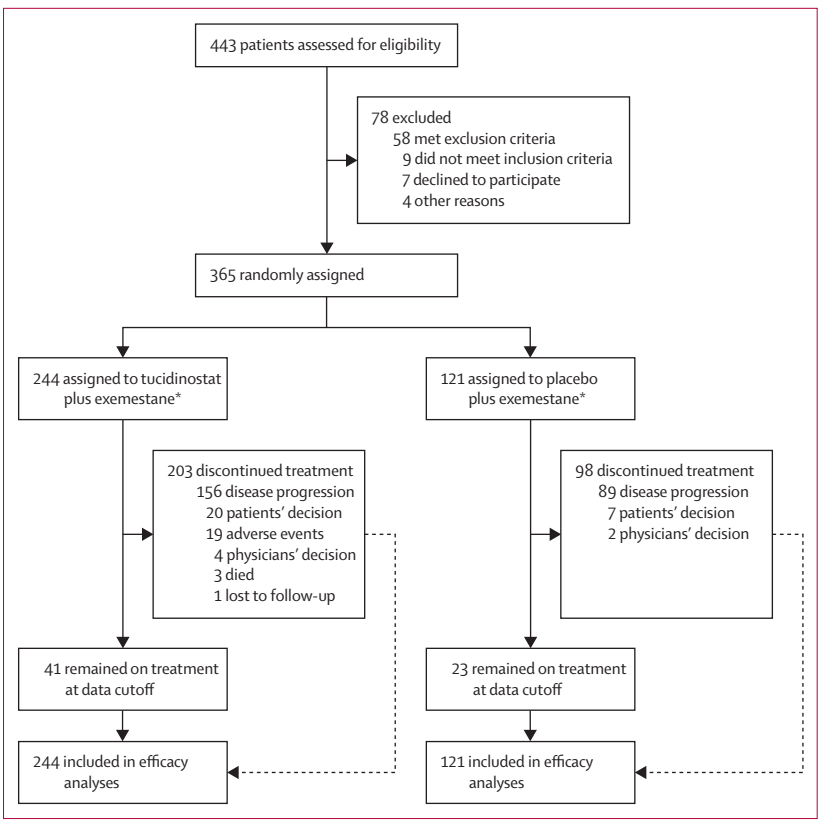

Figure 1: **Trial profile**
*All participants received at least one dose of their assigned treatment.

liver metastases), and serum creatinine concentrations less than or equal to 1·5 times the ULN. Participants were judged to be postmenopausal if they had undergone surgical menopause with bilateral oophorectomy; were aged 60 years or older; were younger than 60 years but had been in natural menopause for at least 12 months, with serum follicle-stimulating hormone (FSH) and oestradiol concentrations within locally used post-menopausal ranges, and had not been treated with chemotherapy, tamoxifen, toremifene, or ovarian suppression for at least 1 year before study entry; or were younger than 60 years and receiving tamoxifen or toremifene, with serum FSH and oestradiol concentrations within the postmenopausal range. Hormone receptor status (ie, expression of estrogen receptor or progesterone receptor, or both) and HER2 status were assessed locally in the most recently obtained tumour tissue sample. Endocrine therapy did not have to be the most recent treatment before enrolment, but recurrence or progression after receipt of the most recent therapy was required for randomisation. Participants were allowed to have received one previous line of chemotherapy for advanced disease. To ensure accurate assessment, patients with bone-only lesions were screened by bone scan, followed by confirmation with CT or MRI. Previous exposure to exemestane was not allowed. Patients with uncontrolled brain metastases, uncontrolled or significant cardiovascular disease, clinically significant gastrointestinal abnormalities, active infection or persistent fever during the 14 days before study entry, or mental disorders that could interfere with treatment compliance were excluded. The full inclusion and exclusion criteria are in the appendix (pp 60–62).

Written, informed consent was obtained from every patient before screening and enrolment. The ethics committee at each participating centre approved the study, which was done in accordance with the principles of Good Clinical Practice, the Declaration of Helsinki, and Chinese regulations. The study protocol is in the appendix (pp 30–86).

Randomisation and masking

Patients were randomly assigned (2:1) to receive oral tucidinostat plus exemestane or placebo plus exemestane. Randomisation was done centrally and stratified according to the presence of visceral metastases (yes *vs* no). We used a dynamic randomisation scheme with an interactive web-response system based on drug pack number allocation. Investigators enrolled participants and used identifying information to register them in the interactive web-response system. Patients were assigned three-digit random numbers and treatment groups. Tucidinostat tablets and the matching placebo had identical packaging, labelling, appearance, and administration schedules. Patients, investigators, study site staff, and the sponsor were masked to treatment assignment until the database was locked. The local and central imaging review pathologists were also masked to treatment allocation.

Procedures

30 mg tucidinostat or matching placebo was given orally (as six 5 mg tablets per day) twice a week (either, Monday and Thursday, Tuesday and Friday, or Wednesday and Saturday) for 4 consecutive weeks in a 4-week cycle. Patients also took 25 mg exemestane orally daily. Efforts were made to maintain the planned schedule and dose—eg, patients had drug diaries to document drug intake. Completed drug diaries were returned to the study site at the next study visit. Patients continued to receive treatment until disease progression, development of unacceptable toxic effects, loss to follow-up, or withdrawal of consent. Dose reduction of tucidinostat or placebo (first to 20 mg, then to 10 mg) was allowed if adverse events occurred, per criteria defined in the study protocol (appendix pp 64–66). Reduction of exemestane doses was not allowed. Delay of tucidinostat or placebo was allowed for up to 2 weeks. Patients in whom grade 4 haematological adverse events, grade 3 neutrophil reductions with body temperatures higher than 38·5°C, or grade 3 non-haematological adverse events stopped tucidinostat or

placebo and received treatment for their symptoms (per investigators' discretion). Study drug was resumed with dose reduction if the adverse events improved to grade 1 or lower within 2 weeks. If adverse events did not improve, study drug was terminated.

CT or MRI, or both, of the chest, abdomen, and pelvis was done to screen patients during the 3 weeks before randomisation, and then repeated every 8 weeks until disease progression. In patients with bone-only lesions, we also assessed extra-bone tissues and organs for new disease lesions. Radiographic bone scans were done at screening and subsequently as clinically indicated or to confirm complete responses. Patients who discontinued study drugs for any reason other than progression continued to undergo imaging assessments every 8 weeks until disease progression or until they started other anti-tumour treatment.

The incidence and severity of adverse events and serious adverse events were monitored by investigators throughout the study period. Haematological laboratory tests were done weekly and biochemical laboratory tests were done every 4 weeks. Adverse events were recorded and graded according to the National Cancer Institute's Common Terminology Criteria for Adverse Events (version 4.0). Suspected causal associations between study drugs and adverse events were recorded on the basis of investigators' judgment.

Outcomes

The primary endpoint was investigator-assessed progression-free survival, defined as the time from randomisation to either the first documented disease progression (according to RECIST version 1.1) or death from any cause. Secondary endpoints were overall survival (defined as the time from randomisation to death from any cause), objective response (defined as the proportion of patients with a complete response or a partial response according to RECIST), clinical benefit (defined as the proportion of patients with an objective response or stable disease lasting 24 weeks or longer per RECIST), duration of response (defined as the time from first documented objective response to death or first documented progression per RECIST), and safety (defined as the frequency and severity of adverse events and laboratory abnormalities).

Statistical analysis

For the primary endpoint, we used a log-rank test adjusted for visceral metastases. We estimated that 255 events (ie, disease progression or death) would be required in the two treatment groups for the study to have 80% power to detect a 44% improvement in median progression-free survival from 4·1 months to 5·9 months (representing an approximately 30% reduction in risk of disease progression or death) with a two-sided 5% level of significance. Furthermore, assuming 24 months of recruitment, 12 months of follow-up, 10% loss to follow-up, and a 2:1 randomisation ratio in favour of the tucidinostat group, we calculated that a sample size of 328 patients was needed.

Efficacy analyses were done in the full analysis set, comprising all randomly assigned patients who received at least one dose of study drug. Safety analyses included all patients who had received at least one dose of study drug and for whom at least one safety case report form had been completed. The Kaplan-Meier method was applied to estimate the distribution of data for progression-free survival, and we used a prespecified Cox proportional-hazards model adjusted for visceral metastases to estimate treatment effect, which was expressed as a hazard ratio (HR) with a 95% CI. Patients who did not have a progression-free survival event were

	Tucidinostat group (n=244)	Placebo group (n=121)
Age, years	55 (48–61)	55 (48–62)
ECOG performance status		
0	95 (39%)	38 (31%)
1	149 (61%)	83 (69%)
Visceral disease*	138 (57%)	62 (51%)
Measurable disease†	193 (79%)	96 (79%)
Metastatic site		
Lung	86 (35%)	36 (30%)
Liver	70 (29%)	32 (26%)
Bone only	52 (21%)	26 (21%)
Number of previous treatments for metastatic disease		
0	106 (43%)	49 (40%)
1	80 (33%)	39 (32%)
2	51 (21%)	29 (24%)
3	7 (3%)	4 (3%)
Previous endocrine treatment for metastatic disease		
Yes	115 (47%)	59 (49%)
Anti-oestrogen	15 (6%)	10 (8%)
Aromatase inhibitor	97 (40%)	50 (41%)
Fulvestrant	17 (7%)	3 (2%)
No	129 (53%)	62 (51%)
Sensitive to previous endocrine therapy‡		
Yes	162 (66%)	83 (69%)
No	82 (34%)	38 (31%)
Previous chemotherapy		
No previous chemotherapy	14 (6%)	8 (7%)
For metastatic disease	69 (28%)	41 (34%)
For adjuvant disease	161 (66%)	72 (60%)
Progesterone receptor status		
Positive	201 (82%)	98 (81%)
Negative	43 (18%)	23 (19%)

Data are median (IQR) or n (%). All participants were of Chinese ethnicity. ECOG=Eastern Cooperative Oncology Group. *Visceral disease was defined as involvement of the lungs, liver, spleen, or adrenal gland. †All other patients had at least one mainly lytic bone lesion. ‡Sensitivity was defined as at least 24 months of endocrine therapy before recurrence in the adjuvant setting or a response or stabilisation for at least 6 months with endocrine therapy for advanced disease.

Table 1: **Baseline characteristics in the full analysis set**

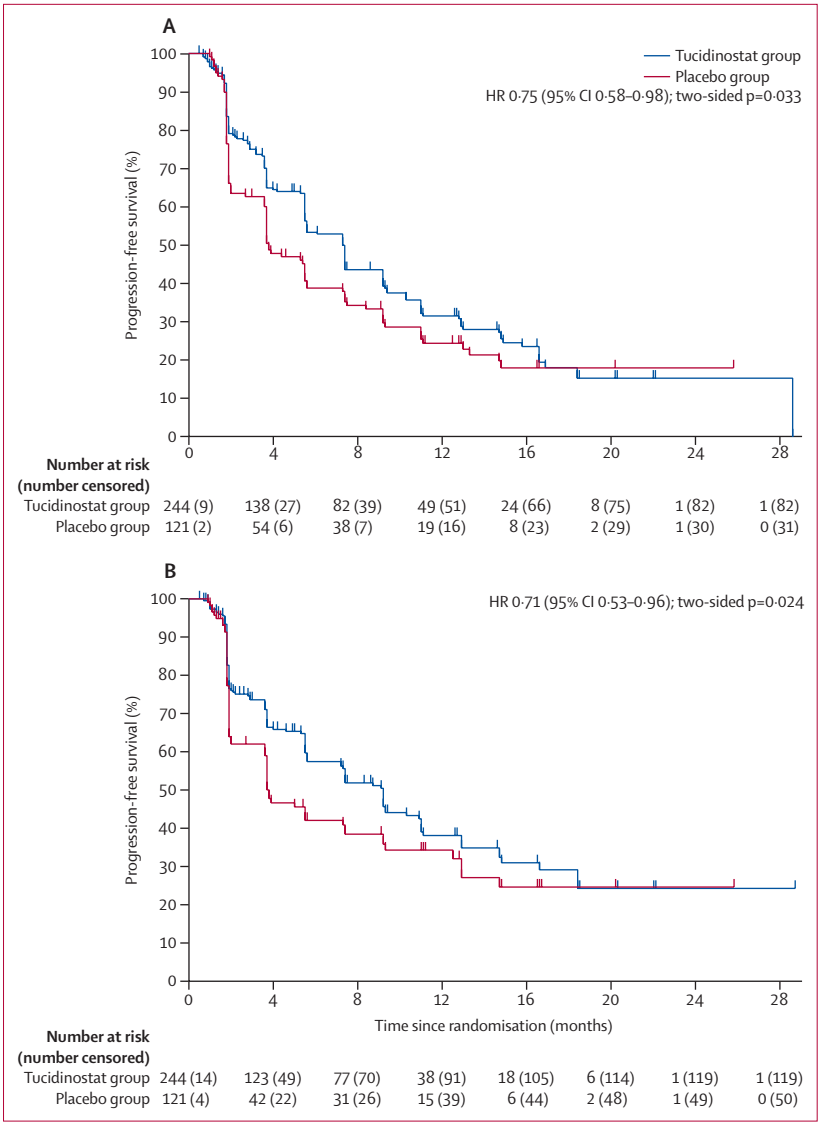

Figure 2: **Kaplan-Meier plots of progression-free survival assessed by (A) investigators and (B) independent central review**

Dashes on the curves represent censored patients. HR=hazard ratio.

censored at the date of their last adequate tumour assessment. We also assessed progression-free survival in prespecified subgroups based on age, stratification factor, and other potential baseline prognostic factors; the effects of treatments by subgroup were reported as HRs with 95% CIs in a forest plot. A blinded, independent central review of progression-free survival was done in all patients as a supportive analysis. The proportions of patients who achieved an objective response or clinical benefit were compared between the two groups with the logistic regression model (adjusted for visceral metastases). The distribution of the data for duration of response was estimated with the Kaplan-Meier method, and the HR was estimated with the Cox proportional-hazards model used to estimate treatment effect. Overall survival was summarised and analysed in the same way as progression-free survival.

On the basis of the calculated sample size and preplanned study duration, no interim analyses were planned or done. All statistical analyses were done in SAS (version 9.4), and all p values reported are two sided. This study is registered with ClinicalTrials.gov, number NCT02482753.

Role of the funding source

The funder of the study had a role in study design, data collection, data analysis, data interpretation, and writing of the report. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

Results

Between July 20, 2015, and June 26, 2017, 365 patients were enrolled and randomly assigned, 244 to tucidinostat plus exemestane (tucidinostat group) and 121 to placebo plus exemestane group (placebo group; figure 1). The rate of loss to follow-up during the study seemed to be higher than 10% (based on a higher-than-expected number of withdrawals), which is why 365 patients were enrolled rather than the projected sample size of 328. Baseline characteristics were well balanced between the treatment groups (table 1). Detailed data for the numbers of patients enrolled in each group meeting the different postmenopausal enrolment criteria are in the appendix (p 4). Because neither CDK4 or CDK6 inhibitors nor everolimus were approved in China during the enrolment period, very few patients had previously received such agents. Three patients in the tucidinostat group and four in the placebo had previously participated in a palbociclib clinical trial, but information about whether the patients had received the active drug or placebo was unavailable. One patient in the tucidinostat group had previously received everolimus treatment. By the data cutoff date (March 9, 2018), 252 events of disease progression or death had occurred, 162 in the tucidinostat group and 90 in the placebo group. 41 (17%) of 244 patients in the tucidinostat group and 23 (19%) of 121 patients in the placebo group were still receiving treatment. Median treatment duration was 24 weeks (IQR 8–44) in the tucidinostat group and 16 weeks (IQR 8–40) in the placebo group. Overall, the median duration of follow-up was 13·9 months (IQR 9·8–17·5).

Investigator-assessed median progression-free survival was 7·4 months (95% CI 5·5–9·2) in the tucidinostat group and 3·8 months (3·7–5·5) in the placebo group (HR 0·75 [95% CI 0·58–0·98]; p=0·033; figure 2A; table 2). In central imaging assessments by blinded independent review, median progression-free survival was 9·2 months (95% CI 7·2–10·9) in the tucidinostat group and 3·8 months (3·6–7·4) in the placebo group (HR 0·71 [95% CI 0·53–0·96]; p=0·024; figure 2B;

table 2). Figure 3 shows progression-free survival in different patient subgroups.

The median relative dose intensity was 92% (IQR 74·3–100·0) for the 244 patients who received tucidinostat (median dose intensity 55·3 mg [IQR 44·6–60·0]) and 100% (98·5–100·0) for 121 patients who received placebo (60·0 mg [59·1–60·0]). The median relative dose intensity of exemestane was 100% (99·2–100·0) for the tucidinostat group (25·0 mg [24·8–25·0]) and 100% (100·0–100·0) for the placebo group (25·0 mg [IQR 25·0–25·0]). 45 (18% [95% CI 14–23]) of 244 patients in the tucidinostat group had an objective response per investigator assessment, compared with 11 (9% [4–14]) in the placebo group (p=0·026). Median duration of response did not differ significantly between groups (table 2). More patients in the tucidinostat group than in the placebo group experienced clinical benefit (table 2). Central assessment of objective response, duration of response, and clinical benefit showed consistent results (table 2). Overall survival results were not mature at the data cutoff date: 55 (23%) of 244 patients in the tucidinostat group and 31 (26%) of 121 patients in the placebo group died. Patients and investigators will remain unaware of study assignments until survival results are mature for analysis.

The most common all-cause adverse events of any grade reported in the tucidinostat group were haematological toxic effects (table 3). Neutropenia, leucopenia, thrombocytopenia, and anaemia. The most common grade 3 or 4 haematological adverse events in either group were neutropenia (124 [51%] of 244 patients in the tucidinostat group *vs* three [2%] of 121 patients in the placebo group), thrombocytopenia (67 [27%] *vs* three [2%]), and leucopenia (46 [19%] *vs* three [2%]). No febrile neutropenia was reported in either treatment group. Most haematological adverse events, such as anaemia and thrombocytopenia, were mostly asymptomatic and manageable by supportive care. Transfusions of red blood cells and platelets were given to one patient in the placebo group for grade 3 anaemia and grade 4 thrombocytopenia, respectively. No remarkable bleeding-related adverse events were reported. The most common non-haematological adverse events of any grade were hypokalaemia, hyperglycaemia, hypocalcaemia, and hypertriglyceridaemia, which were all reported more frequently in the tucidinostat group than in the placebo group (table 3). Gastrointestinal adverse events (including nausea, vomiting, and diarrhoea) and urinary tract infections also occurred more frequently in the tucidinostat group than in the placebo group. Grade 3 or 4 non-haematological adverse events that occurred in more than 2% of patients in the tucidinostat group were hypokalaemia (15 [6%] of 244 patients *vs* one [1%] of 121 patients in the placebo group), hypertriglyceridaemia (12 [5%] *vs* none), increased γ-glutamyl transferase (11 [5%] *vs* three [3%]), increased blood creatine phosphokinase (five [2%] *vs* none), hypophosphataemia (five [2%] *vs* none), hyperglycaemia (five [2%] *vs* none). All adverse events of any grade in both study groups are presented in the appendix (pp 5–16). Treatment-related adverse events are shown in the appendix (pp 17–24).

	Tucidinostat group (n=244)	Placebo group (n=121)	p value	Hazard ratio (95% CI)
Investigator assessment				
Progression-free survival				
Disease progression or death	162 (66%)	90 (74%)	0·033	0·75 (0·58–0·98)
Kaplan-Meier median, months	7·4 (5·5–9·2)	3·8 (3·7–5·5)	..	..
Best overall response				
Complete response	1 (<1%)	0	..	..
Partial response	44 (18%)	11 (9%)	..	..
Stable disease	136 (56%)	65 (54%)	..	..
Progressive disease	49 (20%)	43 (36%)	..	..
Not assessable or unknown	14 (6%)	2 (2%)	..	..
Objective response	45 (18% [14–23])	11 (9% [4–14])	0·026	..
Clinical benefit	114 (47% [41–53])	43 (36% [27–44])	0·034	..
Duration of response				
Disease progression or death after initial objective response	21/45 (47%)	3/11 (27%)	0·574	1·43 (0·41–4·93)
Kaplan-Meier median, months	12·9 (5·6–25·0)	NR (5·6–NE)	..	..
Independent review committee assessment				
Progression-free survival				
Disease progression or death	124 (51%)	71 (59%)	0·024	0·71 (0·53–0·96)
Kaplan-Meier median, months	9·2 (7·2–10·9)	3·8 (3·6–7·4)	..	..
Best overall response				
Complete response	0	0	..	..
Partial response	40 (16%)	9 (7%)	..	..
Stable disease	135 (55%)	66 (55%)	..	..
Progressive disease	53 (22%)	42 (35%)	..	..
Not assessable or unknown	16 (7%)	4 (3%)	..	..
Objective response	40 (16% [12–21])	9 (7% [3–12])	0·024	..
Clinical benefit	104 (43% [36–49])	37 (31% [22–39])	0·020	..
Duration of response				
Disease progression or death after initial objective response	17/40 (44%)	3/9 (33%)	0·997	1·00 (0·28–3·57)
Kaplan-Meier median, months	11·1 (7·4–NE)	NR (1·9–NE)	..	..

Data are n (%), median (95% CI), n (% [95% CI]), or n/N (%). NR=not reached. NE=not estimable.

Table 2: **Efficacy analysis on the basis of investigator and independent review committee assessment**

Serious adverse events of any cause occurred in 51 (21%) of 244 patients in the tucidinostat group and seven (6%) of 121 patients in the placebo group (appendix pp 25–27). No serious adverse event occurred in more than 2% of the patients in either group (appendix pp 25–27). 24 (10%) of 244 patients in the tucidinostat group, and two (2%) of 121 patients in the placebo group, had at least one treatment-related serious adverse event (appendix p 28). No treatment-related deaths were reported in the study. Three deaths occurred in the tucidinostat group during or within 30 days of study treatment because of progression of breast cancer.

The most frequent reason for treatment discontinuation was disease progression, which occurred in 156 (64%) of

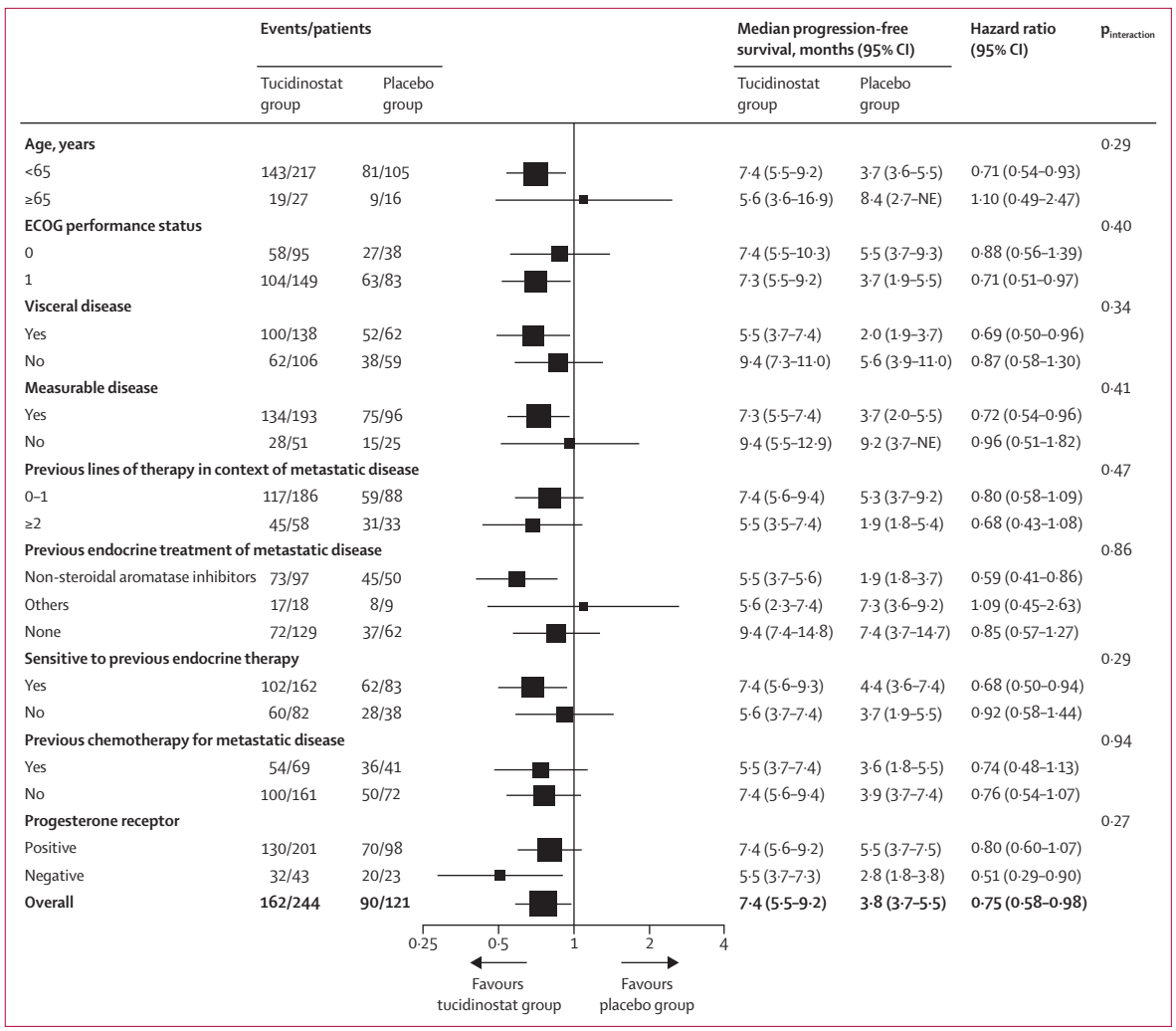

Figure 3: **Analysis of progression-free survival by subgroup**
The size of the effect squares is proportional to the population of the subgroup. ECOG=Eastern Cooperative Oncology Group. NE=not estimable.

244 patients in the tucidinostat group and 89 (74%) of 121 patients in the placebo group. Dose reductions because of adverse events occurred in 81 (33%) patients in the tucidinostat group and three (2%) in the placebo group, 45 (18%) and two (2%), respectively, because of haematological adverse events. 19 (8%) patients in the tucidinostat group and none in the placebo group discontinued treatment because of adverse events. Adverse events that led to treatment discontinuations from any cause in the tucidinostat group are presented in the appendix (p 29).

Discussion

In the prospective, multicentre, phase 3 ACE trial, tucidinostat plus exemestane was associated with improved progression-free survival compared with placebo plus exemestane in a population of patients with hormone receptor-positive metastatic breast cancer who had been exposed to previous endocrine therapy. Investigator-assessed improvements in progression-free survival benefit were confirmed with blinded central imaging assessment. Our findings are consistent with our previously reported results from an exploratory trial in patients with hormone receptor-positive advanced disease treated with tucidinostat and exemestane,[12] in which median progression-free survival was 7·6 months (IQR 3·2–11·3) and four (20%) of 20 participants had an objective response.

Primary and acquired resistance to endocrine therapies is a major challenge in the treatment of hormone receptor-positive breast cancer. To overcome this challenge, combination of targeted drugs with endocrine therapies has emerged as an important strategy in patients with advanced hormone receptor-positive breast cancer in the past 7 years, including inhibitors of PI3 kinase–mTOR (everolimus),[14] and inhibitors of CDK4 and CDK6 (palbociclib,[15] abemaciclib,[16] and ribociclib[17]). However, much remains unknown, such as

	Tucidinostat group (n=244)				Placebo group (n=121)			
	Grade 1–2	Grade 3	Grade 4	Grade 5	Grade 1–2	Grade 3	Grade 4	Grade 5
Any*	55 (23%)	143 (59%)	38 (16%)	3 (1%)†	88 (73%)	15 (12%)	5 (4%)	0
Neutropenia	75 (31%)	102 (42%)	22 (9%)	0	28 (23%)	1 (1%)	2 (2%)	0
Leucopenia	148 (61%)	45 (18%)	1 (<1%)	0	28 (23%)	1 (1%)	2 (2%)	0
Thrombocytopenia	116 (48%)	61 (25%)	6 (2%)	0	13 (11%)	1 (1%)	2 (2%)	0
Anaemia	69 (28%)	9 (4%)	0	0	20 (17%)	2 (2%)	0	0
Hyperglycaemia	55 (23%)	5 (2%)	0	0	17 (14%)	0	0	0
Increased aspartate aminotransferase	50 (20%)	0	0	0	20 (17%)	4 (3%)	0	0
Hypertriglyceridaemia	45 (18%)	10 (4%)	2 (1%)	0	15 (12%)	0	0	0
Increased alanine aminotransferase	49 (20%)	0	0	0	18 (15%)	2 (2%)	0	0
Nausea	60 (25%)	1 (<1%)	0	0	7 (6%)	0	0	0
Hypokalaemia	47 (19%)	14 (6%)	1 (<1%)	0	2 (2%)	1 (1%)	0	0
Diarrhoea	49 (20%)	4 (2%)	0	0	9 (7%)	0	0	0
Hypocalcaemia	56 (23%)	2 (1%)	0	0	3 (2%)	0	0	0
Increased γ-glutamyl transferase	32 (13%)	11 (5%)	0	0	11 (9%)	1 (1%)	2 (2%)	0
Anorexia	42 (17%)	3 (1%)	0	0	11 (9%)	0	0	0
Urinary tract infection	46 (19%)	1 (<1%)	0	0	5 (4%)	0	0	0
Cough	33 (14%)	2 (1%)	0	0	15 (12%)	0	0	0
Fatigue	34 (14%)	1 (<1%)	0	0	13 (11%)	0	0	0
Hypoalbuminaemia	33 (14%)	0	0	0	5 (4%)	0	0	0
Vomiting	34 (14%)	1 (<1%)	0	0	3 (2%)	0	0	0
Upper respiratory tract infection	26 (11%)	1 (<1%)	0	0	7 (6%)	1 (1%)	0	0
Weight loss	27 (11%)	2 (1%)	0	0	5 (4%)	0	0	0
Fever	25 (10%)	0	0	0	7 (6%)	0	0	0
Increase blood creatine phosphokinase	20 (8%)	4 (2%)	1 (<1%)	0	2 (2%)	0	0	0
Hypophosphataemia	10 (4%)	4 (2%)	1 (<1%)	0	0	0	0	0

Data are n (%). Grade 1–2 adverse events that occurred in at least 10% of patients in either study group, and all grade 3, 4, and 5 adverse events that occurred in at least 2% of patients in either study group are listed. For any listed adverse event, each patient was counted according to the worst severity grade. All adverse events were coded according to the Medical Dictionary for Regulatory Activities. A list of all adverse events that occurred is in the appendix (pp 5–16). *Each patient was counted only once according to the worst severity grade of all the adverse events during the study. †Deaths were due to dyspnoea, interstitial pneumonia, and sudden death (possibly due to pulmonary embolism); these events were judged by investigators to be related not to study drug but rather to disease progression.

Table 3: **Adverse events from any cause**

the mechanisms of resistance, the patients most likely to respond, and when best to integrate these therapies into treatment plans. Furthermore, in China, palbociclib is the only CDK4 and CDK6 inhibitor available (approved in July, 2018), and there is a huge unmet need for novel effective therapies in the vast patient population with primary and acquired resistance to endocrine treatment. Thus, further investigation of new agents with different mechanisms of action is still very much needed.

Evidence suggests that altered epigenetic modifications could contribute to development of drug resistance and relapse of different types of cancer, including breast cancers. For instance, mutations of epigenetic modulating genes are frequently noted in recurrent breast cancer lesions, and phenotypic or functional changes driven by epigenetic mechanisms, particularly the status of histone modifications and chromatin accessibility, contribute to progression and resistance to endocrine treatment in oestrogen receptor-positive breast cancer cell lines[18,19] and patients.[20,21] A randomised phase 2 trial[22] showed that the addition of entinostat, a benzamide class of HDAC subtype-selective inhibitor, to exemestane did not improve progression-free survival, but significantly improved overall survival compared with placebo in patients with hormone receptor-positive metastatic breast cancer that had relapsed or progressed after previous endocrine therapy. Our results lend support to evidence that aberrant epigenetic mechanisms can lead to disease progression and resistance to endocrine treatment in breast cancers. Selective targeting of specific HDAC subtypes could thus be a feasible therapeutic strategy across diverse epigenetic mechanisms of acquired resistance to endocrine therapy in advanced breast cancer. Either the steroidal aromatase inhibitor

exemestane or the selective oestrogen receptor degrader fulvestrant could be given as single-agent endocrine therapy after treatment failure with non-steroidal aromatase inhibitors.[23,24] Although data do not strongly support exemestane monotherapy as the most active option in patients pretreated with non-steroidal aromatase inhibitors, the results from our study suggest that a tucidinostat plus exemestane regimen improves progression-free survival in this patient population.

We used a 2:1 allocation ratio in this trial so that the safety of tucidinostat could be assessed in more patients. Haematological adverse events were the most commonly reported adverse events in the tucidinostat plus exemestane group, consistent with the reported safety profile of tucidinostat and other HDAC inhibitors.[7,22,25,26] Although the incidence of neutropenia of any grade in the tucidinostat plus exemestane group in our study was higher than that previously reported for tucidinostat monotherapy, no cases of febrile neutropenia were reported. Other haematological adverse events were mostly asymptomatic and manageable. In terms of non-haematological adverse events, a higher incidence of gastrointestinal adverse events (which are commonly observed with HDAC inhibitors), electrolyte disturbances (such as hypokalaemia and hypocalcaemia) and glucose and fat metabolism alterations (such as hyperglycaemia and hypertriglyceridaemia) was noted in the tucidinostat group compared with the placebo group. Electrolyte disturbances have previously been reported with other HDAC inhibitors,[25] which might be related to the gastrointestinal toxic effects associated with the drug class. HDAC inhibitors interact with the transcription complex of metabolism regulation,[27] but whether or not this mechanism contributed to the high incidence of metabolic adverse events noted in the tucidinostat group has yet to be established. Most non-haematological adverse events in our study were mild and tolerable. 8% of patients in the tucidinostat group discontinued treatment because of adverse events compared with none in the placebo group. The longer duration of treatment in the tucidinostat group could have contributed to this discrepancy in the frequency of adverse event-related discontinuation between groups.

The incidences of symptomatic adverse events in some categories, such as gastrointestinal symptoms, fatigue, and headache, in the placebo plus exemestane group in our study was lower than those in trials with similar study populations given exemestane plus placebo, including BOLERO-2,[14,28] EFFECT,[23] and SoFEA.[24] Although to precisely interpret the reasons for these differences would be difficult, we have identified several issues that could have had a role. Post-hoc analysis of BOLERO-2 showed that symptomatic adverse events were reported less frequently in Asian patients than in non-Asian groups.[29] Second, patients in our trial were younger (median age 55 years) than those in the other three trials (median age 61 years in BOLERO-2, 63 years in EFFECT, and 66 years in SoFEA). Third, the median follow-up duration in our trial (13·9 months) was shorter than that reported for BOLERO-2 (17·7 months). All these factors could have contributed to the different adverse event profiles noted in our study.

Our study had some limitations. The study was done in a single racial group, which could limit the generalisability of our results. For instance, the postmenopausal patients enrolled in this study were considerably younger than those enrolled in many trials done in western countries, consistent with the fact that the mean age at diagnosis of breast cancer in China (45–55 years)[30] is lower than that in western countries. Quality of life was not assessed in this study, which could have resulted in symptomatic adverse events being under-reported in the study. The sample size and event number meant that the power for analysis of progression-free survival by subgroup was insufficient. Overall survival data for the study are still unavailable, pending further follow-up.

In conclusion, to our knowledge, our study is the first phase 3 trial to show that epigenetic modulation plus endocrine blockade is a feasible and adequately tolerated strategy in patients with advanced, hormone receptor-positive, HER2-negative breast cancer that has progressed after previous endocrine therapy. Tucidinostat plus exemestane could be a new treatment option in this patient population.

Contributors

ZJ, ZN, and XL conceived and designed the study and analysed the data. ZJ and ZN wrote the first draft of the Article, with input from MC. ZJ, WL, XH, QZ, TS, SC, ShusW, QO, YY, CG, ZT, YC, YP, YS, HW, TO, KG, JF, XW, ShubW, TL, and JG recruited patients and gathered data. All authors reviewed the results, interpreted the data, contributed substantially to development of the Article, and reviewed and approved the final version for submission.

Declaration of interests

MC is a consultant for Pfizer, Novartis, CytoDyn, and Merus. ZN and XL are employees of Chipscreen Biosciences. All other authors declare no competing interests.

Acknowledgments

This study was partly supported by grants from the Chinese National Major Project for New Drug Innovation (2015ZX09101005) and the Shenzhen municipal science and technology project (JCYJ20120618162903087, JCYJ20160427185121156). We thank all the patients, physicians, and nurses who participated in this study.

References

1 Osborne CK, Schiff R. Mechanisms of endocrine resistance in breast cancer. *Annu Rev Med* 2011; **62:** 233–47.

2 Saxena NK, Sharma D. Epigenetic reactivation of estrogen receptor: promising tools for restoring response to endocrine therapy. *Mol Cell Pharmacol* 2010; **2:** 191–202.

3 Brabletz T. To differentiate or not—routes towards metastasis. *Nat Rev Cancer* 2012; **12:** 425–36.

4 Falkenberg KJ, Johnstone RW. Histone deacetylases and their inhibitors in cancer, neurological diseases and immune disorders. *Nat Rev Drug Discov* 2014; **13:** 673–91.

5 Jones PA, Issa JP, Baylin S. Targeting the cancer epigenome for therapy. *Nat Rev Genet* 2016; **17:** 630–41.

6 Ning ZQ, Li ZB, Newman MJ, et al. Chidamide (CS055/HBI-8000): a new histone deacetylase inhibitor of the benzamide class with antitumour activity and the ability to enhance immune cell-mediated tumour cell cytotoxicity. *Cancer Chemother Pharmacol* 2012; **69:** 901–09.

7 Shi Y, Dong M, Hong X, et al. Results from a multicentre, open-label, pivotal phase II study of chidamide in relapsed or refractory peripheral T-cell lymphoma. *Ann Oncol* 2015; **26:** 1766–71.

8 Pan DS, Yang Q J, Fu X, et al. Discovery of an orally active subtype-selective HDAC inhibitor, chidamide, as an epigenetic modulator for cancer treatment. *Med Chem Commun* 2014; **5:** 1789–96.

9 Yao Y, Zhou J, Wang L, et al. Increased PRAME-specific CTL killing of acute myeloid leukemia cells by either a novel histone deacetylase inhibitor chidamide alone or combined treatment with decitabine. *PLoS One* 2013; **8:** e70522.

10 Shen L, Pili R. Class I histone deacetylase inhibition is a novel mechanism to target regulatory T cells in immunotherapy. *Oncoimmunology* 2012; **1:** 948–50.

11 Zhou Y, Wang YN, Zhang K, et al. Chidamide reverses epidermal growth factor induced endocrine resistance in estrogen receptor-positive breast cancer. *J Shenzhen Univ Sci Eng* 2018; **35:** 339–44.

12 Zhang Q, Wang T, Geng C, et al. Exploratory clinical study of chidamide, an oral subtype-selective histone deacetylase inhibitor, in combination with exemestane in hormone receptor-positive advanced breast cancer. *Chin J Cancer Res* 2018; **30:** 605–12.

13 Eisenhauer EA, Therasse P, Bogaerts J, et al. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). *Eur J Cancer* 2009; **45:** 228–47.

14 Baselga J, Campone M, Piccart M, et al. Everolimus in postmenopausal hormone-receptor-positive advanced breast cancer. *N Engl J Med* 2012; **366:** 520–29.

15 Turner NC, Ro J, André F, et al. Palbociclib in hormone-receptor-positive advanced breast cancer. *N Engl J Med* 2015; **373:** 209–19.

16 Goetz MP, Toi M, Campone M, et al. MONARCH 3: abemaciclib as initial therapy for advanced breast cancer. *J Clin Oncol* 2017; **35:** 3638–46.

17 Hortobagyi GN, Stemmer SM, Burris HA, et al. Ribociclib as first-line therapy for HR-positive, advanced breast cancer. *N Engl J Med* 2016; **375:** 1738–48.

18 Légaré S, Basik M. Minireview: the link between ERα corepressors and histone deacetylases in tamoxifen resistance in breast cancer. *Mol Endocrinol* 2016; **30:** 965–76.

19 Sabnis GJ, Goloubeva OG, Kazi AA, et al. HDAC inhibitor entinostat restores responsiveness of letrozole-resistant MCF-7Ca xenografts to aromatase inhibitors through modulation of Her-2. *Mol Cancer Ther* 2013; **12:** 2804–16.

20 Patten DK, Corleone G, Győrffy B, et al. Enhancer mapping uncovers phenotypic heterogeneity and evolution in patients with luminal breast cancer. *Nat Med* 2018; **24:** 1469–80.

21 Yates LR, Knappskog S, Wedge D, et al. Genomic evolution of breast cancer metastasis and relapse. *Cancer Cell* 2017; **32:** 169–84.

22 Yardley DA, Ismail-Khan RR, Melichar B, et al. Randomized phase II, double-blind, placebo-controlled study of exemestane with or without entinostat in postmenopausal women with locally recurrent or metastatic estrogen receptor-positive breast cancer progressing on treatment with a nonsteroidal aromatase inhibitor. *J Clin Oncol* 2013; **31:** 2128–35.

23 Chia S, Gradishar W, Mauriac L, et al. Double-blind, randomized placebo controlled trial of fulvestrant compared with exemestane after prior nonsteroidal aromatase inhibitor therapy in postmenopausal women with hormone receptor-positive, advanced breast cancer: results from EFECT. *J Clin Oncol* 2008; **26:** 1664–70.

24 Johnston SR, Kilburn LS, Ellis P, et al. Fulvestrant plus anastrozole or placebo versus exemestane alone after progression on non-steroidal aromatase inhibitors in postmenopausal patients with hormone-receptor-positive locally advanced or metastatic breast cancer (SoFEA): a composite, multicentre, phase 3 randomised trial. *Lancet Oncol* 2013; **14:** 989–98.

25 Rasheed W, Bishton M, Johnstone RW, Prince HM. Histone deacetylase inhibitors in lymphoma and solid malignancies. *Expert Rev Anticancer Ther* 2008; **8:** 413–32.

26 Munster PN, Thurn KT, Thomas S, et al. A phase II study of the histone deacetylase inhibitor vorinostat combined with tamoxifen for the treatment of patients with hormone therapy-resistant breast cancer. *Br J Cancer* 2011; **104:** 1828–35.

27 Ye J. Improving insulin sensitivity with HDAC inhibitor. *Diabetes* 2013; **62:** 685–87.

28 Piccart M, Hortobagyi GN, Campone M, et al. Everolimus plus exemestane for hormone-receptor-positive, human epidermal growth factor receptor-2-negative advanced breast cancer: overall survival results from BOLERO-2. *Ann Oncol* 2014; **25:** 2357–62.

29 Noguchi S, Masuda N, Iwata H, et al. Efficacy of everolimus with exemestane versus exemestane alone in Asian patients with HER2-negative, hormone-receptor-positive breast cancer in BOLERO-2. *Breast Cancer* 2014; **21:** 703–14.

30 Fan L, Strasser-Weippl K, Li JJ, et al. Breast cancer in China. *Lancet Oncol* 2014; **15:** e279–89.