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Epigallocatechin gallate prevents peritoneal fibrosis by inhibiting epithelial-mesenchymal transition in human peritoneal mesothelial cells

Lin Zhang[1,*], Yan Huang[2], Ni Zeng[1], Cangsang Liu[1], Yafen Jiang[1] and Xi Pu[1]
[1]Department of Nephrology, Fourth Changsha Hospital, Changsha, P.R. China;
[2]Department of Emergency, Fourth Changsha Hospital, Institute of Emergency, Changsha, P.R. China

Abstract: The current study set out to elucidate the function of epigallocatechin gallate (EGCG) against peritoneal fibrosis in peritoneal dialysis (PD) patients. Firstly, human peritoneal mesothelial cells (HPMCs) were pretreated with 0, 12.5, 25, 50 or 100μmol/L EGCG. Epithelial-mesenchymal transition (EMT) models were induced by advanced glycation end products (AGEs). Untreated-cells were regarded as the blank control group. Changes in proliferation and migration were analyzed by MTT assay and scratch test and levels of HPMC epithelial and interstitial molecular marker proteins were measured by Western blot assay and immunofluorescence, while trans-endothelial resistance was assessed using an epithelial transmembrane cell resistance meter. Inhibition rates of HPMCs, migration numbers and the levels of Snail, E-cadherin, CK and ZO-1 were all decreased, while the levels of α-SMA and FSP1 and transcellular resistance values were increased in treatment groups ($P<0.05$). With the increase of EGCG concentrations, HPMCs growth inhibition rates and migration numbers, the levels of α-SMA and FSP1 and TER values were decreased and the levels of Snail, E-cadherin, CK and ZO-1 were enhanced ($P<0.05$). Overall, the current study highlights that EGCG effectively inhibits the proliferation and migration of HPMCs, increases permeability, suppresses EMT and ultimately delays peritoneal fibrosis.

Keywords: Epigallocatechin gallate, peritoneal fibrosis, ultrafiltration, epithelial-mesenchymal transition, end-stage renal disease.

INTRODUCTION

Peritoneal dialysis (PD) is one of the leading renal replacement strategies for end-stage renal disease (ESRD) patients, accounting for over 10% of all renal replacement therapies (Hamad *et al.*, 2018). PD exerts its beneficial effects on the quality of life of ESRD patients by removing metabolites and toxic substances and correcting water and electrolyte imbalances (Chuasuwan *et al.*, 2020). As a conduit for blood vessels, lymphatic vessels and nerves, the peritoneum serves to support the abdominal organs (Strippoli *et al.*, 2016). Continuous exposure to hypertonic, high-glucose and acid dialysate solutions can precipitate peritoneal inflammation and injury, further increase the risk of peritoneal fibrosis, angiogenesis and vascular diseases and ultimately lead to failure in PD treatment (Kariya *et al.*, 2018; Roumeliotis *et al.*, 2020). Epithelial-mesenchymal transition (EMT) of human peritoneal mesothelial cells (HPMCs) represents a cytological change of peritoneal fibrosis (Strippoli *et al.*, 2016; Kang *et al.*, 2019). Epigallocatechin gallate (EGCG) is the major monomer and active content of green tea polyphenols and renowned for its antibacterial, antiviral, antioxidant, anti-atherosclerotic, antithrombotic, antiangiogenic, anti-inflammatory and anti-tumor effects (Fujiki *et al.*, 2018; Gan *et al.*, 2018; Wang and Tian, 2018). What's noteworthy is that EGCG can inhibit polypeptide fibrosis and further be transformed into stable oxidation products under neutral and weak alkaline conditions, which produce even stronger effects against protein fibrosis (Sneideris *et al.*, 2019). To the best of our knowledge, there is a scarcity in regard to the role and mechanism of EGCG in EMT during PD.

In light of the same, the current study set out to elucidate the preventive and therapeutic effects of EGCG on peritoneal fibrosis in PD patients, in addition to exploration of the action mechanism of EGCG. Therefore, this study discussed the preventive effect and functional mechanism of EGCG on peritoneal fibrosis in PD patients with the expectation to supply theoretical reference on clinical application.

MATERIALS AND METHODS

Cell culture

HPMCs (LMAI Bio, Shanghai, P.R. China) were seeded and maintained in high-glucose DMEM medium (PM150210B; Procell, Wuhan, China) supplemented with 10% FBS (fetal bovine serum). Flasks were cultured in a humidified incubator at 37°C with an atmosphere of 5% CO_2. Upon reaching 80% confluency, the cells were passaged after trypsinization (2.5%; CC0134; Leagene, Beijing, China) and the medium was replenished every 24h. Next, the HPMCs were incubated with EGCG (dosages of 0, 12.5, 25, 50 or 100μmol/L; Sigma-Aldrich Inc., St. Louis, MO, USA) for 60 minutes, prior to

*Corresponding author: e-mail: 397639438@qq.com

treatment with advanced glycation end products (AGEs; EY-D1072; Shanghai Yiyan Biological Technology, Shanghai, China) at a dosage of 500μg/mL for 72 hours. Afterwards, the cells were collected for testing. Untreated cells were adopted as the blank control group.

MTT assay

HPMCs at good growth state were digested with trypsin and resuspended in PBS. The cell suspension (100μL; 5×10^3 cells) was subsequently added into 96-well cell culture plates. Next, the wells were labeled as 0h, 24 h and 48h and 3 replicates were set for each condition. Cells were cultivated at 37℃ with 5% CO_2 in air for 24 h or 48 h prior to the addition of 10μL MTT solution. Afterwards, these cells were cultivated at 37℃ with 5% CO_2 in air for an additional 4h before discarding the supernatant. Following the addition of 150μL DMSO, the plates containing cells were uniformly-mixed for 600 seconds before determining optical density (OD) under 490 nm. Inhibition rate of cell growth (%) = (1 - OD of experimental group / OD of blank group) × 100%. Each test was carried out thrice to obtain the mean value.

Scratch testg

HPMCs (100μL; 5×10^4 cells) were added into 6-well cell culture plates and labeled as 0 h, 24 h and 48 h. Three replicate wells were set for each condition. The cells were cultured at 37ºC with 5% CO_2 in air overnight. The following day, the cell layer was scratched with a pipette tip and rinsed thrice with PBS. After removing the detached cells, fresh medium was added, followed additional incubation at 37ºC with 5% CO_2. At 0h, 24h and 48h, the scratches were photographed at four randomly chosen fields.

Western blot assay

HPMCs were lysed with 200μL cell lysis solution for 30 min. Next, the lysed cells were precipitated by centrifugation (4ºC; 12,000 rpm) for 10 min. The upper clear liquid above the cell sediments was subsequently tested for the concentration of protein with the help of BCA kits (P0012; Beyotime, Shanghai, P.R. China). The cells were denaturized with boiling in water for 5 min in the presence of sodium dodecyl sulfate loading buffer (5×). Later, 10% SDS-PAGE was performed to process 20 microgram samples at 100 V, after which the proteins were transferred onto polyvinylidene difluoride membranes (100V, 2h, 0ºC) blocked with skimmed milk (5%; 25˚C; one hour). Rabbit anti-human Snail (diluted a 1000-fold; Abcam, Cambridge, UK), E-cadherin (diluted a 1000-fold; Abcam), CK (diluted a 1000-fold; Abcam), ZO-1 (dilution ratio of 1:1,000; Abcam), α-SMA (diluted a 1000-fold; Abcam), FSP1 (diluted a 1000-fold; Abcam) or β-actin (diluted 5000-fold; Abcam) polyclonal primary antibodies were incubated with the membranes at 4ºC for 16 hours. Later, the secondary antibody (goat anti-rabbit horseradish peroxidase-conjugated; diluted 3000-fold; Abcam) was further incubated with the membranes at 25˚C for one hour. After washing, enhanced chemiluminescence detection kits (Solarbio, Beijing, China) were adopted to develop the membranes, which were photographed and analyzed with a Gel Image Processing System (1600 series; Tanon, Shanghai, China). The contents of target proteins were calculated relative to that of β-actin.

Immunofluorescence

HPMCs were seeded onto coverslips. After reaching confluency, the cells on coverslips were rinsed with PBS twice before fixation with 4% paraformaldehyde for 600 seconds. Next, the cells were rinsed with PBS for 300 seconds and then perforated with 0.5% Triton for 900 seconds, followed by another two rinses with PBS. Following blockade with 1% BSA for 30 min, the primary antibody was added to the cells and cultured at 37^{O}C for two hours. Subsequently, the cells were rinsed again with PBS two times and treated with fluorescence-labelled secondary antibody at 37℃ for one hour. Following another two rinses with PBS, the cells were stained with 5% DAPI (2 min) and then mounted. Fluorescent photos of the cells were obtained using a fluorescence microscope (Olympus, Tokyo, Japan).

Transcellular resistance test

An epithelial transmembrane cell resistance meter (Millipore, Merck, Billerica, MA, USA) was utilized for transcellular resistance test. Briefly, 12-well plates (outer well) were used in combination with Millicell wells (inner well) to culture cells at 37^{O}C overnight. HPMCs were digested with trypsin and resuspended in PBS. Next, the HPMCs (5×10^4 cells/cm^2) were seeded into inner wells, followed by incubation at 37ºC with 5% CO_2 in air. Three replicate wells were set for each condition. The resistance was determined at 0 h and 24h, respectively. For single-layer HPMCs, the transcellular resistance (TER)= (resistance-basic resistance)/bottom area of Millicell wells. The resistance value after power on was the basic resistance value.

STATISTICAL ANALYSIS

Measurement data in the present study were analyzed using the SPSS 22.0 software (IBM, Armonk, NY, USA). In addition, measurement data were illustrated as *means* ± *standard deviations* and underwent normality test. One-way ANOVA was adopted to compare differences among groups, followed by the Student-Newman-Keuls test. A value of $p<0.05$ was regarded statistically significant.

RESULTS

EGCG suppresses HPMCs proliferation in a concentration-dependent manner

In order to examine how EGCG affects HPMCs proliferation, the inhibition rate of cell growth was

examined with a MTT assay. Cell growth inhibition rates after treatment with different concentrations of EGCG were markedly smaller than those in blank control ($P < 0.05$) and the rates of cell growth inhibition were significantly different among various dosage groups ($P< 0.05$) (table 1). Overall, these findings indicated that EGCG inhibits the proliferation of HPMCs in a dose-dependent manner.

EGCG reduces the number of migrated HPMCs according to dose

The effects of EGCG on the migration of HPMCs were assessed with a Scratch test. The numbers of migrated cells after treatment with different doses of EGCG were markedly lower than those in the blank control ($P<0.05$) and the number of migrated cells were significantly different among various dosage groups ($P<0.05$) in a dose-dependent manner (fig. 1). Together, these findings highlighted that EGCG reduces the number of migrated HPMCs in a dose-dependent manner.

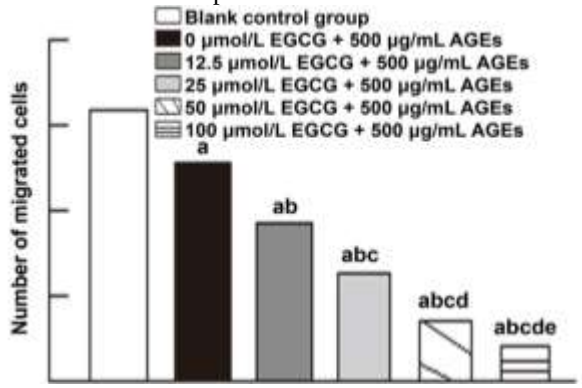

Fig. 1: EGCG affects migration of HPMCs. Note: a, $P<0.05$ v.s. blank control; b, $P<0.05$ v.s. 0μmol/L EGCG+500μg/mL AGEs group; c, $P<0.05$ v.s. 12.5μmol/L EGCG+500μg/mL AGEs group; d, $P<0.05$ v.s. 25μmol/L EGCG+500μg/mL AGEs group; e, $P<0.05$ v.s. 50μmol/L EGCG+500μg/mL AGEs group.

Treatment with EGCG enhances expression of epithelial cell molecular marker proteins in HPMCs

The effects of EGCG on the expression of epithelial cell molecular marker proteins in HPMCs were assessed with a Western blot assay. Relative to the blank control group, the levels of Snail, E-cadherin, CK and ZO-1 protein expression were significantly reduced in all treatment groups ($P<0.05$) and the expression of these proteins was significantly increased with increasing dosages of EGCG ($P<0.05$; fig. 2). Together, these findings suggested that treatment with EGCG promotes epithelial cell molecular marker protein expression levels in HPMCs.

Treatment with EGCG reduces the expression of interstitial cell molecular marker proteins in HPMCs

Additionally, the effects of EGCG on the expression of interstitial cell molecular marker protein levels in HPMCs were further explored with a Western blot assay. Compared to the blank control group, α-SMA and FSP1 levels were markedly enhanced in all treatment groups ($P<0.05$) and the levels of these proteins were drastically decreased with increasing dosages of EGCG ($P<0.05$; fig. 3). Overall, these findings highlighted that EGCG treatment diminishes the expression of interstitial cell molecular marker proteins in HPMCs.

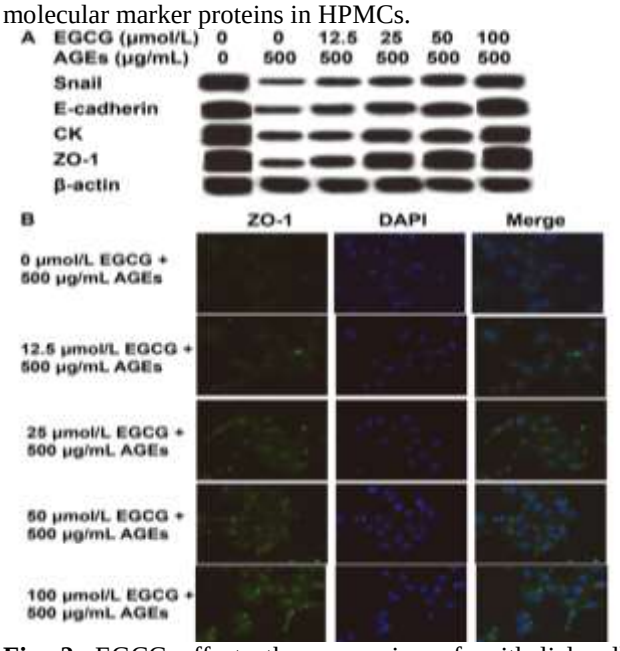

Fig. 2: EGCG affects the expression of epithelial cell molecular marker proteins in HPMCs, which was determined by (A) Western blotting and (B) immunofluorescence. Note: a, $P<0.05$ v.s. blank control; b, $P<0.05$ v.s. 0μmol/L EGCG + 500μg/mL AGEs group; c, $P<0.05$ v.s. 12.5μmol/L EGCG + 500μg/mL AGEs group; d, $P<0.05$ v.s. 25μmol/L EGCG + 500μg/mL AGEs group; e, $P<0.05$ v.s. 50μmol/L EGCG + 500μg/mL AGEs group.

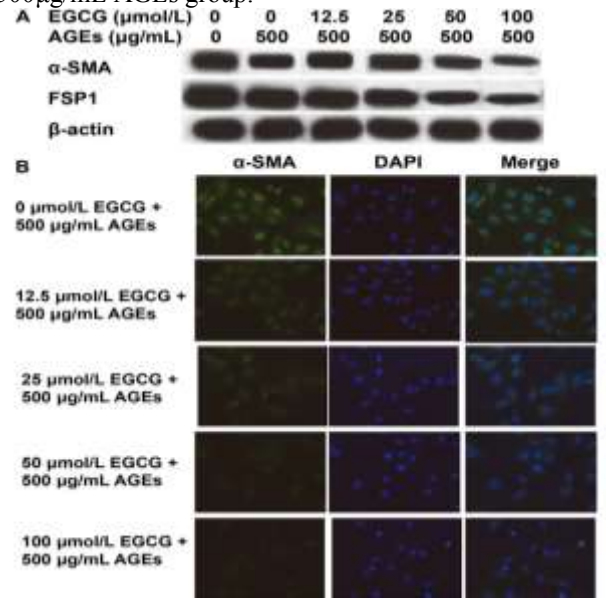

Fig. 3: EGCG affects the expression of interstitial cell molecular marker proteins in HPMCs, which was

determined by (A) Western blotting and (B) immunofluorescence. Note: a, P<0.05 v.s. blank control group; b, P<0.05 v.s. 0μmol/L EGCG+500μg/mL AGEs group; c, P<0.05 v.s. 12.5μmol/L EGCG+500μg/mL AGEs group; d, P<0.05 v.s. 25μmol/L EGCG+500μg/mL AGEs group; e, P<0.05 v.s. 50μmol/L EGCG+500μg/mL AGEs group.

EGCG decreases the trans-endothelial resistance of HPMCs in a dose-dependent manner

Lastly, the influence of EGCG on the trans-endothelial resistance of HPMCs was assessed using an epithelial transmembrane cell resistance meter. The TER values for HPMCs in different dosage groups were markedly greater than those in the blank control group (P<0.05) (table 2). In addition, the TER values were decreased with increasing dosages of EGCG (P<0.05) (table 2). Together, these findings highlighted that EGCG decreases trans-endothelial resistance of HPMCs in a dose-dependent manner.

DISCUSSION

ESRD, also known as uremia, represents the terminal stage of renal disease that can be caused by various etiologies (Lu *et al.*, 2022). The incidence rate of ESRD has been on a steady-incline in line with increases in the incidence rate of hypertension, atherosclerosis and diabetes and the proportion of elderly populaces (Abu-Odah *et al.*, 2017). In the pathogenesis of ESRD, patients not only present with water and electrolyte metabolic disorders and acid-base imbalance, but also exhibit a series of uremic symptoms due to retention of large amounts of metabolic end products and toxic substances and can cause deteriorating effects on their physical and mental health (Shen and Norris, 2016). Although renal transplantation is regarded as the gold-standard treatment for ESRD, dialysis has been widely-adopted as a renal replacement therapy for ESRD patients due to the lack of donors (Radić *et al.*, 2010). Peritoneal dialysis (PD) is a well-established and cost-effective form of such treatment and based on the infusion of a sterile solution into the peritoneal cavity through a catheter to remove toxins and excessive water using the peritoneal membrane as the exchange surface (Yuan *et al.*, 2020). Moreover, mounting evidence suggests that PD can effectively protect the residual renal function of ESRD patients, realize satisfactory clinical effects and also has a high safety profile (Sukul *et al.*, 2019). On the other hand, ultrafiltration failure represents a severe complication caused by the loss of ultrafiltration function of peritoneum and not only the key reason why PD patients withdraw from treatment, but also an important cause of death (Kim, 2009). Unsurprisingly, the incidence of ultrafiltration failure increases with chronic PD duration and almost 50% patients with PD develop ultrafiltration for over 6 years (Aguirre and Abensur, 2011), which underscores the correlation between prolongation of PD and incidence of ultrafiltration failure. Interestingly, the hard-done work of our peers has indicated that peritoneal fibrosis serves as a crucial factor in the pathogenesis of ultrafiltration filtration (Kariya *et al.*, 2018; Li *et al.*, 2019). Finding an effective way to prevent and treat PD has become an urgent problem to be solved in clinic.

EGCG is a catechin monomer isolated from green tea and well-known for its antioxidant, anticancer and antimutagenic properties (Snitsarev *et al.*, 2013). HPMCs are an important component of the peritoneum and as such confer roles in PD and peritoneal fibrosis (Elsurer *et al.*, 2010; Kang *et al.*, 2019). Findings uncovered in our study demonstrated that EGCG inhibits the proliferation and migration of HPMCs, suggesting that EGCG can repair the damage of HPMCs caused by AGEs and hinder the process of peritoneal fibrosis *via* inhibition of HPMC proliferation and migration. EMT is a biological process wherein epithelial cells transform into mesenchymal phenotype cells through specific procedures, whereas the occurrence of EMT in PD has been previously indicated in the progression of peritoneal fibrosis (Strippoli *et al.*, 2016). What's more, EGCG can protect against renal tubular EMT induced by oxalic acid through the Nrf2 pathway (Kanlaya *et al.*, 2016). Moreover, EGCG was recently shown to inhibit thyroid cancer cell invasion and EMT *via* blockade of the TGF-β1/Smad pathway (Li *et al.*, 2019). Additionally, EMT is associated with an increase in epithelial cell molecular marker protein expression and decreased mesenchymal cell molecular marker protein expressions (María *et al.*, 2003). During the course of our study, we discovered that following treatment with EGCG, the expressions of Snail, E-cadherin, CK and ZO-1 proteins were all diminished and the levels of α-SMA and FSP1 proteins were all enhanced relative to the blank control group, which in line with previous reports. Furthermore, increasing dosages of EGCG brought about a gradual enhancement in the expression levels of Snail, E-cadherin, CK and ZO-1 proteins, while the levels of α-SMA and FSP1 were gradually decreased, highlighting that EGCG can inhibit the EMT of HPMCs by up-regulating the expression of epithelial cell molecular marker proteins and down-regulating the expression of mesenchymal cell molecular marker proteins in a dose-dependent manner. Meanwhile, trans-endothelial resistance is a widely-used index for the evaluation of permeability of endothelial cells (Srinivasan *et al.*, 2015). Accumulating studies have indicated that peritoneal fibrosis can decrease peritoneal permeability, increase matrix accumulation and reduce PD efficacy (Zhou *et al.*, 2016; Yanai *et al.*, 2018). Herein, we found that in contrast to the blank control group, treatment with EGCG led to an increase in TER values in the experimental groups, which is again in accordance with previous research results. Lastly, increasing dosages of EGCG led to a gradual decline in the TER values,

Table 1: Effect of EGCG on the proliferation of HPMCs (means ± standard deviations).

Groups	Cell growth inhibition rate (%)	
	24 h	48 h
Blank control	0	0
0 μmol/L EGCG + 500 μg/mL AGEs	0	0
12.5 μmol/L EGCG + 500 μg/mL AGEs	83.24 ± 4.68^{ab}	76.53 ± 4.54^{ab}
25 μmol/L EGCG + 500 μg/mL AGEs	59.63 ± 4.52^{abc}	52.34 ± 4.33^{abc}
50 μmol/L EGCG + 500 μg/mL AGEs	44.56 ± 4.35^{abcd}	42.42 ± 4.21^{abcd}
100 μmol/L EGCG + 500 μg/mL AGEs	36.22 ± 4.21^{abcde}	28.25 ± 4.13^{abcde}
F	246.984	258.48
P	< 0.01	< 0.01

Table 2: Effect of EGCG on the trans epithelial resistance of HPMCs (means ± standard deviations)

Groups	TER value ($\Omega \cdot cm^2$)
Blank control	7.53 ± 2.24
0 μmol/L EGCG + 500 μg/mL AGEs	34.25 ± 2.25
12.5 μmol/L EGCG + 500 μg/mL AGEs	28.72 ± 2.18^{ab}
25 μmol/L EGCG + 500 μg/mL AGEs	24.53 ± 2.21^{abc}
50 μmol/L EGCG + 500 μg/mL AGEs	16.36 ± 1.86^{abcd}
100 μmol/L EGCG + 500 μg/mL AGEs	11.23 ± 1.75^{abcde}
F	38.745
P	< 0.01

Note: a, *P*<0.05 v.s. blank control group; b, *P*<0.05 v.s. 0μmol/L EGCG+500μg/mL AGEs group; c, *P*<0.05v.s. 12.5μmol/L EGCG+500μg/mL AGEs group; d, *P*<0.05 v.s. 25μmol/L EGCG+500μg/mL AGEs group; e, *P*<0.05 v.s. 50μmol/L EGCG+500μg/mL AGEs group.

suggesting that EGCG can effectively enhance the permeability of HPMCs, improve dialysis efficiency and attenuate peritoneal fibrosis.

CONCLUSION

Collectively, findings uncovered in the current study highlight that EGCG can effectively inhibit the proliferation and migration of HPMCs. In addition, through up-regulating epithelial cell molecular marker protein expression and down-regulating expression of mesenchymal cell molecular marker proteins, EGCG increases the permeability of HPMCs, inhibits the EMT of HPMCs and delays peritoneal fibrosis *via* up-regulation of epithelial cell molecular marker protein expression and down-regulation of mesenchymal cell molecular marker protein expression. However, the study is only an *in vitro* experiment and presents with its certain limitations. Further research incorporating *in vivo* experiments is warranted to validate the clinical application of our findings.

REFERENCES

Abu-Odah H, Abed Y and El-Nems K (2017). Factors associated with end-stage renal disease in the gaza strip: A case-control study. *The Lancet.*, **390**: S34.

Aguirre AR and Abensur H (2011). Protective measures against ultrafiltration failure in peritoneal dialysis patients. *Clinics (Sao Paulo).*, **66**(12): 2151-2157.

Chuasuwan A, Pooripussarakul S, Thakkinstian A, Ingsathit A and Pattanaprateep O (2020). Comparisons of quality of life between patients underwent peritoneal dialysis and hemodialysis: A systematic review and meta-analysis. *Health Qual Life Outcomes.*, **18**(1): 191.

Elsurer R, Afsar B, Sezer S and Ozdemir FN (2010). Peritoneal cells at admission: Do they have prognostic significance in peritonitis? *Ren Fail.*, **32**(3): 335-342.

Fujiki H, Watanabe T, Sueoka E, Rawangkan A and Suganuma M (2018). Cancer prevention with green tea and its principal constituent, egcg: From early investigations to current focus on human cancer stem cells. *Molecules and cells.*, **41**(2): 73-82.

Gan RY, Li HB, Sui ZQ and Corke H (2018). Absorption, metabolism, anti-cancer effect and molecular targets of epigallocatechin gallate (egcg): An updated review. *Critical reviews in food science and nutrition.*, **58**(6): 924-941.

Hamad A, Ismail H, Elsayed M, Kaddourah A, Ahmed H, Ibrahim R, Ali A and Alali F (2018). The epidemiology of acute peritonitis in end-stage renal disease patients on peritoneal dialysis in qatar: An 8-year follow-up study. *Saudi J Kidney Dis Transpl.*,, **29**(1): 88-94.

Kang SH, Kim SW, Kim KJ, Cho KH, Park JW, Kim CD and Do JY (2019). Effects of tranilast on the epithelial-to-mesenchymal transition in peritoneal mesothelial cells. *Kidney Res Clin Pract.*, **38**(4): 472-480.

Kanlaya R, Khamchun S, Kapincharanon C and Thongboonkerd V (2016). Protective effect of epigallocatechin-3-gallate (egcg) via nrf2 pathway against oxalate-induced epithelial mesenchymal transition (emt) of renal tubular cells. *Sci Rep.*, **6**: 30233.

Kariya T, Nishimura H, Mizuno M, Suzuki Y, Matsukawa Y, Sakata F, Maruyama S, Takei Y and Ito Y (2018). Tgf-β1-vegf-a pathway induces neoangiogenesis with peritoneal fibrosis in patients undergoing peritoneal dialysis. American journal of physiology. *Am. J. Physiol Renal Physiol.*, **314**(2): 167-180.

Kim YL (2009). Update on mechanisms of ultrafiltration failure. *Peritoneal dialysis international: Perit Dial Int.*, **29**(Suppl 2): 123-127.

Li T, Zhao N, Lu J, Zhu Q, Liu X, Hao F and Jiao X (2019). Epigallocatechin gallate (egcg) suppresses epithelial-mesenchymal transition (emt) and invasion in anaplastic thyroid carcinoma cells through blocking of tgf-β1/smad signaling pathways. *Bioengineered.*, **10**(1): 282-291.

Li X, Liu H, Sun L, Zhou X, Yuan X, Chen Y, Liu F, Liu Y and Xiao L (2019). Microrna-302c modulates peritoneal dialysis-associated fibrosis by targeting connective tissue growth factor. *J Cell Mol Med.*, **23**(4): 2372-2383.

Lu J, Shi D, Zhao X, Xi M, Qi H and He Q (2022). Comparative Analysis of Efficacy and Prognosis of Hemodialysis and Peritoneal Dialysis for End-Stage Renal Disease: A Meta-analysis. *Comput Math Methods Med.*, 6007698.

Maria, Yanez-Mo, Enrique, Lara-Pezzi, Rafael, Selgas, Marta, Ramírez-Huesca, Carmen and Domínguez-Jiménez (2003). Peritoneal dialysis and epithelial-to-mesenchymal transition of mesothelial cells. *N Engl J Med*. 348(5): 403-413.

Radic J, Ljutic D, Radic M, Kovacic V, Sain M and Curkovic KD (2010). The possible impact of dialysis modality on cognitive function in chronic dialysis patients. *Neth J Med.*, **68**(4): 153-157.

Roumeliotis S, Dounousi E, Salmas M, Eleftheriadis T and Liakopoulos V (2020). Unfavorable effects of peritoneal dialysis solutions on the peritoneal membrane: The role of oxidative stress. *Biomolecules.*, **10**(5): 768.

Shen JI and Norris KC (2016). Esrd payment reform: First do no harm. *J Am Soc Nephrol.*, **27**(10): 2924-2926.

Sneideris T, Sakalauskas A, Sternke-Hoffmann R, Peduzzo A, Ziaunys M, Buell AK and Smirnovas V (2019). The environment is a key factor in determining the anti-amyloid efficacy of EGCG. *Biomolecules.*, **9**(12): 855.

Snitsarev V, Young MN, Miller RM and Rotella DP (2013). The spectral properties of (-)-epigallocatechin 3-O-gallate (EGCG) fluorescence in different solvents: dependence on solvent polarity. *PloS one.*, **8**(11): 79834.

Srinivasan B, Kolli AR, Esch MB, Abaci HE, Shuler ML and Hickman JJ (2015). TEER measurement techniques for *in vitro* barrier model systems. *J Lab Autom.*, **20**(2): 107-126.

Strippoli R, Moreno-Vicente R, Battistelli C, Cicchini C, Noce V, Amicone L, Marchetti A, Del Pozo MA and Tripodi M (2016). Molecular mechanisms underlying peritoneal emt and fibrosis. *Stem Cells Int.*, 3543678.

Sukul N, Zhao J, Fuller DS, Karaboyas A, Bieber B, Sloand JA, Subramanian L, Johnson DW, Oliver MJ, Tungsanga K, Tomo T, Morton RL, Morgenstern H, Robinson BM and Perl J (2019). Patient-reported advantages and disadvantages of peritoneal dialysis: Results from the pdopps. *BMC Nephrol.*, **20**(1): 116.

Wang L and Tian X (2018). Epigallocatechin-3-gallate protects against homocysteine-induced brain damage in rats. *Planta Med.*, **84**(1): 34-41.

Yanai K, Ishii H, Aomatsu A, Ishibashi K and Morishita Y (2018). Micrornas in peritoneal fibrosis: A systematic review. *Discov Med.*, **26**(145): 271-280.

Yuan X, Li H, Zhou L and Huang Y (2020). A bibliometric analysis of the 100 most influential papers on peritoneal dialysis. *Medicine (Baltimore).*, **99**(47): e23115.

Zhou Q, Bajo MA, Del Peso G, Yu X and Selgas R (2016). Preventing peritoneal membrane fibrosis in peritoneal dialysis patients. *Kidney Int.*, **90**(3): 515-524.