cancers

MDPI

Article

Assessment of PI3K/mTOR/AKT Pathway Elements to Serve as Biomarkers and Therapeutic Targets in Penile Cancer

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Supplementary Materials

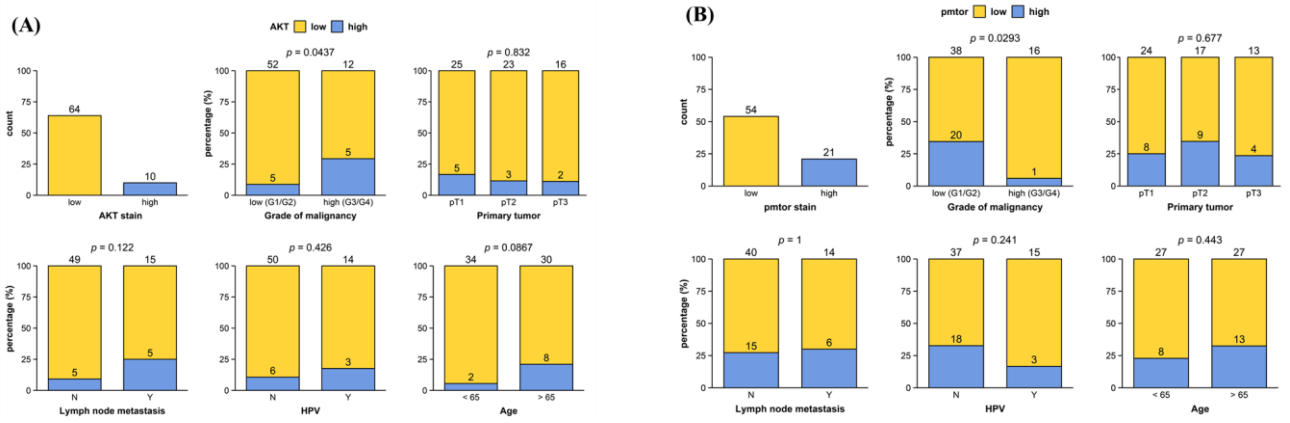

Figure S1. Dichotomized biomarker expressions of AKT (A) and pmTOR (B) correlated with clinical and histopathological data (grading, primary tumor, lymph node metastases, HPV infection and age). The Fischer's exact test was used for statistical analysis. High expression of AKT (p = 0.044) and low expression of pmTOR (p = 0.029) were associated with high-grade primary tumors

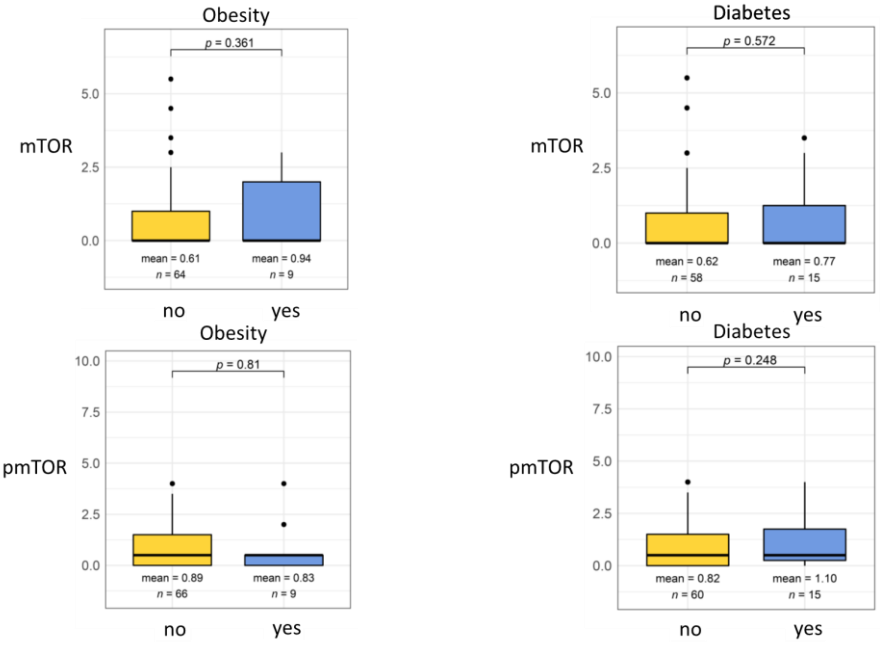

Figure S2. Expressions of mTOR and pmTOR correlated with clinical data (diabetes and obesity). *Y*-axis: immunoreactive score (IRS). *X*-axis: yellow boxplots—low expression of the respective biomarker, blue boxplots—high expression of the respective biomarker. Box (represents the interquartile range (IQR)): lower line—quartile Q1 (25%-quantile); middle line—median; upper line—quartile Q3 (75%-quantile); whisker—separate values lying outside the IQR; circles—outliers. The Fischer's exact test was used for statistical analysis.

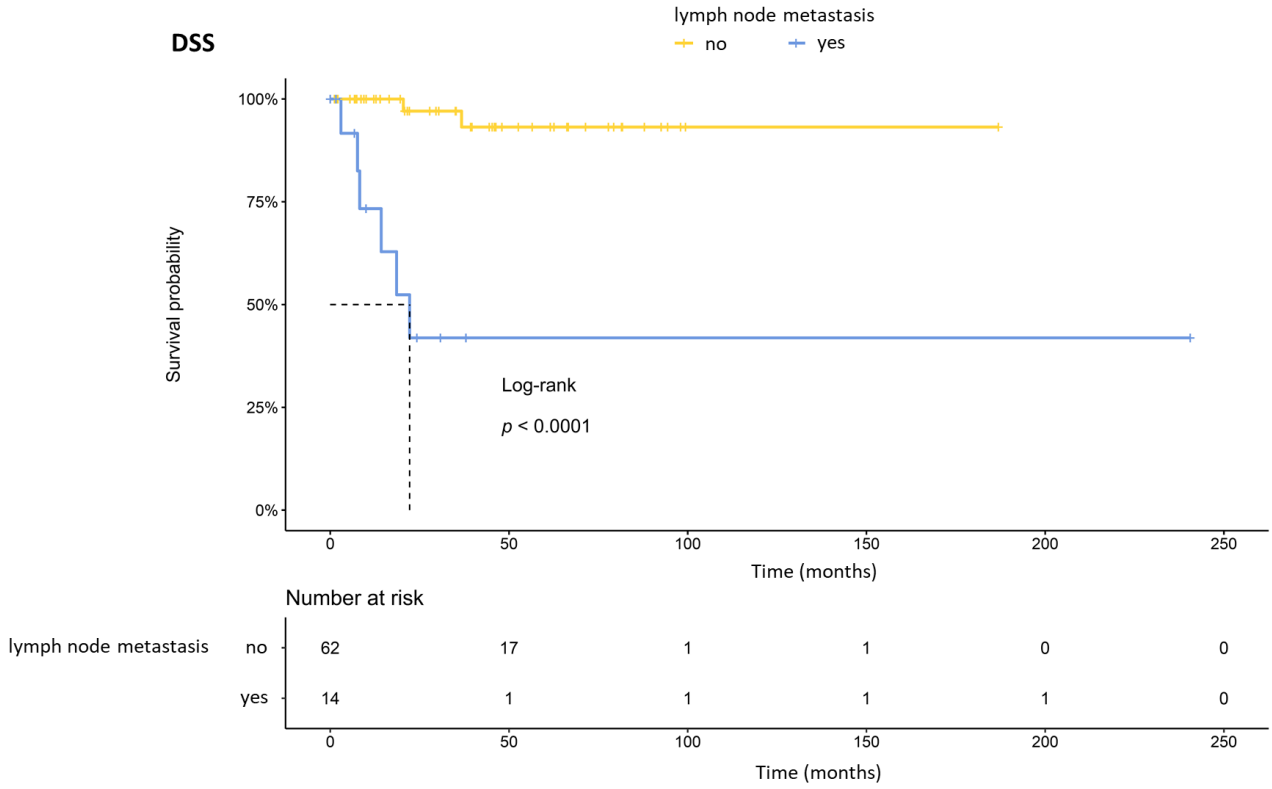

Figure S3. Univariate COX regression analysis: lymph node metastasis was associated with a shortened disease-specific survival (DSS) ($p < 0.0001$).

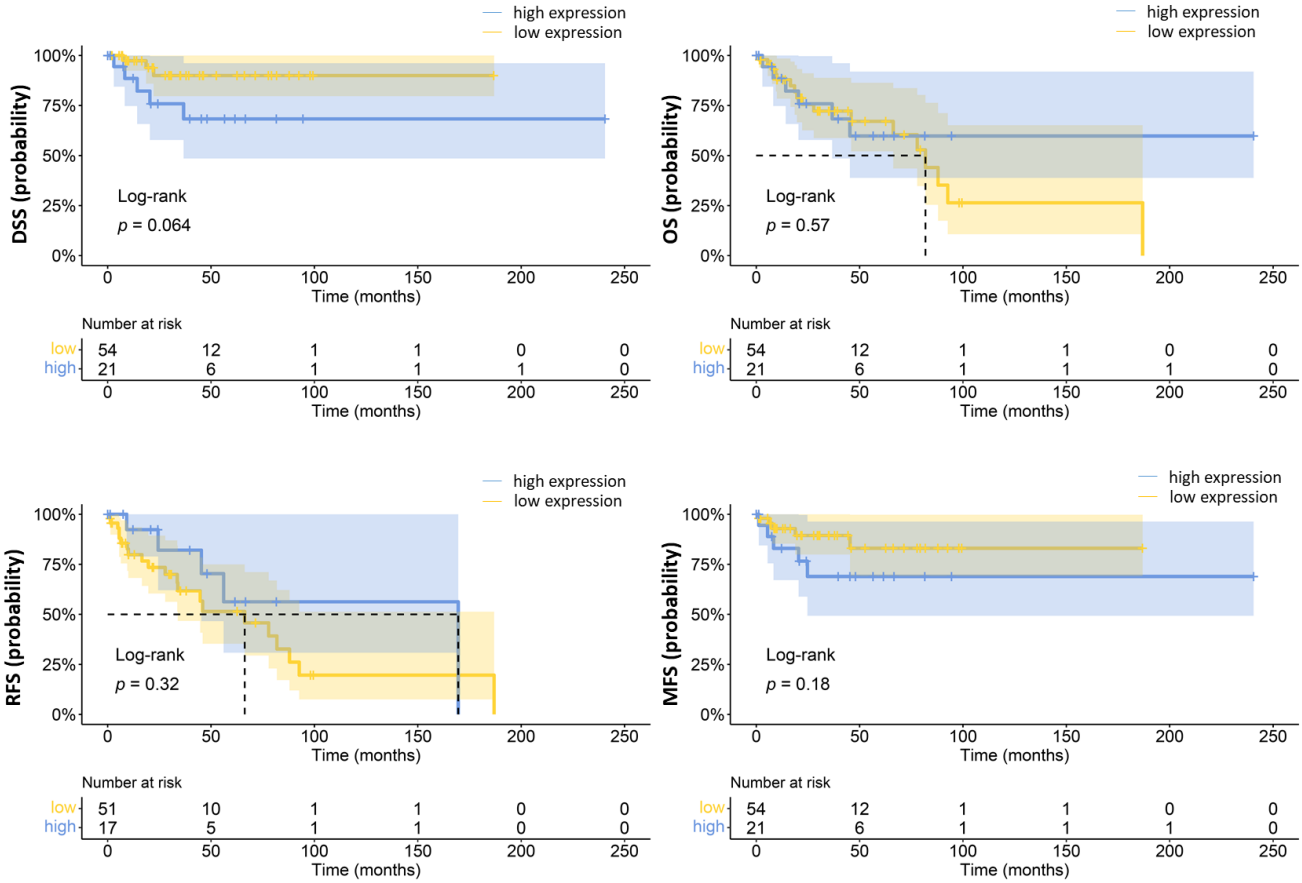

Figure S4. Kaplan–Meier Plots of disease-specific survival (DSS) (**A**), overall survival (OS) (**B**), recurrence-free survival (RFS) (**C**) and metastasis-free survival (MFS) (**D**) according to pmTOR expression.

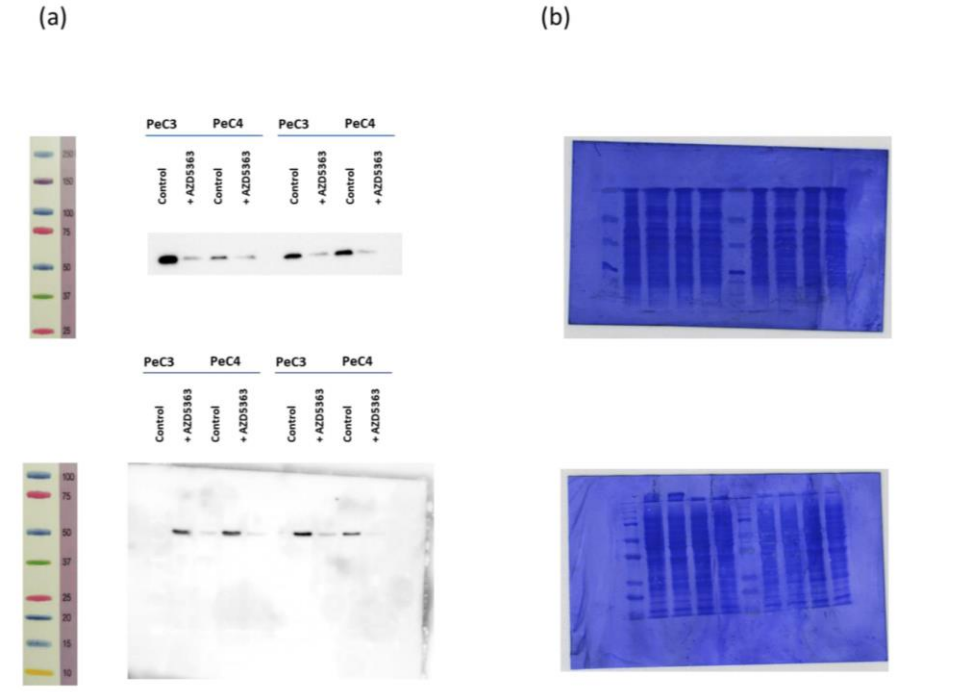

Figure S5. Protein expression profile in UKF-PeC-3 and UKF-PeC-4 cells without/after treatment with capivasertib. Protein expression of AKT (**a**), corresponding Coomassie blue staining of total protein (**b**).

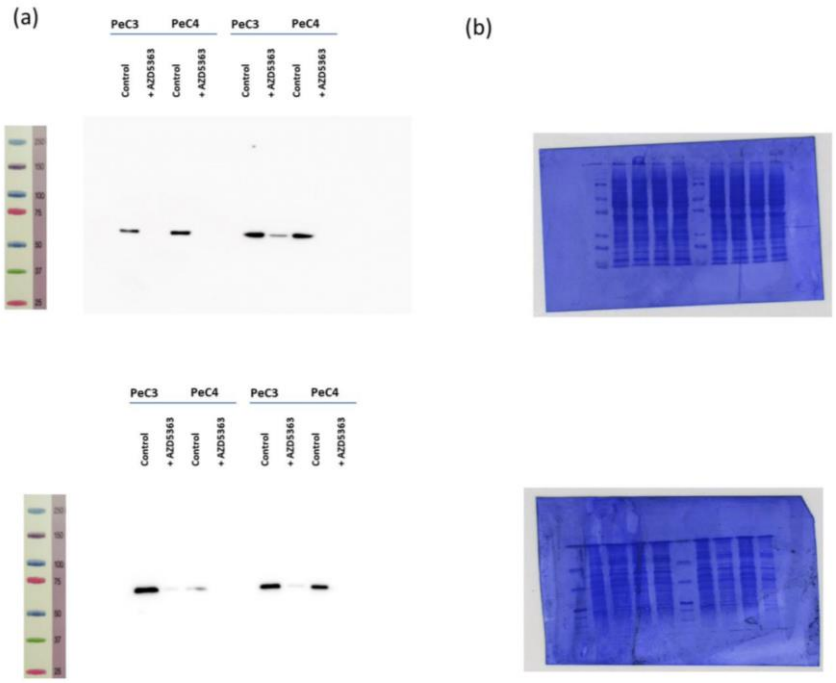

Figure S6. Protein expression profile in UKF-PeC-3 and UKF-PeC-4 cells without/after treatment with capivasertib. Protein expression of pAKT (**a**), corresponding Coomassie blue staining of total protein (**b**).

Table S1: Immunoreactive scores (IRS). IRS determined as cutoff levels (low and high) after graphically depicting the survival curves of each of the scores separately by receiver operating characteristic (ROC).

Biomarker	IRS cut-off
AKT	4,75
pAKT	0,75
pmTOR	1,25
pS6	1,25
p4EBP1	6,25
pPRAS	0,75
pp70S6K	0,25
mTOR	1,25
S6K1	0,25

Table S2. Immunohistochemical staining results with a respective group classification. Values in brackets: percentage from total number.

Biomarker	Overall (*n* = 76)
panAKT	
Low expression	64 (84.2%)
High expression	10 (13.2%)
Missing	2 (2.6%)
pAKT	
Low expression	20 (26.3%)

High expression	53 (69.7%)
Missing	3 (3.9%)
mTor	
Low expression	58 (76.3%)
High expression	15 (19.7%)
Missing	3 (3.9%)
pmTor	
Low expression	54 (71.1%)
High expression	21 (27.6%)
Missing	1 (1.3%)
pS6	
Low expression	23 (30.3%)
High expression	50 (65.8%)
Missing	3 (3.9%)
p4epb1	
Low expression	63 (82.9%)
High expression	10 (13.2%)
Missing	3 (3.9%)
pp70S6K	
Low expression	47 (61.8%)
High expression	26 (34.2%)
Missing	3 (3.9%)
s6k1	
Low expression	3 (3.9%)
High expression	44 (57.9%)
Missing	29 (38.2%)
pPRAS	
Low expression	71 (93.4%)
High expression	5 (6.6%)