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Impact of Ki 67 and Progesterone receptor status on the outcome of metastatic breast cancer patients treated with cyclin dependant kinase inhibitors

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Abstract: Background: Breast cancer is the most common malignancy in females worldwide and is the second cause of cancer related mortality after lung cancer. Outcome of treatment of metastatic breast cancer is still controverse , up till now no clear data about effect of ki67 an PR status on treatment outcome. we aimed to determine the prognostic and predictive value of Ki67 and PR status among metastatic breast cancer patients on CDK4\6 inhibitor treatment. **Patients and Methods:** This observational prospective cohort study was conducted on 50 female patients with hormone- receptor-positive (HR+) and human epidermal growth factor receptor 2 (HER2)-negative metastatic breast cancer (BC) from January 2022 till May 2024 with median follow up period 19.88 months (10.89-28.09), treated and followed up at the Department of Medical Oncology, faculty of medicine, Zagazig University. Ki67 and progesterone receptor status were assessed by immunohistochemistry in primary tumor samples before starting the treatment. **Results:** Considering biobsy from primary tumour, IDC was the commonest pathological type (90.0%), Grade II was the commonest (64.0%) followed by Grade III (28.5%), ER positive was the commonest (93.0%), PR ≥ 20 % was (60.0%), Ki67 ≥ 14 % was (62.0%), The biobsy from metastatic site showed that, all the studied group were ER positive (100%), PR ≥ 20 % was (62%) and ki67 Low (<14%) in (48%) of the studied group, Concerning survival rate of the whole group, the median follow up was 19.88 months ranging (10.89-28.09) months. The 5-years OS rate was (95.5%) with the median not reached. There was statistically significant relation between PR and KI67 status and 5 years OS rate. PR ≥20% was associated with statistically significant longer OS rate at 5 years (100% vs 87.5%) (p=0.009) and the median survival estimate not reached. Ki67 ≥14% had marginally significant shorter OS rate at 5 years than those with Ki67 <14% (91.3% vs 100%) and the median not reached(p=.026). As regard PFS, the median survival was 10.56 months and 8-month PFS rate was 88 %, and 32% of patients remained progression-free at 12 months as CDK4/6 inhibitors was not used in first line treatment in this study. There was statistically significant correlation between PR, KI67(proliferation index) and HER-2 status.. PR ≥20% was associated with statistically significant longer PFS rate at 12 months (45.2% vs 10.5%) and the median survival estimate was (11.23 vs 10.50) (p 0.032). Also, KI67<14% was associated with statistically highly significant longer PFS rate at 12 months (58.3% vs 7.7%) and the median survival estimate was (10.36 vs 12.43) (p < 0.001). **Conclusion:** Outcome of treatment of metastatic breast cancer is still controverse , Ki-67 and progesterone receptor status may be reliable predictors of response to CDK4/6 inhibitors in metastatic breast cancer, highlighting the need for further research to identify robust biomarkers. Alternative predictive biomarkers and patient stratification strategies are therefore crucial for optimizing treatment selection and improving outcomes.

Keywords: *Ki 67, Progesterone receptor, metastatic breast cancer, cyclin dependant kinase inhibitors*

Introduction.

Breast cancer is the most common women's cancer worldwide, comprising 25% of total new cases diagnosed in 2018, and is the second leading cause of cancer-related deaths. In Egypt, it is the most common cancer among females and constitutes 29% of National Cancer Institute cases **(1)**

Metastatic breast cancer (MBC) is an incurable disease, but survival improvements have been reported with appropriate therapeutic strategies **(2).**

Although metastatic hormone receptor positive (HR+) Human epidermal growth factor receptor 2 negative (HER2 –ve) breast cancer has a favorable prognosis with respect to other BC subtypes, the outcome of patients with metastatic disease remains poor. **(3).**

Hormone therapy is the main treatment for these patients and is routinely used in the adjuvant setting but also recommended for patients with advanced disease. However, primary resistance to hormone therapy occurs in around 20% of cases and virtually all patients eventually develop secondary resistance **(4).**

In the past decade, randomized controlled trials have led to the introduction of several innovative therapeutic strategies into clinical practice consisting of new targeted therapies combined with hormone treatments for both endocrine-sensitive and endocrine-resistant metastatic breast cancer **(3)**

Cyclin dependant kinase 4\6 inhibitor (CDK4/6i) are a class of small molecule-targeted drugs that selectively inhibit CDK4 and CDK6, the regulators that advance the cell cycle from G1 to S phase. The combination of CDK4 or CDK6 with cyclin D1 forms a cyclin D1–CDK4/6 complex that phosphorylates retinoblastoma protein (pRb), releasing the transcription factor E2F and leading to the transcription of S-phase specific genes and cell cycle progression. Blocking this cyclin D1–CDK4/6-pRb signaling pathway enables CDK4/6i to arrest the proliferation of tumor cells **(5)**

In human breast cancer, the subtype for which CDK4/6 inhibition has the strongest rationale is estrogen receptor (ER)-positive disease. Pre clinically, strong synergy has been observed when CDK4/6 inhibitors are added to standard anti-estrogen therapies, and large, randomized clinical trials have confirmed that the addition of CDK4/6 inhibitors to hormonal therapy is a valuable clinical approach in metastatic breast cancer. Three CDK4/6 inhibitors have now been approved by the FDA for the treatment of ER-positive metastatic breast cancer: palbociclib, ribociclib and abemaciclib. The addition of these agents to endocrine therapy has resulted in the longest improvement in progression-free survival (PFS) seen to date in this subtype of breast cancer **(6).**

We aimed to determine the prognostic and predictive value of Ki67 and PR status among metastatic breast cancer patients on CDK4\6 inhibitor treatment.

Patients and Methods

This study is a prospective cohort study that aimed to determine the prognostic and predictive value of Ki67 and PR status among HR positive \ HER2 negative metastatic breast cancer patients on CDK4\6 inhibitor treatment at time of presentation. Assuming all cases fulfill the inclusion and exclusion criteria included in the study. During the study period 50 cases included as a comprehensive sample from January 2022 till May 2024 who admitted, treated and follow up in Medical Oncology Department, Zagazig University.

Inclusion criteria: Female patients with metastatic breast cancer, Age ≥ 18 years old, Eastern Cooperative Oncology Group (ECOG) performance status ≤ 3, Metastatic HR+/HER2− breast cancer patients eligible for treatment with CDK4\6 inhibitor, women of childbearing period, a serum pregnancy test must be done.

Exclusion criteria: Patients diagnosed with malignancy other than breast cancer, Metastatic breast cancer patients indicated for chemotherapy treatment, Known pregnancy or lactating female patient, Early breast cancer, also Male breast cancer.

The following variables of patients medical condition followed up, reviewed and documented in medical records then will be transcribed into an Excel spreadsheet:

1. Personal data, Medical history (ex. Diabetic, Hypertensive) and clinical examination.
2. Demographic features including age & residence, date of diagnosis with breast cancer, detailed history and full physical examination at time of the diagnosis and at every follow up visit during CDK4\6 inhibitor administration.
3. Eastern Cooperative Oncology Group (ECOG) performance status at the time of starting therapy.
4. Calculation of Body mass index (BMI=kg/m2) for all patients in the study.
5. All participants in this study will be subjected to baseline complete blood picture, liver function tests: Serum alanine and aspartate amino-transferases (ALT and AST respectively), serum bilirubin (total and direct), serum albumin, serum creatinine, tumour marker (CA 15-3) electrocardiogram (ECG) , pulmonary function test and blood urea.
6. Ki67 and progesterone receptor status were asssed by immunohistochemistry in primary tumour samples before starting the treatment.

Diagnosis of metastatic breast cancer

Patients with newly diagnosed or recurrent MBC should have a biopsy, if technically feasible, to confirm histology and to re-assess estrogen receptor (ER), progesterone receptor (PgR), KI 67 and human epidermal growth factor receptor 2 (HER2) status , The standard work-up for staging includes computed tomography (CT) of the chest and abdomen and bone scintigraphy, [18F]2-fluoro-2-deoxy-D-glucose (18F-FDG) positron emission tomography (PET)\CT may be used instead of CT and bone scans. There is no evidence that any staging or monitoring approach provides an overall survival (OS) benefit over another. The imaging modality chosen at baseline should be applied for disease monitoring to ensure comparability. The interval between imaging and treatment start should be ≤ 4 weeks. **(13)**

Immunohistochemistry:

Estrogen (ER) and progesterone (PR) receptors: For the identification of BC, BC tissues are investigated for estrogen and progesterone receptors. Samples with 1% to 100% of tumor nuclei are considered as positive ER an PR. For reporting of ER(not PR) , if 1 to 10 % of tumor nuclei are immunoreactive , the sample is reported as low positive ER and if < 1% , it is considered negative for hormonal receptors. **(7).**

Ki67 and PR were analyzed both as continuous and dichotomized variables; i.e., low (<14%) *vs.* high (≥14%) for Ki67, low (<20%) *vs.* high (≥20%) for PR in accordance with the St. Gallen guidelines **(8).**

HER2 testing: positive HER2 is IHC +3 (staining > 30% of tumor cells), Low HER2 is IHC +1 or +2 but +2 need additional testing by ISH or FISH to detect positivity or negativity and negative HER2 is IHC 0. **(9)**.

By ISH, HER2 is considered positive when ratio of HER2 to CEP17 is >2.2 or average copy number> six signals per nucleus, and is considered negative ratio is <1.8 or average copy number < four signals per nucleus. **(10)**

Treatment regimen, evaluation of response, and adverse events

All patients received continuous oral treatment with CDK 4\6 inhibitors initially modified according to the adverse effects during the treatment follow-up

Follow-up evaluation schedules of treatment include the following:

Response evaluation was done every 3 months by clinical evaluation, subjective symptom evaluation, blood tests and repeating the initially abnormal radiological examinations with comparative measurements (in accordance with the conventional and modified Response Evaluation Criteria in solid Tumors (RECIST). However, the interval between assessments should be tailored to the clinical needs of the patient and to the aggressiveness of the disease. In the case of clinical suspicion of progressive disease appropriate tests (imaging and laboratory) should be performed irrespective of scheduled examinations, if necessary including areas not imaged in previous tests.

Serum tumour markers (CA 15-3) may be helpful in monitoring response, particularly in the case of not easily measurable disease, but not used as the only determinant for treatment decision.

Maintenance of a good quality of life was paramount and best achieved with prompt amelioration of symptoms and side-effects of treatment.

Dose reduction allowed in cases of drug-related grade 3 or 4 toxicities. The toxicity grade assessed before each treatment cycle using the National Cancer Institute Common Toxicity Criteria for Adverse Events (version 4.0) **(2)**

End points

Primary:

Progression free survival (PFS) and overall survival (OS) in relation to PR and KI67 status

Progression free survival (PFS): The time elapsed between treatment initiation and tumor progression or death from any cause, with censoring of patients who are lost to follow-up **(11)**

Secondary:

Toxicity profile related to CDK4\6i and overall survival (OS) in relation to PR and KI67 status

Overall survival (OS): the time from date of diagnosis to the date of death due to any cause, or to the date of censoring at the last time the subject was known to be alive**.(12)**

Protection of privacy and confidentiality of patient's information

Data collection and presentation were anonymous and both privacy and confidentiality were protected to the maximum possible standards.

Statistical analysis

Data were analyzed using SPSS win statistical package version 24. Numerical data was expressed as mean and standard deviation (SD), median, range as appropriate. Qualitative data was expressed as frequency and percentage. Survival analysis was done using Kaplan-Meier method. Comparison between two survival curves was done using log rank test. P value less than or equal to 0.05 will be considered significant. All tests were two tailed. Overall survival (OS) calculated from date of diagnosis until date of death or last follow up. Progression free survival calculated from date of metastasis until date of progression, death or last follow up.

Results

***Socio-demographic data and clinical history of the studied group*:**

Table (1); Socio-demographic data and clinical history of the studied group

Demographic data		Studied group N =50 %
Age (years)	Mean ± SD	56.00±7.24
	Median	55
	Range	(40-71)
BMI	Mean ± SD	36.44 ±7.14
	Median	35.5
	Range	(26-46)
Residence	Urban	18(36%)
	Rural	32(64%)
Marital status	Married	44(88%)
	Unmarried	6(12%)
Performance status	0	3(6%)
	1	15(30%)
	2	28(56%)
	3	4(8%)
Family history of breast cancer	No	46(92%)
	Yes	4(8%)
Menstrual history	Premenopausal	9 (18.0%)
	Postmenopausal	41(82%)
Comorbidities	Diabetes mellitus	9(18%)
	Hypertension	17(34%)
	HTN and DM	6(12%)

BMI= Body Mass Index, HTN= hypertension , D.M = diabetes mellitus.

Table (1) shows that the mean age of the studied group was (56.00±7.24) ranging from (40-71) years with the median age was 55 years , about two-thirds (64.0%) were rural resident, about (82%) of them were postmenopausal and (88%) of them were married. Regarding the clinical data, the mean BMI of the studied

group was (36.44 ±7.14) ranging from 26 to 46.0 and more than half of them (56.0%) had performance status 2, the majority (92.0%) didn't have family history, (34%.) had HTN and (18%) had DM.

Clinical and pathological features of the studied group

Table (2) Clinical and pathological features of the studied group

Pathology and markers of the primary tumor tissue		**The studied group (n=42)**
		No ≈ (%)
Pathology*	IDC	38 (90.0%)
	ILC	4 (9.0%)
Grading*	G 1	3 (6%)
	G II	27(64.0%)
	G III	12 (28.5%)
ER*	Negative	3 (7.0%)
	Positive	39 (93.0%)
PR*	< 20 %	17 (40.0%)
	≥ 20 %	25 (60.0%)
HER-2*	Negative (score 0)	15 (36.0%)
	Low	27 (64.0%)
Ki67*	Low (<14%)	16 (38.0%)
	High (≥14%)	26 (62.0%)
Tumor size (cT)*	TI	3(7.0%)
	T2	15 (35.0%)
	T3	19 (45.0%)
	T4	5 (11.0%)
Lymph node staging (cN)*	N0	2 (5.0%)
	N1	13 (31.0%)
	N2	17 (40.0%)
	N3	10 (24.0%)
Type of surgery*	MRM	35 (70.0%)
	BCS	7 (14.0%)
Site of breast mass *	Right	20 (48.0%)
	Left	22 (52.0%)
Enrollment classification (n=50)	Denovo metastatic	8 (16.0%)
	Recurrent metastatic	42 (84.0%)
ER (n=50)	Negative	0 (0.0%)
	Positive	50(100.0%)
PR (n=50)	< 20 %	19 (38.0%)
	≥ 20 %	31 (62.0%)
HER-2 (n=50)	Negative (score 0)	21 (42.0%)
	Low	29 (58.0%)
Ki67 (n=50)	Low (<14%)	24 (48.0%)
	High (≥14%)	26 (52.0%)
Metastatic sites (n=50)	Bone	36 (72.0%)
	Visceral disease	29 (58.0%)
	Soft tissue	6 (12.0%)
No. of organ metastasis (n=50)	≤2	23 (46.0%)
	>2	27 (54.0%)

IDC = invasive ductal carcinoma, ILC = invasive lobular carcinoma, ER=estrogen receptor, PR = progesterone receptor, HER2 = human epidermal receptor, IHC = immunohistochemistry, * = primary tumor, MRM=Modified radical mastectomy, BCS=breast conservative surgery

Table (2), Total number of the metastatic group was 50. Only 8 patients (16.0%) were denovo metastatic and 42 patients (84.0%) were recurrent metastatic breast cancer. As regard primary tumor tissue ,old data of the pathology was available and included that , IDC was the commonest pathological type (90.0%). Grade ll was the commonest (64.0%). ER positive was the commonest (93.0%), PR ≥ 20 % was (60.0%), Ki67 ≥ 14 % was (62.0%).

Regarding primary tumor size, T3 was the commonest (45.0%) followed by T2 (35.0%). As regard Lymph node metastasis, N2 was the commonest (40%). The majority of the recurrent group did modified radical mastectomy (70%) and (14%) did breast conservative surgery with nearly half of them on left side (52%)
At time of randomization, all the studied group (100.0%) were ER positive, PR ≥ 20 % was (62.0%), Ki67 ≥ 14 % was (52.0%), and HER-2 low positive was (58.0%).
Nearly half of the patients (54.0%) had more than two sites of metastasis with bone the most common site (72.0%)

Treatment modality of the primary tumor and metastatic breast cancer

Table 3 Treatment characteristics of the 50 recurrence and metastasis breast cancer patients

Treatment characteristics	Cases, n (%)
Treatment of the primary disease (No = 42)	
Adjuvant chemotherapy	
Anthracyclines + taxanes	36 (85.0%)
TC(taxotere\cyclophosphamide)	6 (14.0%))
Adjuvant endocrine therapy	
SERM	3 (7.14%)
SERM + AI	13 (30.9%)
AI	26 (61.9%)
Adjuvant radiotherapy	
No	0 (0%)
Yes	42 (100%)
Treatment of metastatic disease (No = 50)	
Chemotherapy	First line 21 (42.0%) Second line 0
Endocrine therapy	First line 29 (58.0%) Second line 21 (42.0%)
CDK4/6i treatment line	
1	0 (0.0%)
2	29 (58.0%)
≥3	21 (42.0%)
CDK4/6i treatment regimen	
CDK4\6 + AI	1 (2.0%)
CDK4\6 + fulvestrant	49 (98.0%)

SERM, selective estrogen receptor modulators; AI, aromatase inhibitors

Table(3), Regarding line of treatment, 42 patients received adjuvant treatment in the form of adjuvant chemotherapy(85 % recieved AC every 21 days for 4 cycle followed by weekly paclitaxel for 12 weeks and the remaining received TC for 4 cycle) followed by endocrine therapy (61% AI, 30.9% SERM+AI and 7.14% SERM) and radiotherapy.
In the metastatic setting, endocrine therapy was the commonest treatment used in first line (58.0%) followed by chemotherapy (42%) but CDK4\6i never used in the first line treatment in the study group only used as second or third line after progression on hormonal treatment.
In the second line, (42.0%) received endocrine therapy after chemotherapy and (58%) received CDK4\6i
Only one patient (2.0%) received CDK4\6 + AI who was progressed on adjuvant SERM and 49 patients (98%) received CDK4\6 + fulvestrant.

Side effect

Table (4) Toxicity profile among studied group

Toxicity		N	%
Hematological toxicity			
Neutropnia (n=33)	Grade 1	8	24.2
	Grade 2	20	60.6
	Grade 3	5	15.2
Anemia (n=13)	Grade 1	9	69
	Grade 2	4	30
Non hematological toxicity			
GIT(diarrhea)	Grade1	4	57
(n=7)	Grade2	3	43

Table (4) As regard hematological toxicity, neutropenia was the most common complication occurred in 33 patients (66.0%) and most of them was grade 2 (60.0%). Anemia occurred in 13 patients (26.0%) wich was grade 1&2 As regard nonhematological toxicity, diarrhea occurs in (14.0%) and was only grade 1&2.

Survival analysis among studied group and factors affecting it

Table (5): Overall survival rate and factors affecting it:

Variables	No	No of events	Median survival estimate	3 years OS rate	5 years OS rate	10 years OS rate	P value
OS	50	4	NR	100%	95.5%	91.6%	-
Age group							
>55	21	2	NR	100%	94.4%	86.6%	0.846
<=55	29	2		100%	96.2%	96.2%	
Menstrual history							
Premenopausal	9	0	NR	100%	100%	100%	0.379
Postmenopausal	41	4		100%	94.6%	90.1%	
PR							
<20%	19	4	NR	100%	87.5%	79.5%	0.009
≥20%	31	0		100%	100%	100%	
HER2 score							
Score 0	21	2	NR	100%	95.0%	86.4%	0.869
Low	29	2		100%	95.8%	95.8%	
KI67							
<14	24	0	NR	100%	100%	100%	0.026
≥14	26	4		100%	91.3%	83.0%	
Current treatment							
CDK 4\6 + AI	1	0	NR	100%	100%	100%	0.828
CDK 4\6+fasoldex	49	4		100%	95.3%	91.5%	

P value ≤0.5 Statistically significant. OS, overall survival.

P value ≤ 0.01 Statistically highly significant

This table shows that there was statistically significant relation between overall survival and progesterone receptor status and KI67

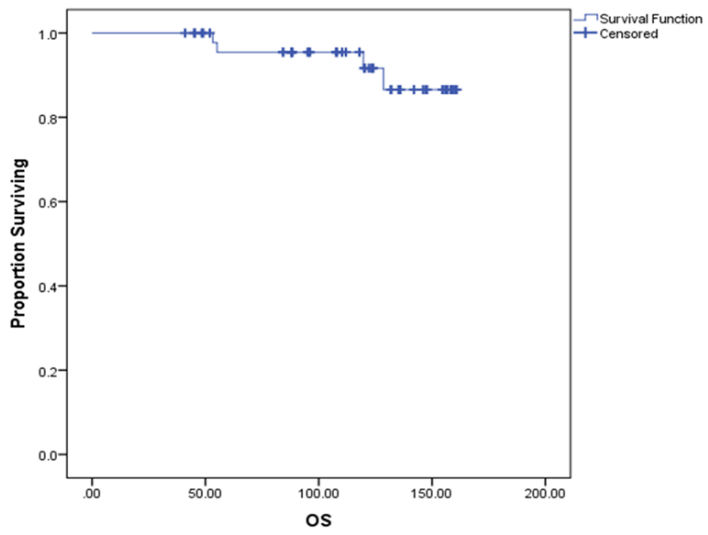
Table (6) Progression Free Survival (PFS) and factors affecting it:

Variables	No	No of events	Median survival estimate	8 months PFS rate	12 month PFS rate	15month PFS rate	P value
PFS	50	50	10.56	88.0%	32.0%	6.0%	-
Age group							
>55	21	21	10.56	86.0%	38.0%	4.0%	0.567
<=55	29	29	10.58	89.7%	27.6%	10.0%	
Menstrual history							
Premenopausal	9	9	9.98	77.8%	11.0%	0%	0.065
Postmenopausal	41	41	10.95	90.2%	34.1%	7.3%	
PR							
<20%	19	19	10.50	89.5%	10.5%	0%	**0.032**
≥20%	31	31	11.23	87.1%	45.2%	9.7%	
HER2 score							
score 0	21	21	11.25	85.7%	42.9%	14.3%	
Low	29	29	10.50	89.7%	24.1%	6.9%	0.041
KI67							
<14%	24	24	12.43	83.3%	58.3%	12.5%	**<0.001**
≥14%	26	26	10.36	92.3%	7.7%	3.8%	
Current treatment							
CDK 4\6 + AI	1	1	11.10	100%	0%	0%	0.870
CDK 4\6+fasoldex	49	49	10.56	87.8%	32.7%	6.1%	

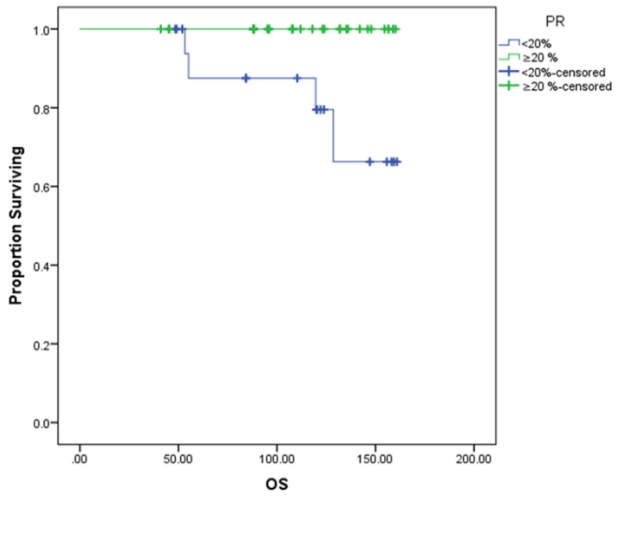
P value ≤0.5 Statistically significant. P value ≤ 0.01 Statistically highly significant

Table (6) show that there was statistically significant relation between progression free survival and progesterone receptor status, KI67 and HER-2 status.

After a median follow up period of 19.88 months (Range 10.89-28.09 months), the progression free survival (pFS) rate at 12 months was **32%** and the overall survival (OS) rate at 5 years was **95.5%** for the whole group of patients **(n=50).**



(A)



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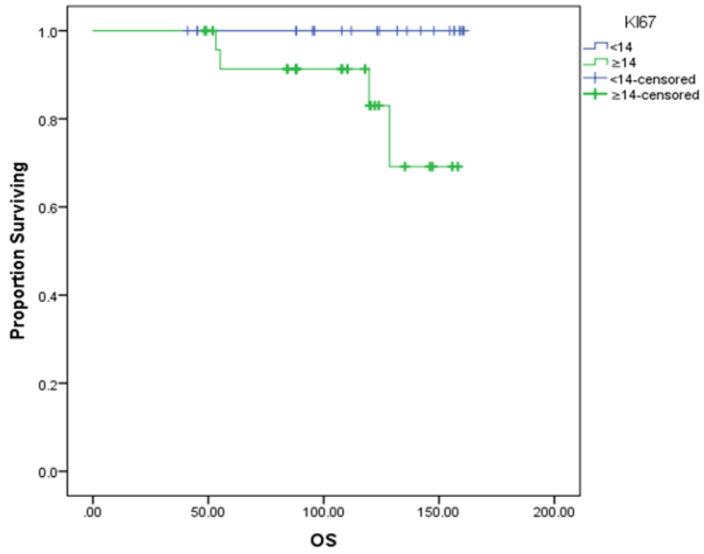
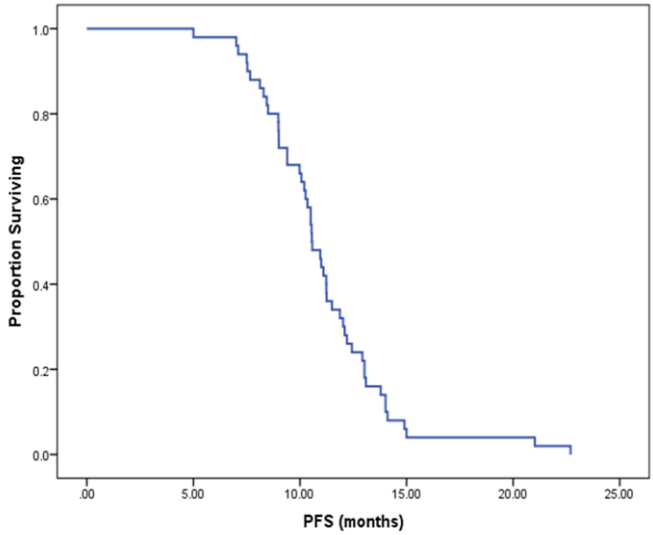
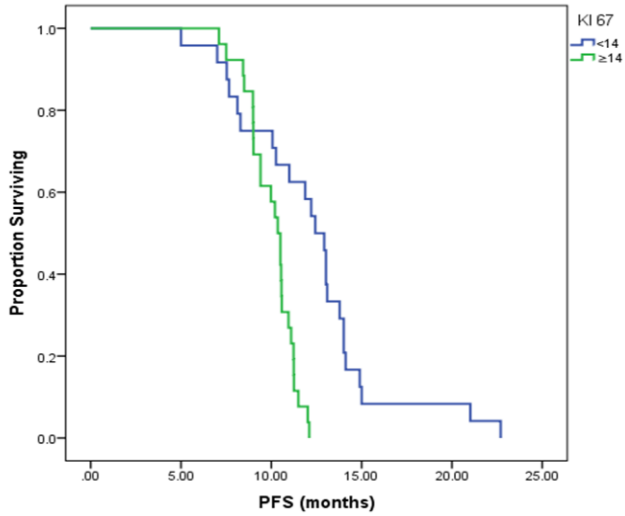

KI67
<14
≥14
<14-censored
≥14-censored
Proportion Surviving
OS

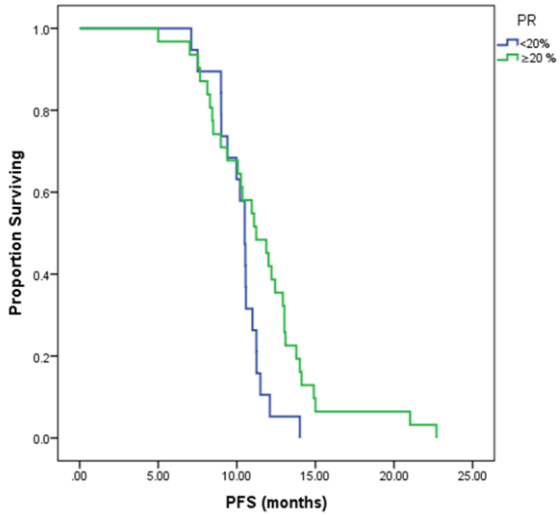
(C)


Proportion Surviving
PFS (months)

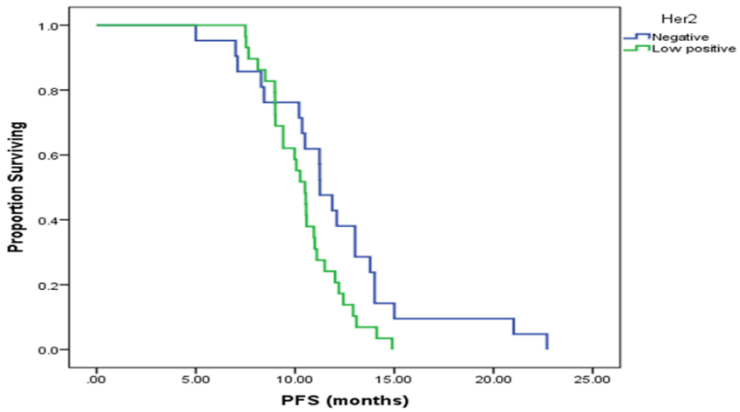
(D)


KI 67
<14
≥14
Proportion Surviving
PFS (months)

(E)


PR
<20%
≥20 %
Proportion Surviving
PFS (months)

(F)


Her2
Negative
Low positive
Proportion Surviving
PFS (months)

(G)

Graph (A): Represent OS among study group, (B) Comparison Between PR <20% and PR ≥20% as regard to OS among study group (p= 0.009), (C) Comparison between KI67 levels as regard to OS among study group (p= 0.026), (D) Progression Free Survival among study group. (E) Progression Free Survival among KI67 <14, ≥14 (p=0.001), (F) Progression Free Survival among PR <20, ≥20 (p=0.023), (G) Progression Free Survival among Her-2 (p=0.041)

Discussion

Resistance to endocrine therapy has been a major obstacle in the management of hormone receptor (HR)-positive metastatic breast cancer (MBC). Meanwhile, a number of treatments are available to such patients, and physicians often encounter difficulties in choosing the most appropriate treatments for individual patients. The combination of CDK 4/6 inhibitors (CDKi) and endocrine therapy has now become a standard treatment for HR-positive and human epidermal growth factor receptor 2 (HER2)-negative MBC. However, no predictive markers for CDKi-based treatments have been established. Considering their side effects and the financial burden on patients, identifying such markers is crucial. **(14)**

The main aim of the study was to evaluate predictive value of PR, KI67 and HER-2 status on sensitivity to CDK4/6 I

This observational prospective cohort study was conducted on 50 female patients with hormone- receptor-positive (HR+) and human epidermal growth factor receptor 2 (HER2)-negative metastatic breast cancer (BC) from January 2022 till May 2024 with median follow up period 19.88 months (10.89-28.09), treated and follow up at the Department of Medical Oncology, faculty of medicine, Zagazig University.

The current study showed that the mean **age** of the studied group was 56.00±7.24 ranging from (40 to 71) and the median is 55 years. Comparable with the current study **Zhong et al., (15)** showed that the mean age was 54.2 (11.3) years of females with HR+/HER-2 negative metastatic breast cancer received palbociclib plus ET as the first-line treatment for ABC**. Salmon et al., (16)** found that the median age of patient treated with CDK4\6i was 63 years ranging (31-89) years.

About (82%) of them were postmenopausal and (88%) of them were married about two-thirds (64.0%) were rural resident. **Salmon et al., (16)** found that 100% of patients were postmenopausal and **Finn et al., (17)** all patients included in PALOMA-2 trial were postmenopausal. In **PALOMA-3 trial, Iwata et al., (18)** include **(**42%) premenopausal and (58%) postmenopausal to determine efficacy and safety of fulvestrant with or without palbociclib in premenopausal and postmenopausal women with hormone receptor–positive, human epidermal growth factor receptor 2–negative metastatic breast cancer that progressed on prior endocrine therapy in Asian patients

Regarding the clinical data, the mean BMI of the studied group was (36.44 ±7.14) ranging from 26 to 46.0. **Sanchez-Covarrubias** who found significant association between obesity and the incidence of BC in a systematic review and meta-analysis.

More than half of them (56.0%) had performance status 2. **Islam et al., (19)** evaluated the patients with metastatic breast cancer for depression and anxiety and found that more than half (52.6%) of the patients had ECOG performance status grade II**.**

According to family history, 8 % of the studied cases had positive family history of BC, also **Won et al., (20)** in a nationwide study from Korea showed that there were 7% of women with BC have positive family history.

In this study (34%) and (18 %) of patients had comorbidities (DM, HTN respectively), however, **Ewertz et al., (21)** revealed that 16% of BC patients had comorbidities and the study showed that the risk of dying from all causes was significantly increased for all types of comorbidities, but the risk of dying from BC was significantly increased only for peripheral vascular disease, dementia, chronic pulmonary disease, liver, and renal diseases. **Anwar et al., (22)** found that metabolic comorbidities that include were frequently found in breast cancer patients and were associated with higher risks to develop recurrent metastatic disease, particularly in post-menopausal women.

Considering biopsy from primary tumor, IDC was the commonest pathological type (90.0%). This come in agreement of **Shao et al., (23)** who found that IDC was the commonest type in patent treated with CDK4\6i.
Grade II was the commonest (64.0%) followed by Grade III (28.5%). Also **Alves et al., (24)** found that most patient was grade II (50%) followed by grade III (36%). ER positive was the commonest (93.0%), PR ≥ 20 % was (60.0%), Ki67 ≥ 14 % was (62.0%). **Shao et al., (23)** founded that PR ≥ 20% in (53.5%) and and Ki67 ≥ 14% in more than half of the patients (52%).
Most of the studied group were recurrent metastatic disease (84%) while only (16%) were de novo metastatic breast cancer**.** Results from a reference center in Brazil to determine treatment outcomes in HR+ HER2- metastatic breast cancer patients treated with CDK4/6 inhibitors founded that (19%) were denovo metastatic breast cancer **(25)**.
The biopsy from metastatic site showed that, all the studied group were ER positive (100%), PR ≥ 20 % was (62%) and ki67 Low (<14%) in (48%) of the studied group. In agreement with our study **Palleschi et al., (3)** revealed that PR ≥ 20% was (61.2%) and ki67 low <14% was (57%). **Rocca et al., 2015(30)** reported discordance rates of 20 % for ER, 33 % for PgR, and 8 % for HER2 between the primary tumor and metastasis that affect choice of treatment.

Concerning survival rate of the whole group, the median follow up was 19.88 months ranging (10.89-28.09) months. The 5-years **OS** rate was (95.5%)(from start of the diagnosis as breast cancer till death or last follow up) with the median not reached. There was statistically significant relation between PR and KI67 status and 5 years OS rate. **PR ≥20%** was associated with statistically significant longer OS rate at 5 years (100% vs 87.5%) (p=0.009) and the median survival estimate not reached. **Ki67 ≥14**% had marginally significant shorter OS rate at 5 years than those with Ki67 <14% (91.3% vs 100%) and the median not reached(p=.026)
Inari et al., (26) found that high **Ki-67** immunohistochemical expression levels in distant metastatic lesions were independently associated with poorer overall survival outcomes that (hazard ratio 2.307; 95% confidence interval 1.207–4.407, *P* = 0.011).
Contrary to our study, A retrospective study included 314 patients with breast cancer who underwent various types of breast surgeries. Analyzed was done to find any possible correlation between the level of Ki67 and various patient and tumor characteristics and the survival rates.ki67 is correlated with the grade of the tumor, and is not a predictor for the survival of breast cancer patients. It may predict aggressive behavior of the tumor and higher histopathological grades. **(27)**
Another study conducted by **Xiao et al., (28)** found all patients with negative HER-2 status, the 5-year OS and BCSS rates of ER+/PR+ patients were the best (92.65% and 96.07%, respectively), followed by ER+/PR− patients (5-year OS: 85.40%, 5-year BCSS: 89.67%)
Also **Li et al., (29)** found that high expression of PR is more frequently observed in tumors with a better baseline prognosis (ie, luminal A) than tumors with a poor baseline prognosis (ie, luminal B). PR is an upregulated target gene of ER, its expression is dependent on estrogen, and PR can modulate ER action. PR is also a valuable prognostic biomarker of overall survival or disease-free survival (DFS) in breast cancer.
As regard **PFS**, the median survival was 10.56 months (median time on CDK4/6 I) months and 8-month PFS rate was 88 %, and 32% of patients remained progression-free at 12 months as CDK4/6 inhibitors was not used in 1st line treatment..
On the other hand, Witkiewicz et al., (31) showed that the progression-free survival (PFS) for patients treated with fulvestrant(FUL) combinations were 17.2 months. The difference in survival between different studies may be due to clinical variables, visceral involvement, prior endocrine therapy, recurrent disease and CDK4\6i treatment line of treatment as most study group use CDK4/6i as first line treatment.
PR ≥20% was associated with statistically significant longer PFS rate at 12 months (45.2% vs 10.5%) and the median survival estimate was (11.23 vs 10.50) (p=0.032). Also, **KI67<14%** was associated with statistically highly significant longer PFS rate at 12 months (58.3% vs 7.7%) and the median survival estimate was (10.36 vs 12.43) (p < 0.001). In agreement with the current study, **Shao et al., (23)** found that PR ≥20% was associated

with longer PFS (8.5 vs. 6.7 months, P=0.08) In the total population, patients with Ki67 ≥14% had marginally significant shorter PFS than those with Ki67 <14% [6.0 *vs.* 10.8 months; P=0.062]

In contrary to the current study, **Palleschi et al., (3)** found that PR, Ki67 did not significantly influence the PFS. The difference from the current study may be due to clinical variables, site and number of metastasis, prior endocrine therapy, recurrent disease and CDK4\6i treatment line.

Her-2 negative tumor (score 0) was associated with statistically significant longer PFS rate at 12 months (42.9% vs 24.1%) (p=0.041). **Guven & Sahin., (32)** found that the risk of progression and/or death was higher in patients with HER2-low tumors compared to HER2-zero (HR: 1.22, 95% CI 1.10–1.35, $p < 0.001$). In the pooled analysis of five studies, although the median follow-up was short, the risk of death was higher in the HER2-low group compared to the HER2-zero group (HR: 1.22, 95% CI 1.04–1.44, $p = 0.010$).

On the other hand**, Guliyev et al., (33)** found that the HER2-zero and low patients showed no statistically significant differences. This multicenter retrospective study included patients with HR + /HER2-negative MBC cancer who were treated with first-line CDK 4/6i in combination with ET. Patients were divided into two groups (HER2-low and zero), and survival and safety analyses were performed. The difference in result may be due to difference in clinicopathological features, site, number of metastasis, previous hormonal treatment and sensitivity.

In spite of this explicit prognostic information, Ki-67 measurement remains inconsistent and irreproducible between patients, limiting updates to current guidelines surrounding the routine inclusion of Ki-67 staining in standard breast cancer immuno-histochemical workup

Conclusion

Outcome of treatment of metastatic breast cancer is still controverse , Ki-67 and progesterone receptor status may be reliable predictors of response to CDK4/6 inhibitors in metastatic breast cancer, highlighting the need for further research to identify robust biomarkers. Alternative predictive biomarkers and patient stratification strategies are therefore crucial for optimizing treatment selection and improving outcomes.

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