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Evaluation of CEP55, SERPINE1 and SMPD3 genes and proteins as diagnostic and prognostic biomarkers in gastric carcinoma in Egyptian patients

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Abstract

Background Gastric carcinoma (GC) is a fatal disease. Detection of new biomarkers that can be utilized in the early diagnosis of GC is a pressing need. This present study assessed centrosomal protein-55 (CEP55) serpin family E member 1 (SERPINE1) and sphingomyelin phosphodiesterase 3 (SMPD3) genes and proteins in gastric adenocarcinoma with different tumor progression features. Thirty surgically resected gastric tissue samples from thirty patients suffered from gastric cancers were obtained. The gastric tissue samples were divided into tumorous (with different stages and grades) and adjacent non-tumorous samples. CEP55, SERPINE1 and SMPD3 genes were assessed by quantitative qRT-PCR, and their proteins were assessed by ELISA in the gastric tissue samples.

Results As regards SERPINE1, CEP55 genes and proteins, results revealed significant elevations in the GC samples ($p < 0.0001$). On the contrary, SMPD3 gene and protein revealed significant decreases as compared to non-tumorous samples. The studied genes and proteins showed highly significant specificity and sensitivity in the early detection of GC. SERPINE1 gene and protein revealed highly significant increases and positive correlations, while SMPD3 gene and protein revealed highly significant decreases and negative correlations as the tumor progresses.

Conclusion CEP55, SERPINE1 and SMPD3 genes and proteins could be used as useful biomarkers for the early detection of GC. SERPINE1 and SMPD3 genes and proteins might be used as risk and protective prognostic factors in GC, respectively.

Keywords CEP55, Gastric carcinoma, SERPINE1, SMPD3, Genes, Proteins

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1 Background

Gastric cancer (GC) is a fatal cancer with marked mortality [1]. In Egypt, GC has very low incidence, but the patients are often diagnosed at late stages with poor prognosis [2]. The pathogenesis of gastric carcinoma is due to multiple factors including the genetic and proteomic alterations [1].

Abnormal cell cycle activity enhances the progression of cancers. This abnormality is generally results from disorders in the signaling pathways of the cell proliferation or genetic alteration in the protein encoding genes in the cell cycle [1].

Some bioinformatic analysis studies identified several diagnostic and prognostic signatures for gastric carcinoma [3, 4]. Centrosomal protein-55 (CEP55), serpin family E member 1 (SERPINE1) and sphingomyelin phosphodiesterase 3 (SMPD3) genes were identified and included in these signatures. Few studies experimentally assessed them in GC at genomic and proteomic levels.

CEP55 is one of the centrosomal protein families which have a regulatory role in the cell cycle [5]. CEP55 was reported to significantly upregulated and related to the pathogenesis and the progression of GC. The CEP55-induced gastric cell transformation is mediated by the AKT signaling pathway [5].

SERPINE1 gene encodes plasminogen activator inhibitor 1 (PAI-1) [6]. PAI-1 inhibits the plasminogen activators and is involved in the pathogenesis, spread and anti-apoptotic mechanisms of cancers [7, 8].

SMPD3 gene encodes neutral sphingomyelinase-2 (nSMase2 protein) which catalyzes sphingomyelin hydrolysis in the plasma membrane into phosphorylcholine and ceramide [9] which are 2nd messengers signaling molecules involved in cellular differentiation, proliferation, adhesion and apoptosis, and the cellular responses to stress [10, 11]. SMPD3 is a potential tumor suppressor gene that is involved in various cancers as breast cancer, hepatocellular carcinoma and leukemia [12–14].

The present work was aimed to assess SERPINE1, SMPD3 CEP55 genes and proteins in gastric adenocarcinoma in different stages and grades and to reveal their correlations with other clinicopathological features of GC.

2 Methods

2.1 Sample collection

Thirty fresh frozen gastric tissue samples were obtained from the pathology department from January 2022 to June 2022. These samples were surgically resected from thirty patients who suffered from GC after having written informed consents. The clinical and pathology data were regained for the study after obtaining a written permission from the Head of the Pathology Department, Faculty of Medicine, Beni-Suef University. The studied samples were thirty gastric carcinoma samples (at different grades and stages) and thirty adjacent non-tumorous samples. Two professional pathologists performed the histopathological diagnoses independently according to the American Joint Committee on Cancer criteria. The study was carried in compliance with Helsinki Declaration guidelines and principles [15]. The research is approved by Research Ethical Committee, Faculty of Medicine, Beni-Suef University, with approval number: FMBSUREC/02012022/Abdel-Tawab.

2.2 RNA extraction

The paraffin-embedded tissue sections were used for total RNA extraction using RNeasy FFPE kit (Cat No. 73504, Qiagen, USA) according to the manufacturer's protocol. The spectrophotometry (JENWAY, USA) was used to quantify the extracted RNA at 260 nm. All procedures were done in triplicate.

2.3 PCR primers

PCR primers were designed from GenBank RNA sequences (<http://www.ncbi.nlm.nih.gov/tools/primer-blast>).

Table 1 PCR primers

Gene	Sequence	Gene bank accession number
CEP55	Forward primer. 5'-CCGTTGTCTCTTCGATCGCT-3' Reverse primer. 5'-CTCAAGGACTCGAATTTCTCCA-3'	NM_018131.4
SERPINE1	Forward primer. 5'-GCAAGGCACCTCTGAGAACT-3' Reverse primer. 5'-GGGTGAGAAAACACGTTGC-3'	NM_000602.4
SMPD3	Forward primer. 5'-TGAGGAGCCGGGAGGAG-3' Reverse primer. 5'-GCCCTACTACATGGTGTCCG-3'	NM_018667.4
GAPDH	Forward primer 5'-GATGCTGGTGTGAGTATGTCG-3' Reverse primer 5'-GTGGTCAGGATGCATTGCTGA-3'	XR: 598,347.1

The designated gene primers are given in Table 1. Ideal primer pair was optimized with considered conditions including Tm: 61–63 °C and 75–200 bp length.

2.4 RT-qPCR

Software version 3.1 (Applied Biosystems, USA) of Step One plus real-time PCR system was used to analyze the studied genes. Optimum annealing temperature was organized for the protocol of PCR and for the selected primers. The housekeeping gene was GAPDH. cDNA was obtained by using of antisense sequence specific primer (20 pmol), total RNA (five microliters), AMV reverse transcriptase (0.8 µL) for 1 h at 37 °C. SYBR® Green method (Applied Biosystems, CA, USA) was used to evaluate the mRNA expression. A negative control sample was used. The optimum annealing temperature was 60 °C for all selected primers. 25 µL reaction volume (Mater Mix of SYBR Green), 3 µL cDNA (1X/reaction) and 900 nmol/L of each primer were used in RT-qPCR.

All procedures for amplification were performed in compliance with the manufacturer protocol; the initial activation step was for 2 min at 50 °C (Reverse transcription) and for 10 min at 95 °C (initial activation of PCR) followed by 40 repeated cycles (15 s for denaturation and 10 min for annealing/extension at 60 °C).

The comparative cycle threshold (Ct) method was used for calculation of the studied gene expression. Passive reference dye (ROX) was used for normalization and GAPDH fluorescence. The Ct values of the studied genes and the reference gene (GAPDH) were determined using Applied Biosystems Step One plus software. GAPDH housekeeping gene expression data were used for normalization of the studied gene expression data.

Relative quantitation of target gene expressions (RQ) was calculated according to the following equation [16]:

$$\Delta C_t = C_t \text{ assessed gene} - C_t \text{ reference gene}$$

$$\Delta \Delta C_t = \Delta C_t \text{ sample} - \Delta C_t \text{ control gene}$$

$$RQ = 2^{-(\Delta \Delta C_t)}$$

2.5 Assessment of CEP55, SERPINE1 and SMPD3 proteins in the gastric tissue samples

Tissues weighed before homogenization then were rinsed in ice-cold PBS (0.02 mol/L, pH 7.0–7.2) and. The tissues were cut to small pieces and homogenized them in PBS with a glass homogenizer on ice. Two freeze–thaw cycles were done for the resulting suspension for further lysis of the cell membranes. The homogenates were centrifugated for 15 minutes at 5000 rpm then the supernatant was immediately removed to be assayed. SERPINE1

AND SMPD3 proteins were assessed in tissue samples using ELISA kits according to the manufacturer's recommendations [CEP55 ELISA kit (Abnova, UK, Catalog number abx386471), SERPINE1 ELISA kit (R&D Systems, USA, Catalog number DSE100) and SMPD3 ELISA kit (Abnova, Taiwan, catalog number H00055512-PW1)].

2.6 Statistical analysis

SPSS version 22 (SPSS Inc., Chicago, IL, USA) was utilized. Data were described as mean ± SD. When comparing between groups, we used analysis of variance (ANOVA) with multiple comparisons post hoc test. Simple linear correlation (Pearson correlation coefficient test) (r) was also done to test for linear relations between the studied genes and clinical variables. P-value is considered significant if $p < 0.05$. Receiver operating characteristic curves (ROC curves) were utilized detection of reliability of the studied parameters as a diagnostic tool and their best cutoff values were calculated. AUC (area under the curve) was considered significant if > 0.60 .

3 Results

3.1 CEP55, SERPINE1 and SMPD3 genes and protein expressions in the studied samples

In the GC samples, there were highly significant ($p < 0.001$) elevations in the mean levels of CEP55, SERPINE1 genes and proteins with highly significant ($p < 0.001$) decreases in the mean levels of SMPD3 gene and protein as compared to non-tumorous samples as shown in Fig. 1.

3.2 The relations between CEP55, SERPINE1 and SMPD3 genes, protein expression levels and the tumor progression features and demographic data

The results demonstrated highly significant ($p < 0.001$) elevations in the mean levels of genes and protein expression of SERPINE1 and highly significant ($p < 0.001$) decreases in the mean levels of gene and protein expression of SMPD3 in advanced features of the tumor, while the mean levels of gene and protein expression levels of CEP55 did not reveal any significant differences with the tumor progression signs as shown in Table 2. There were no significant differences between the studied parameters and the demographic data as illustrated in Table 2.

3.3 Significant correlations in the study

The results revealed significant correlations between the studied parameters as demonstrated in Table 2. Moreover, there were significant positive correlations between the mean levels of genes, protein expression of SERPINE1 and the advanced tumor features as shown in Table 3.

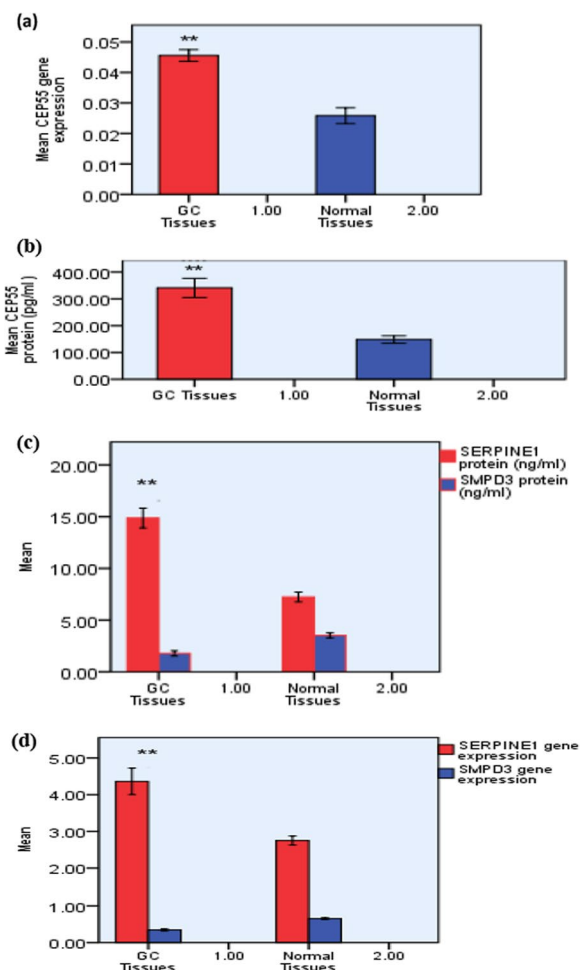


Fig. 1 SERPINE1, CEP55, and SMPD3 genes and proteins expressions in the studied gastric tissues. **a** CEP55 gene in the studied gastric tissues. **b** CEP55 protein in the studied gastric tissues. **c** SERPINE1 and SMPD3 proteins in the studied gastric tissues **d** SERPINE1 and SMPD3 genes in the studied gastric tissues **; high significant differences ($p < 0.001$) in gastric cancer tissues than normal tissues

Furthermore, there were significant negative correlations between the mean levels of genes, protein expression of SMPD3 and the advanced tumor features as shown in Table 3.

3.4 ROC curve analysis of the studied parameters in the early detection of GC

ROC curve analysis demonstrated that the optimum cutoff values for CEP55 gene expression were ≥ 0.035 (AUC 0.998, 93.3% specificity and 100% sensitivity), for CEP55 protein levels, they were ≥ 181 (AUC=0.983, 90% specificity and 100% sensitivity), for SERPINE1 gene expression, they were ≥ 3.5 (AUC 0.950, 90% specificity and 86.7% sensitivity), for SERPINE1 protein levels, they were ≥ 9.9 (AUC 0.998, 97% specificity and 100%

sensitivity), for SMPD3 gene expression, they were ≤ 0.55 (AUC 0.998, 96.7% specificity and 100% sensitivity), and for SMPD3 protein levels, they were ≤ 2.75 (AUC 0.978, 93.3% specificity and 93.3% sensitivity) as shown in Fig. 2.

4 Discussion

Gastric carcinoma (GC) accounted for 1.09 million cases worldwide [17]. The dysfunction of mRNA is reported to be involved in numerous cancers and is strongly associated with cancer development [18] Some bioinformatic analysis studies identified several diagnostic and prognostic signatures for gastric carcinoma [3, 4]. CEP55, SERPINE1 and SMPD3 genes were some of these signatures. Few studies experimentally assessed them in GC at genomic and proteomic levels. Therefore, the present work aimed to experimentally assess CEP55, SERPINE1 and SMPD3 genes and serum proteins in GC; to our best knowledge, this study is the first one to do.

The present results showed highly significant increases of CEP55 gene expression and protein levels in GC, but no significant differences were found in CEP55 gene expression and protein levels in advanced characteristics of GC. Highly significant specificity and sensitivity were found in the ROC curve analysis of the diagnostic reliability of gene expression and protein of CEP55 in the early detection of GC. These results suggested that CEP55 gene might be used as a potential diagnostic but not prognostic biomarker in GC.

The present findings coincided with Tao et al. [5] who revealed upregulation of CEP55 gene and protein expressions in GC tissues. They reported that CEP55 gene knockdown inhibited tumor cells proliferation and explained that high expression of CEP55 enhanced AKT phosphorylation and inhibited the activity of p21.

The present results may experimentally confirm the findings of Liu et al. [3] who reported that CEP55 gene was one of the nine hub genes which were upregulated in GC tissues via their integrated analysis of multiple gene expression profile datasets. They suggested that CEP55 gene was strongly involved in GC pathogenesis.

Moreover, CEP55 was reported to be upregulated in oral cavity carcinoma [19], colorectal carcinoma [20], hepatocellular carcinoma [21] and lung cancer [22]. The explanation of the previous findings regarding to CEP55 was done by Li et al. [23] who reported that CEP55 augmented the cell proliferation and blocked the apoptotic PI3K/Akt/p21 pathway in some human glioma cells. Activation of this pathway leads to suppression of p21 dysregulating cell cycle, but on the contrary, Wang et al. [24, 25] stated that CEP55 was involved in glucose metabolism regulation, surviving and apoptosis of glioma cells through Akt/mTOR signaling pathway. These findings

Table 2 Relations between the studied parameters, the demographic data and the tumor progression signs

	N	CEP55 gene expression M±SD	CEP55 protein (pg/ml) M±SD	SERPINE1 gene expression M±SD	SERPINE1 protein (ng/ml) M±SD	SMPD3 gene expression M±SD	SMPD3 protein (ng/ml) M±SD
Studies samples							
GC Tissues	30	0.046±.005	340.2±95	4.4±0.97	14.9±2.6	0.35±0.09	1.8±0.66
Normal tissues	30	0.026±.007	148±37	2.8±0.33	7.2±1.3	0.65±0.07	3.5±0.67
<i>p</i> value		<i>p</i> < 0.001	<i>p</i> < 0.001	<i>p</i> < 0.001	<i>p</i> < 0.001	<i>p</i> < 0.05	<i>p</i> < 0.001
Tumor size (cm)							
Tumor size < 3	14	0.0453±.005	350.2±108	3.7±0.76	12.7±1.6	0.39±0.09	2.2±0.48
Tumor size ≥ 3	16	0.0458±.006	332.5±85	5±0.67	16.8±1.5	0.30±0.07	1.38±0.52
<i>p</i> value		<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> < 0.001	<i>p</i> < 0.001	<i>p</i> < 0.05	<i>p</i> < 0.001
Lymph nodes							
Lymph nodes ≤ 3	12	0.046±.006	344±100	3.8±0.72	13.4±1.6	0.39±0.05	2.2±0.48
Lymph nodes > 3	18	0.046±.005	338.8±94.5	4.7±0.95	15.9±2.7	0.31±0.09	1.48±0.59
<i>p</i> value		<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> < 0.05	<i>p</i> < 0.05	<i>p</i> < 0.05	<i>p</i> < 0.05
Metastasis(organ)							
Metastasis = 1	16	0.044±.005	342±96	3.8±0.88	13.6±2.4	0.39±0.08	2.2±0.58
Metastasis > 1	14	0.047±.005	339±97	5±0.65	16.2±2	0.29±0.06	1.29±0.34
<i>p</i> value		<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> < 0.001	<i>p</i> < 0.05	<i>p</i> < 0.05	<i>p</i> < 0.001
Tumor stages							
Stage II	12	0.045±.005	343±98	3.4±0.52	12.6±1.5	0.43±0.05	2.5±0.35
Stage III	8	0.047±.005	311±87	4.8±0.39	14.9±1.8	0.33±0.07	1.7±0.20
Stage IV	10	0.046±.006	361±99	5.1±0.78	17.5±1.3	0.26±0.04	1±0.18
<i>p</i> value		<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> < 0.001	<i>p</i> < 0.001	<i>p</i> < 0.001	<i>p</i> < 0.001
Tumor grades							
G1	5	0.047±.007	323±33	3.1±0.51	12±1.7	0.47±0.06	2.6±0.38
G2	9	0.047±.006	352±107	3.8±0.81	13.6±2.3	0.39±0.07	2.2±0.60
G3	11	0.045±.003	306±63	4.98±0.51	15.8±2	0.29±0.06	1.45±0.38
G4	5	0.044±.001	426±134	4.92±1.1	17±1.7	0.28±0.02	1.08±0.27
<i>p</i> value		<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> < 0.001	<i>p</i> < 0.05	<i>p</i> < 0.001	<i>p</i> < 0.001
Age							
< 60 years	22	0.046±.006	337±99	4.4±0.9	14.9±2.8	0.35±0.09	1.7±0.68
≥ 60 years	8	0.044±.003	351±87	4.3±1.1	14.9±2	0.34±0.05	1.9±0.62
<i>p</i> value		<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> > 0.05
Gender							
Male	17	0.044±.003	318±79	4.3±1	14.8±2.8	0.35±0.08	1.8±0.75
Female	13	0.045±.005	370±109	4.5±0.9	14.9±2.4	0.34±0.09	1.7±0.55
<i>p</i> value		<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> > 0.05	<i>p</i> > 0.05

CEP55; centrosomal protein-55, SERPINE1; serpin family E member 1, SMPD3; sphingomyelin phosphodiesterase 3, *p* < 0.001; Highly significant difference, *p* < 0.05; Significant difference, *p* > 0.05; Insignificant difference, M±SD; mean ± standard deviation, N; number of cases

concluded that CEP55 could be used as an effective therapeutic target in several types of malignant tumors.

In the present study, SERPINE1 gene and protein were found to be significantly increased in GC and positively correlated with tumor progression features. ROC curve analysis of the diagnostic reliability of SERPINE1 gene expression and protein in the early detection of GC showed highly significant specificity and sensitivity.

These results suggested that SERPINE1 gene and protein might be used as useful diagnostic, prognostic biomarkers and therapeutic targets in GC.

The present findings coincided with Chen et al. [26] who revealed that SERPINE1 overexpression promotes malignant progression and poor prognosis of GC.

Li L et al. [27] agreed with the present results as they stated that SERPINE1 overexpression was

Table 3 Correlations of the studied parameters in the studied samples

	CEP55 gene expression	CEP55 protein (pg/ml)	SERPINE1 gene expression	SERPINE1 protein (ng/ml)	SMPD3 gene expression	SMPD3 protein (ng/ml)
Tumor size (cm)	$r=0.051$ $p>0.05$	$r=-0.094$ $p>0.05$	$r=0.693$ $p<0.001$	$r=0.797$ $p<0.001$	$r=-0.511$ $p<0.05$	$r=-0.656$ $p<0.001$
Lymph nodes	$r=-0.001$ $p>0.05$	$r=0.019$ $p>0.05$	$r=0.630$ $p<0.001$	$r=0.559$ $p<0.05$	$r=-0.613$ $p<0.001$	$r=-0.646$ $p<0.001$
Metastasis (organ)	$r=0.289$ $p>0.05$	$r=-0.017$ $p>0.05$	$r=0.611$ $p<0.001$	$r=0.525$ $p<0.05$	$r=-0.567$ $p<0.05$	$r=-0.696$ $p<0.001$
Tumor stages	$r=0.078$ $p>0.05$	$r=0.076$ $p>0.05$	$r=0.771$ $p<0.001$	$r=0.815$ $p<0.001$	$r=-0.819$ $p<0.001$	$r=-0.919$ $p<0.001$
Tumor grades	$r=-0.191$ $p>0.05$	$r=0.135$ $p>0.05$	$r=0.643$ $p<0.001$	$r=0.645$ $p<0.001$	$r=-0.681$ $p<0.001$	$r=-0.726$ $p<0.001$
CEP55 gene expression	1	$r=0.659$ $p<0.001$	$r=0.685$ $p<0.001$	$r=0.759$ $p<0.001$	$r=-0.788$ $p<0.001$	$r=-0.688$ $p<0.001$
CEP55 protein (pg/ml)	$r=0.659$ $p<0.001$	1	$r=0.684$ $p<0.001$	$r=0.719$ $p<0.001$	$r=-0.747$ $p<0.001$	$r=-0.647$ $p<0.001$
SERPINE1 gene expression	$r=0.685$ $p<0.001$	$r=0.684$ $p<0.001$	1	$r=0.837$ $p<0.001$	$r=-0.822$ $p<0.001$	$r=-0.770$ $p<0.001$
SERPINE1 protein (ng/ml)	$r=0.759$ $p<0.001$	$r=0.719$ $p<0.001$	$r=0.837$ $p<0.001$	1	$r=-0.892$ $p<0.001$	$r=-0.879$ $p<0.001$
SMPD3 gene expression	$r=-0.788$ $p<0.001$	$r=-0.747$ $p<0.001$	$r=-0.822$ $p<0.001$	$r=-0.892$ $p<0.001$	1	$r=0.839$ $p<0.001$
SMPD3 protein (ng/ml)	$r=-0.688$ $p<0.001$	$r=-0.647$ $p<0.001$	$r=-0.770$ $p<0.001$	$r=-0.879$ $p<0.001$	$r=0.839$ $p<0.001$	1

CEP55; centrosomal protein-55, SERPINE1; serpin family E member 1, SMPD3; sphingomyelin phosphodiesterase 3, $p<0.001$; Highly significant difference, $p<0.05$; Significant difference, $p>0.05$; Insignificant difference

significantly associated with GC poor prognosis and tumor progression.

The present findings were correlated with Liao et al. [28] who stated that SERPINE1 gene was overexpressed in gastric cancer and associated with poor prognosis and explained their results by SERPINE1 gene encoding protein that was involved in tumor cells adhesion to the extracellular matrix enhancing tumor spread [29].

Yang et al. [30] coincided with the present results as they reported that SERPINE1 is a cancer-promoting gene in GC facilitating tumor cell proliferation, migration and invasion by regulating EMT. Moreover, xu et al. [31] agreed with the present findings as they identified SERPINE1 as a prognostic biomarker in GC.

The present results correlated with Li et al. [32] who revealed that SERPINE1 was upregulated in GC suggesting its high diagnostic value and its association with poorer prognosis of GC. The present findings agreed with Ma et al. [33] who used bioinformatic analysis reported that SERPINE1 was highly related to the prognosis of GC inhibiting the immune-dominant status of the microenvironment (TME) of the tumor in GC.

Ma et al. [34] correlated with the present results as they revealed that SERPINE1 was associated with poor prognosis of GC by being involved in multiple immune cell infiltrates in gastric cancer. Also, Akhavan et al. [35] agreed with the present findings as they identified SERPINE1 as one of the four genes upregulated in GC suggesting that these genes being predicted targets of hsa-miR-421 and hsa-miR-193a-3p.

The present results coincided with Wang et al. [36] who reported that SERPINE1 gene was upregulated in GC by using GEPIA database. Moreover, they revealed higher levels of SERPINE1 protein in GC tissues suggesting its role in promoting GC proliferation and migration in vitro.

Teng et al. [37] correlated with the present results as they concluded that SERPINE1 was associated with poor prognosis of GC by activating the VEGFR-2 signaling pathway promoting tumor progression and angiogenesis. Furthermore, Zhao et al. [38] agreed with the present findings as they revealed that SERPINE1 was identified to be associated with carcinogenicity and poor prognosis of GC.

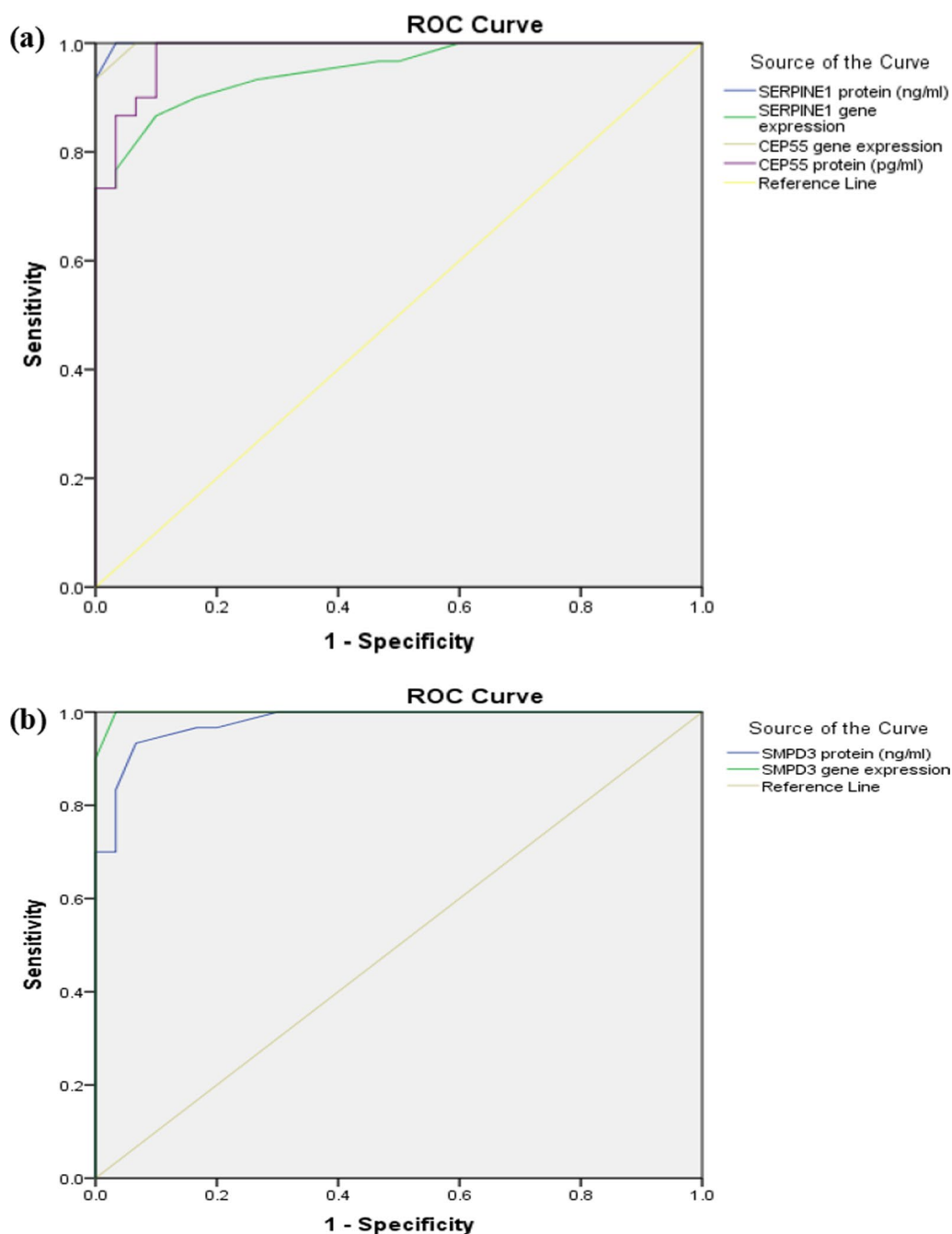


Fig. 2 ROC curve analysis of the studied parameters in the prediction of the presence of gastric carcinoma. **a** ROC curve analysis of SERPINE1 gene, protein, CEP55 gene and protein in the prediction of the presence of gastric carcinoma. **b** ROC curve analysis of SMPD3 gene and protein in the prediction of the presence of gastric carcinoma

On the contrary, SMPD3 gene expression and serum protein levels in the present study were found to be highly significantly decreased in the GC samples compared to non-tumorous samples with highly significant negative correlations with tumor progression signs. ROC curve analysis of diagnostic reliability of SMPD3

gene expression and protein in the early detection of GC showed highly significant specificity and sensitivity. These findings suggested that SMPD3 gene and protein might be used as potential diagnostic and protective prognostic biomarkers in GC.

These findings coincided with Liu et al. (3) who revealed the downregulation of SMPD3 in GC suggesting its protective prognostic role in GC.

Few studies investigated the association of SMPD3 with gastric cancer (3). However, integrative genomic analysis considered SMPD3 as a suppressor gene in other tumors such as HCC [14]. SMPD3 locus is present in chromosome 16q22.1, and this locus genetic or epigenetic modification is associated with the progression of various cancers [39].

The present work revealed highly significant correlations between six studied parameters: CEP55, SERPINE1 and SMPD3 genes and their corresponding serum proteins for further future studies to know explaining molecular mechanisms involved in these correlations.

5 Conclusion

The present study concluded that CEP55 and SERPINE1 genes and proteins were significantly upregulated in gastric carcinoma. On the other hand, SMPD3 gene and protein were significantly downregulated in gastric carcinoma. SERPINE1 was strongly associated with poor prognosis of GC while SMPD3 might have a protective role in GC. Highly significant sensitivity and specificity were reported for CEP55, SERPINE1 and SMPD3 genes and proteins in the prediction of the presence of gastric carcinoma.

These genes and proteins could be used as novel potential biomarkers in early detection of GC. SERPINE1 gene might be a risk prognostic factor while SMPD3 might be a protective prognostic factor in GC. However, further experimental studies are urgently needed to validate the present results on a larger sample size using other experimental techniques.

Abbreviations

GC	Gastric carcinoma
CEP55	Centrosomal protein-55
SERPINE1	Serpin family E member 1
SMPD3	Sphingomyelin phosphodiesterase
nSMase2	Neutral sphingomyelinase 2
PAI-1	Plasminogen activator inhibitor 1
qRT-PCR	Real-time quantitative reverse transcription PCR
Ct	Cycle threshold
VEGFR-2	Vascular endothelial growth factor-2
TME	Tumor microenvironment
EMT	Epithelial–mesenchymal transition
GEPiA	Gene expression profiling interactive analysis
HCC	Hepatocellular carcinoma
GAPDH	Glyceraldehyde 3-phosphate dehydrogenase

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Author contributions

MA is the major contributor to writing, preparation of original draft, data analysis and supervision. HF was involved in conceptualization, methodology, formal analysis, investigation and manuscript writing. NA, AF and SA were responsible for formal analysis, investigation, data analysis, acquisition and manuscript writing. AY, AA and SS took part in acquisition, investigation, data analysis and curation, writing review, resources and editing. All authors approved the final version to be published.

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Availability of data and materials

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

The study was carried in compliance with guidelines and principles of Helsinki Declaration and approved by the research ethic committee. The research is approved by Research Ethical Committee, Faculty of Medicine, Beni-Suef University, with approval number: FMBSUREC/02012022/Abdel-Tawab.

Consent for publication

This manuscript has not been published and is not under consideration for publication elsewhere. We have no conflicts of interest to disclose. All authors have approved the manuscript and agree with submission to Beni-Suef University Journal of Basic and Applied Sciences.

Competing interests

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