Am J Cancer Res 2026;16(8):3476-3487
www.ajcr.us /ISSN:2156-6976/ajcr0174814

Original Article

High-fat diet-responsive DNM1 promotes hepatocellular carcinoma progression and predicts poor prognosis in viral-associated patients

Po-Ming Chen[1,2*], Huai-Yi Huang[3*], Hsin-Hung Chen[4], Rong-Fu Chen[5,6], En-Pei Isabel Chiang[7,8,9,10]

[1]Research Assistant Center, Show Chwan Memorial Hospital, Changhua 500, Taiwan; [2]Department of Nursing, Central Taiwan University of Science and Technology, Taichung 406, Taiwan; [3]Division of Gastroenterology and Hepatology, Chang Bing Show Chwan Memorial Hospital, Changhua 500, Taiwan; [4]Department of Medical Education and Research, Kaohsiung Veterans General Hospital, Kaohsiung 813, Taiwan; [5]Division of Plastic Surgery, Department of Surgery, Kaohsiung Medical University Hospital, Kaohsiung 807, Taiwan; [6]Regenerative Medicine and Cell Therapy Research Center, Kaohsiung Medical University, Kaohsiung 807, Taiwan; [7]Department of Food Science and Biotechnology, National Chung Hsing University, Taichung 404, Taiwan; [8]Graduate Institute of Food Safety, National Chung Hsing University, Taichung 404, Taiwan; [9]Doctoral Program in Translational Medicine, National Chung Hsing University, Taichung 404, Taiwan; [10]Innovation and Development Center of Sustainable Agriculture (IDCSA), National Chung Hsing University, Taichung 404, Taiwan. [*]Equal contributors.

Received June 17, 2026; Accepted July 29, 2026; Epub August 15, 2026; Published August 30, 2026

Abstract: Hepatocellular carcinoma (HCC) arises from diverse etiologies, among which metabolic dysfunction-associated liver disease and chronic viral hepatitis are the two major drivers worldwide. However, the molecular mechanisms linking metabolic stress to HCC progression remain incompletely understood. Dynamin-1 (DNM1), primarily known for its role in vesicular trafficking, has emerged as a potential oncogene, yet its prognostic and functional significance in HCC remains largely unexplored. Here, we investigated the role of DNM1 in high-fat diet (HFD)-associated hepatocarcinogenesis. Transcriptomic profiling was conducted to identify differentially expressed genes between normal and high-fat diet murine models, with human orthologs mapped. Clinical relevance was validated using The Cancer Genome Atlas (TCGA-LIHC) dataset. Survival analysis, GSEA (Gene Set Enrichment Analysis), and subgroup stratifications based on viral hepatitis status were performed. *In vitro*, loss-of-function assays (shRNA knockdown) were executed in HepG2 and SK-Hep1 cell lines to assess cell viability and migration. DNM1 was significantly upregulated in high-fat diet models. In the TCGA-LIHC cohort, high DNM1 expression was an independent risk factor for poor overall survival (HR=1.44, P=0.039) and correlated with advanced tumor stages (Stage III+IV, P=0.010). In vitro knockdown of DNM1 profoundly impaired cell proliferation and migration in HCC cell lines. Strikingly, DNM1 expression was further elevated in patients with concurrent viral hepatitis (P=0.009). GSEA revealed that high DNM1 expression was positively associated with viral infection pathways and negatively correlated with critical immune responses, including interferon-alpha/gamma responses and host immune cytolysis. Survival analysis stratified by four subgroups demonstrated that patients with both viral infection and high DNM1 expression exhibited the worst prognosis (Overall Log-rank P < 0.001). Our findings identify DNM1 as a high-fat diet-responsive regulator that links metabolic stress to hepatocellular carcinoma progression. Elevated DNM1 expression promotes malignant phenotypes in HCC and identifies a subgroup of viral-associated patients with particularly poor prognosis, highlighting DNM1 as a potential prognostic biomarker and therapeutic target.

Keywords: Hepatocellular carcinoma (HCC), dynamin-1 (DNM1), viral hepatitis, high-fat diet, tumor microenvironment, prognosis

Introduction

Hepatocellular carcinoma (HCC) is the most common primary liver malignancy and remains a major global health burden, ranking as the sixth most frequently diagnosed cancer and the third leading cause of cancer-related mortality worldwide [1]. The global incidence of HCC is highly heterogeneous, with a disproportionate burden in Eastern Asia and sub-Saharan Africa, reflecting regional differences in underlying etiologies such as chronic viral hepatitis,

https://doi.org/10.62347/KHKV1727

environmental exposures, and metabolic disorders [2-4]. In recent decades, the epidemiology of HCC has shifted markedly, with metabolic liver disease emerging as a dominant driver of hepatocarcinogenesis.

Nonalcoholic fatty liver disease (NAFLD) has become the most prevalent chronic liver disorder worldwide, affecting approximately 25% of the adult population [5, 6]. NAFLD encompasses a pathological continuum ranging from simple steatosis to nonalcoholic steatohepatitis (NASH), progressive fibrosis, cirrhosis, and ultimately HCC. Importantly, NAFLD-associated HCC can arise even in the absence of cirrhosis, highlighting the complex and multifactorial mechanisms underlying disease progression [7, 8]. Central to NAFLD pathogenesis is hepatic lipid accumulation driven by an imbalance between lipid uptake, de novo lipogenesis, and lipid disposal pathways, including β-oxidation and very-low-density lipoprotein (VLDL) secretion [9, 10]. Beyond simple steatosis, dysregulated lipid metabolism leads to lipotoxicity, oxidative stress, and chronic inflammation, creating a pro-tumorigenic microenvironment that promotes liver cancer development [11, 12].

Emerging evidence further suggests that alterations in intracellular lipid handling - encompassing lipid absorption, cholesteryl ester production, lipid storage, and lipoprotein transport - play a critical role in shaping disease progression from fatty liver to HCC. These processes require dynamic membrane remodeling and vesicular trafficking to coordinate lipid flux and cellular adaptation to metabolic stress. In this context, endocytic pathways have gained increasing attention as regulators not only of receptor signaling but also of nutrient uptake and intracellular lipid trafficking [13]. However, the molecular mediators that integrate lipid metabolic reprogramming with membrane trafficking and tumor progression remain incompletely understood.

By integrating transcriptomic analysis of a high-fat diet (HFD) mouse model with clinical and functional investigations, we identified DNM1 as a lipid-responsive gene upregulated under metabolic stress of high-fat and demonstrated that elevated dynamin 1 (DNM1) expression is associated with poor prognosis in HCC using GSEA. Furthermore, our functional studies revealed that DNM1 promotes hepatocellular carcinoma cell viability and migration in the present study.

The dynamin family of large GTPases plays a central role in membrane fission during endocytosis and vesicular trafficking. Unlike classical small GTPases, dynamins possess unique structural and biochemical properties that enable them to generate mechanical force for membrane remodeling [14, 15]. Among them, DNM1 has been traditionally characterized as a neuron-enriched isoform essential for synaptic vesicle recycling, with genetic alterations linked to severe neurodevelopmental disorders [16, 17]. Nevertheless, emerging studies indicate that DNM1 can be expressed in non-neuronal tissues and may be reactivated under conditions of cellular stress. Beyond its canonical role in vesicle scission, dynamin has also been shown to coordinate membrane dynamics with actin cytoskeleton remodeling, thereby regulating cell migration and invasion [18-20]. Recent evidence further implicates DNM1 in cancer progression, where it contributes to tumor growth, migration, and activation of oncogenic signaling pathways [21, 22].

In this study, we hypothesized that DNM1 acts as an oncogenic driver in HCC, bridging metabolic reprogramming and immune evasion. We initiated our investigation by identifying differentially expressed genes in a high-fat diet murine model and cross-referencing them with human orthologs. Using the TCGA-LIHC dataset, we thoroughly validated the prognostic value of DNM1 and its clinical correlations. Furthermore, we functionally confirmed the pro-tumorigenic role of DNM1 using in vitro cellular models. Ultimately, we uncovered a novel interaction between DNM1 expression, viral hepatitis, and the suppression of host immune responses, highlighting DNM1 as a critical biomarker and therapeutic target in HCC.

Materials and methods

Data acquisition and processing

Clinical and transcriptomic data for the Liver Hepatocellular Carcinoma (LIHC) cohort were retrieved from The Cancer Genome Atlas (TCGA) via the UCSC Xena platform. The dataset included primary tumor samples and matched solid tissue normal samples. Gene expression data (RNA-Seq) was quantified as

log2(RSEM_count+1). Clinical metadata, including overall survival (OS), pathologic stage, and viral hepatitis serology (HBV/HCV), were meticulously matched with the genomic data.

Gene set enrichment analysis (GSEA)

To explore the biological pathways associated with DNM1 expression, GSEA was performed. The primary tumor samples were ranked based on their correlation with DNM1 expression. We queried the Gene Ontology (GO), KEGG, and Hallmark gene sets to identify enriched pathways, specifically focusing on immune response and viral infection signatures.

Cell culture and transfection

HepG2 and SK-Hep1 human hepatocellular carcinoma cell lines were cultured in Dulbecco's Modified Eagle's Medium (DMEM) obtained from the American Type Culture Collection (ATCC; Manassas, VA, USA). The culture medium was supplemented with 10% fetal bovine serum (FBS; Sigma-Aldrich, St. Louis, MO, USA), 100 U/mL penicillin, and 100 µg/mL streptomycin (Invitrogen, Carlsbad, CA, USA). Cells were maintained at 37°C in a humidified incubator with 5% CO_2. Prior to experiments, cells were pre-cultured under standard conditions (37±1°C, 5±1% CO_2). For experiments, cells were seeded into 96-well plates at a density of 1×10^4 cells per well in glucose-containing culture medium and incubated for 24 h. HepG2 and SK-Hep1 cells were then transfected with scrambled control shRNA or DNM1 shRNA plasmids using Lipofectamine™ 3000 (Thermo Fisher Scientific) according to the manufacturer's instructions. The DNM1 shRNA plasmids were purchased from the National RNAi Core Facility, Academia Sinica (Taipei, Taiwan), including TRCN0000288963 (DNM1 shRNA #1; target sequence: CCAGCTACTGTCCATTGAGAA) and TRCN0000296076 (DNM1 shRNA #2; target sequence: GGTCAATGCAACCACAGAATA). For HepG2 cells, transfection was performed using 0.1 µg plasmid DNA combined with 0.1 µL P3000 reagent and 0.15 µL Lipofectamine™ 3000 per well. For SK-Hep1 cells, transfection was conducted using 0.05 µg plasmid DNA with 0.1 µL P3000 reagent and 0.2 µL Lipofectamine™ 3000 per well. Following transfection, cells were incubated for an additional 24 h under standard culture conditions before being subjected to subsequent experiments.

Cell viability assay

Cell viability was assessed using the MTS assay (Promega, Madison, WI, USA) according to the manufacturer's instructions. For drug sensitivity and viability analyses, HepG2 and SK-Hep1 cells were transfected with scrambled control shRNA or DNM1 shRNA (#1 or #2) plasmids for 24 h. After incubation, the culture medium was removed, and cells were gently washed with phosphate-buffered saline (PBS). MTS reagent was then added to each well, followed by incubation for 3 h at 37°C. Absorbance was measured at 490 nm using a microplate reader, and cell viability was calculated relative to the control group.

Wound healing assay

To examine the effect of DNM1 on the migratory capacity of HepG2 and SK-Hep1 cells, DNM1 expression was suppressed using shRNA-mediated knockdown, and cell migration was evaluated by a wound healing assay. Cells were cultured to full confluence and transfected with scrambled control shRNA or DNM1 shRNA (#1 or #2). SK-Hep1 cells were transfected for 24 h, whereas HepG2 cells were transfected for 48 h. A linear wound was created using a sterile pipette tip, and cell migration into the wound area was monitored under a phase-contrast microscope and recorded using a digital camera.

RNA extraction and quantitative real-time PCR

Twenty-four hours after shRNA transfection, total RNA was extracted from cultured cells using the Total RNA Isolation Kit (Blood/Cultured Cell/Fungus; Cat. No. NA017-0100) according to the manufacturer's instructions. RNA concentration and purity were determined spectrophotometrically. Equal amounts of total RNA were reverse-transcribed into complementary DNA (cDNA) using the GScript First-Strand Synthesis Kit (Cat. No. MB305-0050) following the manufacturer's protocol. Quantitative real-time PCR (qPCR) was performed using Fast SYBR™ Green Master Mix in a total reaction

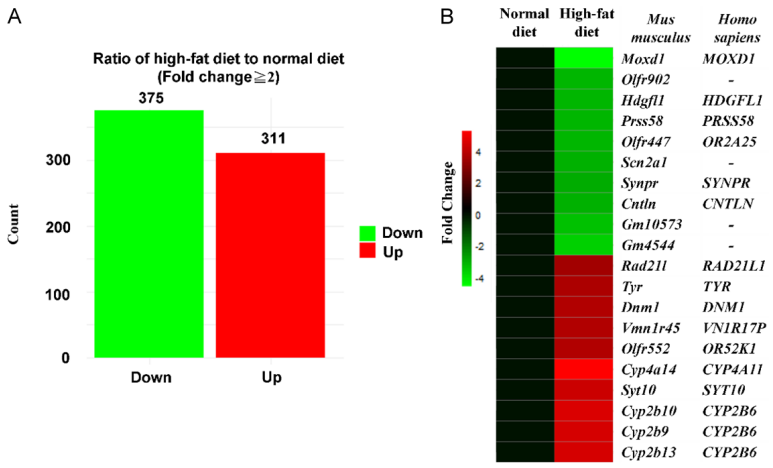

Figure 1. Transcriptomic changes induced by a high-fat diet. A. Bar graph showing the number of differentially expressed genes (DEGs) in liver tissues from mice fed a high-fat diet compared with a normal diet, using a fold change threshold of ≥ 2. A total of 375 genes were downregulated (green) and 311 genes were upregulated (red) under the high-fat diet condition. B. Heatmap illustrating the expression patterns of selected DEGs under normal diet and high-fat diet conditions. Gene expression levels are represented as fold changes, with green indicating downregulation and red indicating upregulation in response to the high-fat diet. Corresponding human homologs (Homo sapiens) are listed alongside mouse genes (Mus musculus) where available.

volume of 20 μL, consisting of 10 μL Fast SYBR™ Green Master Mix, 1 μL forward primer, 1 μL reverse primer, 2 μL cDNA template (5 ng/μL), and 6 μL nuclease-free water. Amplification was performed on a real-time PCR system according to the manufacturer's recommended cycling conditions. The primer sequences for DNM1 were as follows: Forward (5'→3') GTGGACATGGTTATCTCGGAGC and Reverse (5'→3') GGTGGTCACAATTCGCTCCATC. GAPDH was used as the endogenous reference gene. Relative DNM1 mRNA expression was calculated using the $2^{-\Delta\Delta Ct}$ method, with the control group normalized to 100%.

Statistical analysis

Patients were stratified into high and low expression groups based on the median expression value of DNM1. Univariate Cox proportional hazards regression was employed to calculate Hazard Ratios (HR) and 95% confidence intervals (CI) for screening prognostic genes. Survival curves were estimated using the Kaplan-Meier method, and differences were assessed using the Log-rank test. For clinical variable comparisons (e.g., Stage, Viral Status), Student's t-tests and Chi-square tests were utilized for continuous and categorical variables, respectively. Statistical significance was defined as $P < 0.05$. All statistical analyses were performed using R software (version 4.2.2 for Windows).

Results

Comparative hepatic gene expression profiling in mice fed normal vs. high-fat diet

To characterize transcriptional alterations induced by a high-fat diet, differential gene expression analysis was performed using the GSE108169 dataset, which profiles hepatic gene expression in mice fed either a normal diet or a high-fat diet for 11 weeks. Gene expression was measured using Agilent Mouse Oligo Microarrays, and data processing was conducted with GeneSpring GX version 7.3. Using a fold change cutoff of ≥ 2, a total of 686 differentially expressed genes were identified, including 375 downregulated and 311 upregulated genes in the high-fat diet group compared with controls (**Figure 1A**). This distribution indicates substantial transcriptional remodeling under metabolic stress conditions. A heatmap of the top 10 upregulated and top 10 downregulated genes revealed clear segregation between dietary groups (**Figure 1B**), highlighting distinct expression signatures associated with high-fat feeding. Notably, several genes involved in lipid metabolism, intracellular trafficking, and signaling pathways - including *Dnm1, Tyr, Rad21l1, Cyp4a14, Cyp2b10,* and *Syt10* - were markedly upregulated in the high-fat diet group. Cross-species comparison identified conserved human homologs such as *DNM1, TYR, RAD21L1, CYP4A11, CYP2B6,* and *SYT10*, supporting the potential translational relevance of these findings. In contrast, several genes, including *Olfr902, Scn2a1, Gm10573,* and *Gm4544*, lacked identifiable human homologs. Collectively, these data identify a set of evolutionarily conserved hepatic genes responsive to dietary lipid overload and provide a foundation for subsequent functional investigation.

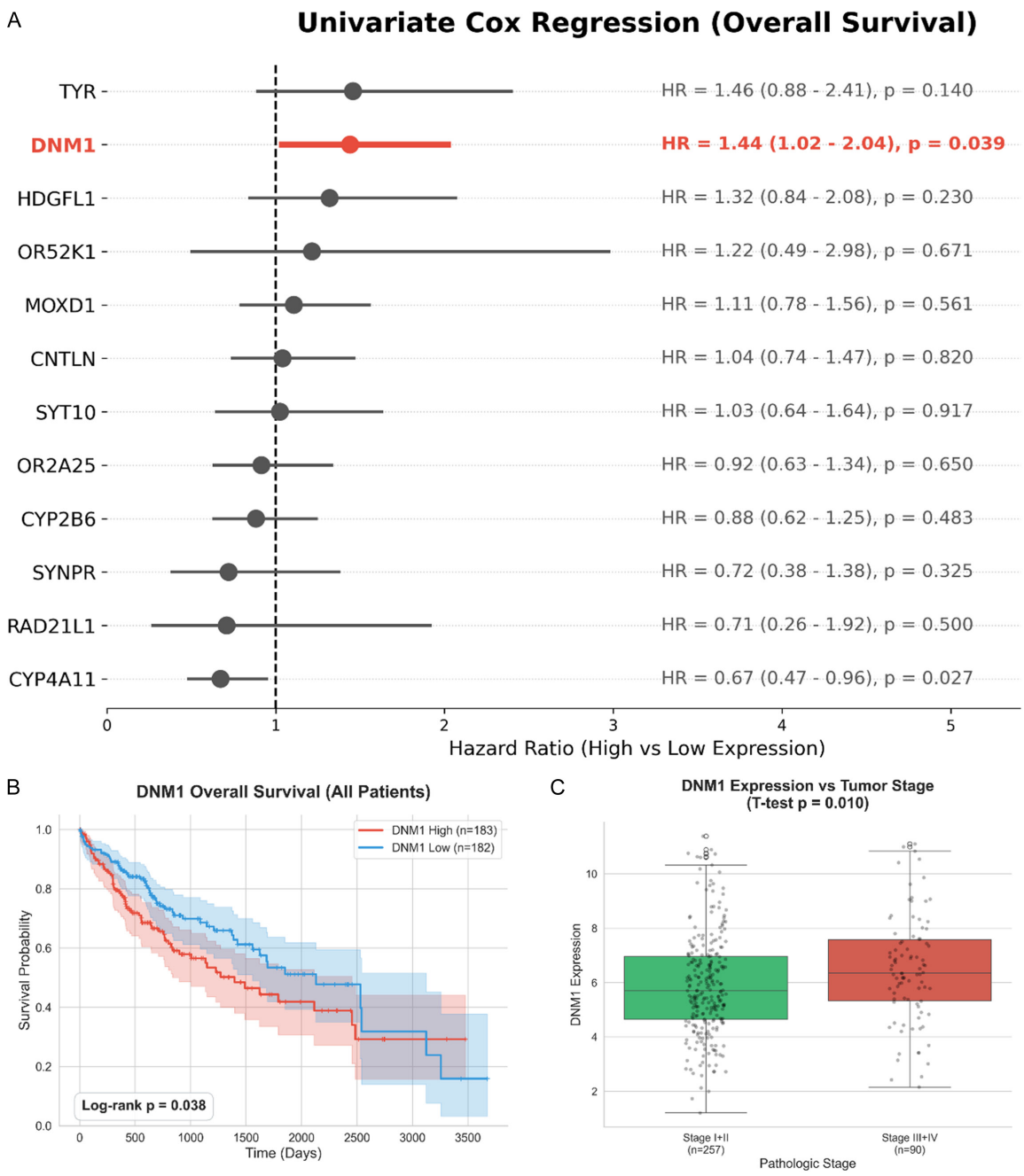

Figure 2. Prognostic and clinical significance of DNM1 in TCGA-LIHC. A. Univariate Cox regression forest plot of the high-fat diet-associated genes for Overall Survival. DNM1 acts as a significant risk factor (HR=1.44, P=0.039). B. Kaplan-Meier survival curve demonstrating that high DNM1 expression is associated with poor overall survival (Log-rank P=0.038). C. Boxplot showing significantly higher DNM1 expression in advanced tumor stages (Stage III+IV) compared to early stages (Stage I+II) (T-test P=0.010).

DNM1 is a poor prognostic factor and correlates with advanced stage in HCC

To determine the clinical significance of these high-fat diet-associated genes in human HCC, we performed Univariate Cox regression analysis using the TCGA-LIHC dataset. Among the up-regulated orthologs, DNM1 was the only gene demonstrating a significant association with poor overall survival (HR=1.44, 95% CI: 1.02-2.04, P=0.039) (**Figure 2A**). Kaplan-Meier analysis confirmed that HCC patients with high

DNM1 expression had significantly shorter overall survival compared to the low expression group (Log-rank P=0.038) (**Figure 2B**). Furthermore, clinical correlation analysis revealed that DNM1 expression was significantly elevated in advanced pathologic stages (Stage III+IV) compared to early stages (Stage I+II) (T-test P=0.010) (**Figure 2C**), confirming its role as a biomarker for disease progression.

Silencing of DNM1 suppresses HCC cell proliferation and migration in vitro

To validate the oncogenic role of DNM1 identified in our clinical and bioinformatic analyses, we performed in vitro loss-of-function assays using two human hepatocellular carcinoma cell lines, HepG2 and SK-Hep1. Quantitative real-time PCR confirmed successful DNM1 knockdown 24 h after transfection in HepG2 and SK-Hep1 cells. In HepG2 cells, DNM1 mRNA expression was reduced to 72.2% and 63.3% in the shRNA DNM1 #1 and shRNA DNM1 #2 groups, corresponding to knockdown efficiencies of 27.8% and 36.7%, respectively. In SK-Hep1 cells, DNM1 expression decreased to 74.2% and 66.0%, with knockdown efficiencies of 25.8% and 34.0%, respectively, compared with the control group. Transfection with two independent short hairpin RNAs (shRNA DNM1 #1 and shRNA DNM1 #2) successfully attenuated DNM1 protein expression compared to the control group, as confirmed by Western blot analysis (**Figure 3A** and **3B**, left panels). We first assessed the impact of DNM1 silencing on tumor cell growth. Notably, knockdown of DNM1 significantly suppressed relative cell viability in both HepG2 (**Figure 3A**, right panel; P=0.015 and P=0.0013) and SK-Hep1 cells (**Figure 3B**, right panel; P= 0.0048 and P=0.0019). Concordant with the bioinformatic predictions, DNM1 depletion markedly impaired the migratory capacity of HCC cells. In HepG2 cells, the wound closure rate at 48 hours was significantly reduced from ~38% in the control group to ~25% in the shRNA-treated groups (**Figure 3C**). Similarly, in SK-Hep1 cells, the robust wound closure observed at 24 hours in the control group was significantly abrogated upon DNM1 knockdown (**Figure 3D**). Collectively, these in vitro experiments demonstrate that DNM1 is functionally essential for maintaining the proliferative and invasive phenotypes of hepatocellular carcinoma cells.

DNM1 facilitates viral infection and immune suppression in the HCC microenvironment

HCC etiology is intimately linked to viral hepatitis (HBV/HCV) and host immune evasion. Intriguingly, Gene Set Enrichment Analysis (GSEA) of the TCGA cohort revealed that high DNM1 expression was positively correlated with viral infection pathways (e.g., Human Papillomavirus Infection) while being significantly negatively correlated with crucial anti-tumor immune responses, including the Interferon Gamma/Alpha responses and Cytolysis by Host of Symbiont Cells (**Figure 4A**). Clinically validating this, we found that DNM1 expression was significantly higher in patients with viral hepatitis compared to non-viral patients (T-test P=0.007). The proportion of 'DNM1 High' patients was also significantly enriched in the viral group (57.2%) compared to the non-viral group (45.2%) (Chi-square P=0.030) (**Figure 4B**). To synthesize these findings, we stratified the patients into four subgroups based on viral status and DNM1 expression. Survival analysis revealed a dramatic synergistic effect: patients suffering from both viral infection and high DNM1 expression exhibited precipitously worse survival outcomes compared to all other groups (Overall Log-rank P < 0.001) (**Figure 4C**). This suggests that DNM1 may exacerbate viral-mediated hepatocarcinogenesis by orchestrating an immunosuppressive microenvironment.

Discussion

In the present study, we systematically unveiled the multifaceted role of DNM1 in the pathogenesis of HCC. Originally identified by our analysis as a highly responsive gene to high-fat diet metabolic stress, we demonstrated that DNM1 possesses robust clinical prognostic value. Elevated DNM1 expression predicts advanced tumor stage and inferior overall survival in the TCGA-LIHC cohort. Importantly, our *in vitro* loss-of-function experiments using HepG2 and SK-Hep1 cell lines solidified these clinical observations, proving that DNM1 is indispensable for the proliferation and migration of HCC cells. Since both HepG2 and SK-Hep1 are established non-viral HCC cell lines, these findings emphasize that DNM1 possesses a fundamental, basal oncogenic function that

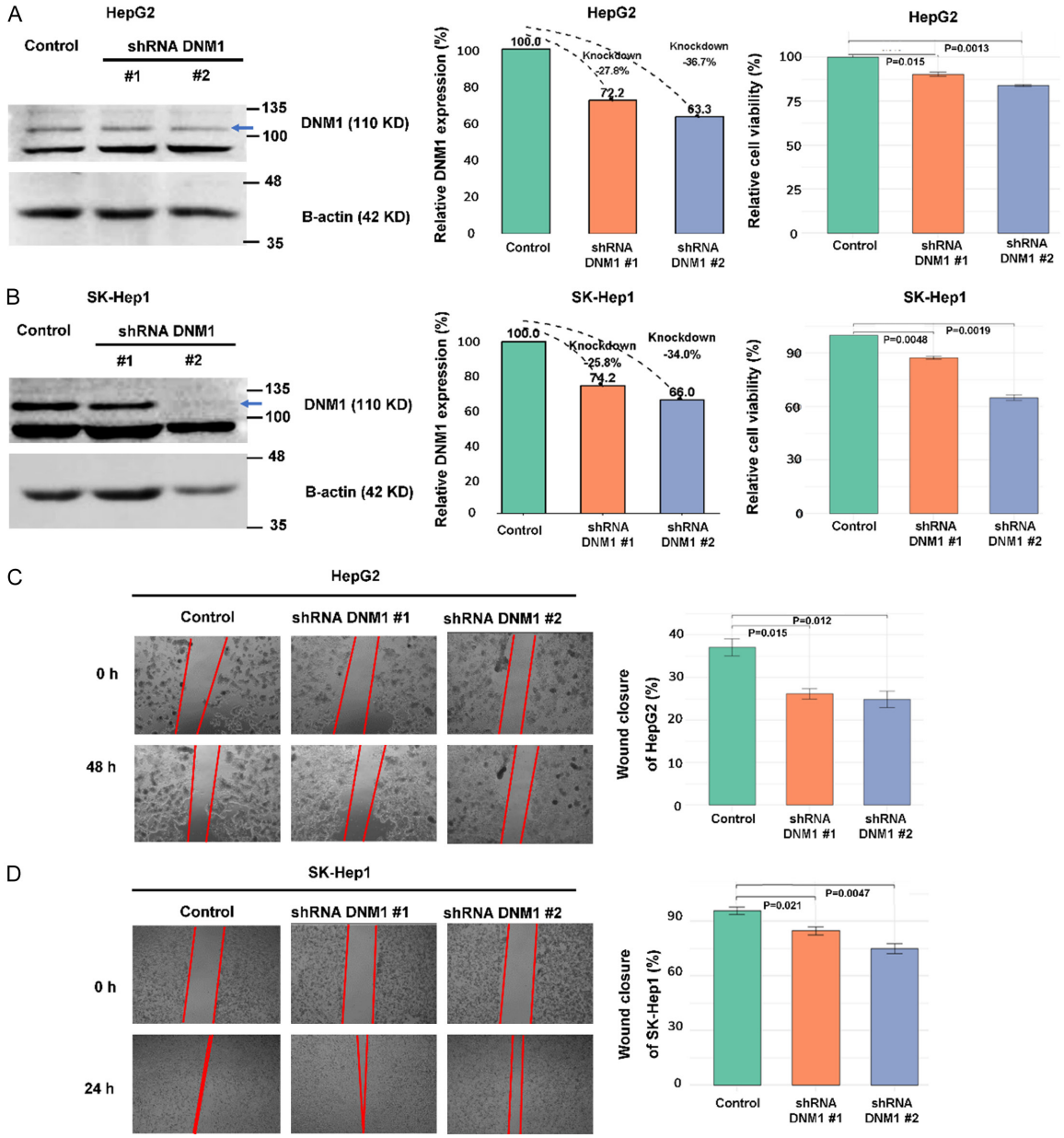

Figure 3. Silencing of DNM1 inhibits the proliferation and migration of hepatocellular carcinoma cells in vitro. A. Left panel: Western blot analysis confirming the knockdown efficiency of DNM1 using two independent shRNAs (shRNA DNM1 #1 and #2) in HepG2 cells. B-actin served as the loading control. Middle panel: DNM1 expression was reduced by 27.8% and 36.7% in HepG2 cells using shDNM1 #1 and shDNM1 #2, respectively. Right panel: Bar chart representing the relative cell viability of HepG2 cells following DNM1 knockdown, indicating a significant decrease in cellular proliferation. B. Left panel: Western blot validation of DNM1 knockdown in SK-Hep1 cells. Middle panel: DNM1 expression was reduced by 25.8% and 34.0% in SK-Hep1 cells using shDNM1 #1 and shDNM1 #2, respectively. Right panel: Relative cell viability assay demonstrating impaired proliferation in SK-Hep1 cells transfected with DNM1 shRNAs. C. Representative micrographs (left) and quantitative bar chart (right) of the wound healing assay in HepG2 cells at 0 h and 48 h. Red lines indicate the migration fronts. DNM1 silencing significantly reduced the percentage of wound closure. D. Representative micrographs (left) and quantitative bar chart (right) of the wound healing assay in SK-Hep1 cells at 0 h and 24 h, showing a significant suppression of migratory capacity upon DNM1 knockdown. Quantitative data are presented as mean ± standard deviation. Statistical significance (*P*-values) between groups is indicated directly above the corresponding bars.

drives tumor progression independently of viral etiology.

The dynamin superfamily comprises large, multi-domain GTPases that are structurally and

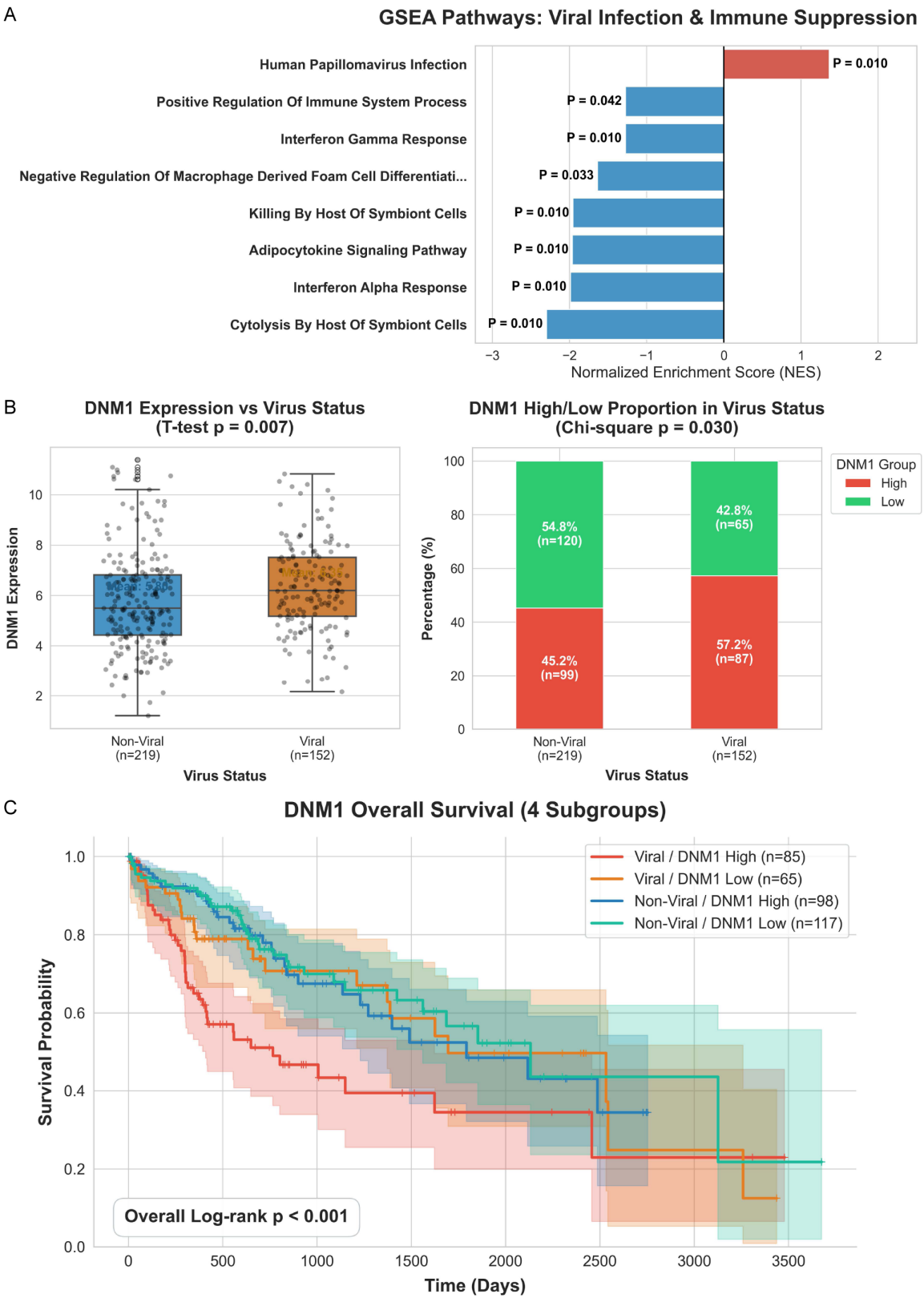

Figure 4. The intersection of DNM1, viral infection, and immune suppression. A. GSEA identifying pathways correlated with DNM1. DNM1 positively correlates with viral infection and negatively correlates with interferon responses and host cell cytolysis. B. Clinical validation showing significantly elevated DNM1 expression (left) and a higher

proportion of DNM1-High patients (right) in the viral hepatitis subgroup. C. Kaplan-Meier analysis of four subgroups combining viral status and DNM1 expression, demonstrating the worst prognosis for patients with concurrent viral infection and high DNM1 expression (Overall Log-rank P < 0.001).

functionally distinct from classical small GTP-binding proteins such as Ras, Rab, and heterotrimeric G proteins. Unlike these canonical GTPases, dynamins exhibit a modular domain architecture, relatively large molecular size, and low nucleotide affinity, enabling them to generate mechanical force for membrane remodeling, particularly membrane fission during clathrin-mediated endocytosis and vesicular trafficking [14]. These unique biochemical properties position dynamins as central regulators of intracellular membrane dynamics, and dysregulation of dynamin activity has been implicated in a wide spectrum of human diseases, reflecting their tissue-specific and context-dependent functions in maintaining cellular homeostasis [15, 23].

DNM1 has historically been characterized as a neuron-enriched isoform essential for synaptic vesicle recycling and neurotransmission. Consistent with this role, pathogenic variants in DNM1 are strongly associated with neurodevelopmental disorders, including developmental and epileptic encephalopathies, as well as domain-specific phenotypes linked to mutations in the pleckstrin homology region [16, 17, 24]. These observations underscore the fundamental importance of DNM1-mediated membrane trafficking in high-demand physiological contexts. However, emerging evidence indicates that DNM1 is not strictly neuron-specific and may be reactivated in non-neuronal tissues under conditions of cellular stress or increased membrane trafficking demand.

A growing body of literature suggests that dynamin family proteins play a critical role in coordinating membrane dynamics with actin cytoskeleton remodeling, thereby regulating cell motility and invasive behavior. Mechanistically, dynamin has been shown to directly modulate actin polymerization and organization at membrane sites, coupling membrane deformation with cytoskeletal force generation [18, 19, 25]. This coupling is particularly important in migratory processes, where coordinated membrane extension and actin remodeling are required for directional movement. Indeed, dynamin-dependent regulation of actin dynamics has been demonstrated to facilitate cell migration in complex three-dimensional environments [20]. Furthermore, interactions between dynamin and cytoskeletal regulators, such as α-actinin-4, enhance invasive capacity by promoting focal adhesion turnover and actin reorganization [26]. In cancer contexts, dynamin-containing complexes - including β-Pix-dynamin signaling modules - have been shown to drive tumor progression by integrating membrane trafficking with cytoskeletal remodeling [27], while direct interactions with membrane proteins such as podocalyxin further promote migratory and metastatic phenotypes [28]. These mechanistic insights provide a coherent framework for our observation that DNM1 enhances hepatocellular carcinoma cell migration.

Beyond its role in cell motility, DNM1 has increasingly been implicated in cancer biology as a regulator of tumor progression and clinical outcome, although its functional impact appears to be highly context-dependent. Elevated DNM1 expression has been associated with enhanced tumor growth, migration, and poor prognosis in colorectal cancer, in part through activation of the PI3K/AKT signaling pathway [22]. More broadly, DNM1 has been incorporated into prognostic and predictive biomarker panels across multiple malignancies, including bladder cancer, glioma, and lung adenocarcinoma, where it contributes to models predicting survival or metastatic risk [29-31]. However, contrasting observations have also been reported. For example, DNM1 has been suggested to function as a protective factor in acute myeloid leukemia [32], but downregulation of DNM1 has been associated with survival outcomes in glioma [33]. These findings highlight the possibility that DNM1 functions as a context-dependent regulator whose role in tumor biology is shaped by tissue-specific signaling networks, cellular differentiation states, and metabolic environments. In addition, recent work has implicated DNM1 in cancer-related systemic phenotypes, such as cognitive impairment, further emphasizing its mul-

tifaceted role in cancer biology [34]. Collectively, these studies reinforce the concept that dynamin-mediated endocytosis is not merely a degradative pathway but a dynamic regulator of receptor signaling and cellular adaptation [13].

Importantly, our findings support a conceptual framework linking dietary lipid overload to hepatocellular carcinoma progression through DNM1-dependent mechanisms. By analyzing a high-fat diet-induced transcriptomic dataset, we identified DNM1 as a lipid-responsive gene that is significantly upregulated under conditions of metabolic stress. This observation suggests that chronic exposure to a high-fat diet may induce DNM1 expression during the pathological continuum from hepatic steatosis to fibrosis and ultimately HCC, reflecting an adaptive response to increased membrane trafficking demand in lipid-rich environments. Beyond its canonical role in membrane fission, dynamin proteins have been implicated in lipid handling processes. Notably, dynamins facilitate lipid absorption in yolk sac endodermal epithelial cells, contributing to cholesteryl ester production, lipid storage, and very-low-density lipoprotein (VLDL) transport [35]. In the context of a high-fat diet, upregulation of DNM1 may therefore enhance hepatocellular lipid uptake, esterification, and lipid transport pathways, potentially exacerbating hepatic steatosis and accelerating disease progression.

Furthermore, we observed that DNM1 expression was inherently higher in HCC patients with viral hepatitis. When stratifying patients, the 'Viral+DNM1 High' subgroup displayed a devastatingly poor prognosis compared to all other groups, including the non-viral groups. We hypothesize that while DNM1 intrinsically promotes tumor cell proliferation and migration (as seen in our non-viral in vitro models), it additionally plays a synergistic role in viral-infected patients. Potentially through its canonical role in endocytosis, DNM1 might facilitate viral entry or downregulate surface expression of immune receptors (such as MHC molecules or cytokine receptors), thereby simultaneously promoting viral persistence and shielding the tumor from cytotoxic T-cells. This dual function positions DNM1 as a critical molecular bridge between basic metabolic oncogenesis and viral immune evasion.

Several limitations of this study should be acknowledged. First, the clinical findings rely heavily on the retrospective TCGA-LIHC dataset; therefore, multi-center prospective cohorts are required to further validate DNM1 as an independent prognostic biomarker. Second, although DNM1 knockdown reduced cellular proliferation and migration, the precise molecular binding partners and downstream signaling pathways (e.g., PI3K/AKT or Wnt/β-catenin) underlying these effects were not investigated. Finally, the association between DNM1 and immune suppression was inferred primarily from GSEA results. Future *in vivo* studies using immunocompetent mouse models with orthotopic HCC are needed to clarify the mechanisms by which DNM1 regulates immune cell infiltration and interferon signaling within the tumor microenvironment. Additionally, HFD-driven HCC models combined with DNM1 genetic manipulation will be necessary to establish the causal relationship between HFD-induced DNM1 activation and HCC progression.

Although our study does not directly establish that HFD-induced DNM1 upregulation accelerates hepatocarcinogenesis *in vivo*, the integration of transcriptomic, clinical, and functional analyses supports a model in which DNM1 represents a critical molecular link between metabolic stress and tumor progression. Future studies using diet-induced HCC models combined with hepatocyte-specific DNM1 deletion or overexpression will be required to determine whether DNM1 is not only a prognostic biomarker but also a functional driver of NAFLD-associated hepatocarcinogenesis.

Acknowledgements

This work was supported by Show Chwan Memorial Hospital (Grant No. SRD-111039 and SRD-111018) and in part by the National Science and Technology Council (NSTC), Taiwan (Grant No. NSTC 115-2321-B-005-009).

Disclosure of conflict of interest

None.

Abbreviations

HCC, Hepatocellular Carcinoma; DNM1, Dynamin-1; TCGA, The Cancer Genome Atlas; LIHC, Liver Hepatocellular Carcinoma; NAFLD, Non-Alcoholic Fatty Liver Disease; HBV, He-

patitis B Virus; HCV, Hepatitis C Virus; GSEA, Gene Set Enrichment Analysis; OS, Overall Survival; HR, Hazard Ratio; CI, Confidence Interval; TME, Tumor Microenvironment; shRNA, Short Hairpin RNA; CCK-8, Cell Counting Kit-8; NES, Normalized Enrichment Score; FBS, Fetal Bovine Serum.

Address correspondence to: En-Pei Isabel Chiang, Department of Food Science and Biotechnology, National Chung Hsing University, Taichung 404, Taiwan. Tel: +886-4-22853049; E-mail: chiangisabel@nchu.edu.tw

References

[1] Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A and Bray F. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin 2021; 71: 209-249.

[2] McGlynn KA, Petrick JL and London WT. Global epidemiology of hepatocellular carcinoma: an emphasis on demographic and regional variability. Clin Liver Dis 2015; 19: 223-238.

[3] Llovet JM, Kelley RK, Villanueva A, Singal AG, Pikarsky E, Roayaie S, Lencioni R, Koike K, Zucman-Rossi J and Finn RS. Hepatocellular carcinoma. Nat Rev Dis Primers 2021; 7: 6.

[4] Rumgay H, Arnold M, Ferlay J, Lesi O, Cabasag CJ, Vignat J, Laversanne M, McGlynn KA and Soerjomataram I. Global burden of primary liver cancer in 2020 and predictions to 2040. J Hepatol 2022; 77: 1598-1606.

[5] Younossi ZM, Koenig AB, Abdelatif D, Fazel Y, Henry L and Wymer M. Global epidemiology of nonalcoholic fatty liver disease-Meta-analytic assessment of prevalence, incidence, and outcomes. Hepatology 2016; 64: 73-84.

[6] Huang DQ, El-Serag HB and Loomba R. Global epidemiology of NAFLD-related HCC: trends, predictions, risk factors and prevention. Nat Rev Gastroenterol Hepatol 2021; 18: 223-238.

[7] Mittal S and El-Serag HB. Epidemiology of hepatocellular carcinoma: consider the population. J Clin Gastroenterol 2013; 47 Suppl: S2-S6.

[8] Younossi Z, Anstee QM, Marietti M, Hardy T, Henry L, Eslam M, George J and Bugianesi E. Global burden of NAFLD and NASH: trends, predictions, risk factors and prevention. Nat Rev Gastroenterol Hepatol 2018; 15: 11-20.

[9] Donnelly KL, Smith CI, Schwarzenberg SJ, Jessurun J, Boldt MD and Parks EJ. Sources of fatty acids stored in liver and secreted via lipoproteins in patients with nonalcoholic fatty liver disease. J Clin Invest 2005; 115: 1343-1351.

[10] Koyama Y and Brenner DA. Liver inflammation and fibrosis. J Clin Invest 2017; 127: 55-64.

[11] Michelotti GA, Machado MV and Diehl AM. NAFLD, NASH and liver cancer. Nat Rev Gastroenterol Hepatol 2013; 10: 656-665.

[12] Silberner J. Prescription for loneliness: five minutes with... Helen Stokes-Lampard. BMJ 2020; 368: m613.

[13] Banushi B, Joseph SR, Lum B, Lee JJ and Simpson F. Endocytosis in cancer and cancer therapy. Nat Rev Cancer 2023; 23: 450-473.

[14] Ferguson SM and De Camilli P. Dynamin, a membrane-remodelling GTPase. Nat Rev Mol Cell Biol 2012; 13: 75-88.

[15] Maxfield FR and McGraw TE. Endocytic recycling. Nat Rev Mol Cell Biol 2004; 5: 121-132.

[16] von Spiczak S, Helbig KL, Shinde DN, Huether R, Pendziwiat M, Lourenço C, Nunes ME, Sarco DP, Kaplan RA, Dlugos DJ, Kirsch H, Slavotinek A, Cilio MR, Cervenka MC, Cohen JS, McClellan R, Fatemi A, Yuen A, Sagawa Y, Littlejohn R, McLean SD, Hernandez-Hernandez L, Maher B, Møller RS, Palmer E, Lawson JA, Campbell CA, Joshi CN, Kolbe DL, Hollingsworth G, Neubauer BA, Muhle H, Stephani U, Scheffer IE, Pena SDJ, Sisodiya SM and Helbig I; Epi4K Consortium; EuroEPINOMICS-RES NLES Working Group. DNM1 encephalopathy: a new disease of vesicle fission. Neurology 2017; 89: 385-394.

[17] Yigit G, Sheffer R, Daana M, Li Y, Kaygusuz E, Mor-Shakad H, Altmüller J, Nürnberg P, Douiev L, Kaulfuss S, Burfeind P, Wollnik B and Brockmann K. Loss-of-function variants in DNM1 cause a specific form of developmental and epileptic encephalopathy only in biallelic state. J Med Genet 2022; 59: 549-553.

[18] Schafer DA. Regulating actin dynamics at membranes: a focus on dynamin. Traffic 2004; 5: 463-469.

[19] Orth JD, Krueger EW, Cao H and McNiven MA. The large GTPase dynamin regulates actin comet formation and movement in living cells. Proc Natl Acad Sci U S A 2002; 99: 167-172.

[20] Lees JG, Gorgani NN, Ammit AJ, McCluskey A, Robinson PJ and O'Neill GM. Role of dynamin in elongated cell migration in a 3D matrix. Biochim Biophys Acta 2015; 1853: 611-618.

[21] Hu M, Gu J, Su W, Zhang Z, Zhu B, Wang Q and Xing C. DNM1: a prognostic biomarker associated with immune infiltration in colon cancer-a study based on TCGA database. Biomed Res Int 2021; 2021: 4896106.

[22] Chen R, Hong R, Chen L, Liu Q, Cao Z, Wang H, Han S and Ke C. Dynamin 1 promotes colorectal cancer progression by activating the PI3K/

Akt signaling pathway. J Transl Med 2025; 24: 212.
[23] Laiman J, Lin SS and Liu YW. Dynamins in human diseases: differential requirement of dynamin activity in distinct tissues. Curr Opin Cell Biol 2023; 81: 102174.
[24] Brereton E, Fassi E, Araujo GC, Dodd J, Telegrafi A, Pathak SJ and Shinawi M. Mutations in the PH Domain of DNM1 are associated with a nonepileptic phenotype characterized by developmental delay and neurobehavioral abnormalities. Mol Genet Genomic Med 2018; 6: 294-300.
[25] Schafer DA. Coupling actin dynamics and membrane dynamics during endocytosis. Curr Opin Cell Biol 2002; 14: 76-81.
[26] Burton KM, Cao H, Chen J, Qiang L, Krueger EW, Johnson KM, Bamlet WR, Zhang L, McNiven MA and Razidlo GL. Dynamin 2 interacts with alpha-actinin 4 to drive tumor cell invasion. Mol Biol Cell 2020; 31: 439-451.
[27] Keum S, Yang SJ, Park E, Kang T, Choi JH, Jeong J, Hwang YE, Kim JW, Park D and Rhee S. Beta-Pix-dynamin 2 complex promotes colorectal cancer progression by facilitating membrane dynamics. Cell Oncol (Dordr) 2021; 44: 1287-1305.
[28] Wong BS, Shea DJ, Mistriotis P, Tuntithavornwat S, Law RA, Bieber JM, Zheng L and Konstantopoulos K. A direct podocalyxin-dynamin-2 Interaction regulates cytoskeletal dynamics to promote migration and metastasis in pancreatic cancer cells. Cancer Res 2019; 79: 2878-2891.
[29] Lin J, Yang J, Xu X, Wang Y, Yu M and Zhu Y. A robust 11-genes prognostic model can predict overall survival in bladder cancer patients based on five cohorts. Cancer Cell Int 2020; 20: 402.
[30] Patel VN, Gokulrangan G, Chowdhury SA, Chen Y, Sloan AE, Koyutürk M, Barnholtz-Sloan J and Chance MR. Network signatures of survival in glioblastoma multiforme. PLoS Comput Biol 2013; 9: e1003237.
[31] Deng Q, Wang F, Song L, Chen L, Huang Y, Guo Z and Yang H. Proteomics-based model for predicting the risk of brain metastasis in patients with resected lung adenocarcinoma carrying the EGFR mutation. Int J Med Sci 2024; 21: 765-774.
[32] Cheng Y, Yang X, Wang Y, Li Q, Chen W, Dai R and Zhang C. Multiple machine-learning tools identifying prognostic biomarkers for acute myeloid Leukemia. BMC Med Inform Decis Mak 2024; 24: 2.
[33] Haghshenas Z, Nazari E, Khalili-Tanha G and Razzaghi Z. Identification of prognostic and diagnostic biomarkers for glioma utilizing immune system gene profiling. Med J Islam Repub Iran 2025; 39: 49.
[34] Ng DQ, Hudson C, Nguyen T, Gupta SK, Koh YQ, Acharya MM and Chan A. Dynamin-1 is a potential mediator in cancer-related cognitive impairment. Neurotherapeutics 2025; 22: e00480.
[35] Tung CT, Lin HJ, Lin CW, Mersmann HJ and Ding ST. The role of dynamin in absorbing lipids into endodermal epithelial cells of yolk sac membranes during embryonic development in Japanese quail. Poult Sci 2021; 100: 101470.