ORIGINAL ARTICLE

Correlation between signal transducer and activator of transcription 1 (STAT 1), vascular endothelial growth factor (VEGF), and signal to noise ratio (SNR) value in otoacoustic emission (OAE) examination on organ of Corti cochlea due to cisplatin

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ABSTRACT

Aim To assess the cochlear damage caused by cisplatin in the rat cochlea based on decreased signal to noise ratio (SNR) values on otoacoustic emission (OAE) examination and increased expression of signal transducer and activator of transcription 1 (STAT 1) and vascular endothelial growth factor (VEGF) on immunohistochemical examination.

Methods Twenty-four *Rattus norvegicus* were divided into 4 groups and injected with 8 mg/kgBW of cisplatin intraperitoneally except for control group. The SNR on OAE examination were checked before the treatment and on day 3, 4, and 7 after the treatment. The cochleas were stained immunohistochemically, followed by assessment of the cochlear organ of Corti damage based on STAT 1 and VEGF expression.

Results A decrease in the mean of SNR value was found in accordance with the length of cisplatin exposure. The STAT 1 and VEGF expression increased with duration of cisplatin exposure. A correlation was found between SNR values, STAT 1, and also VEGF expression ($p<0.05$).

Conclusion An increase of STAT 1 and VEGF expression influences cochlear damage due to cisplatin administration. There was a correlation between STAT 1 and VEGF expression with SNR values in the cochlear organ of Corti of *Rattus norvegicus* exposed to cisplatin.

Key words: apoptosis, experimental, immunohistochemistry, rat

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INTRODUCTION

Ototoxicity is hearing impairment caused by damage to inner ear after ototoxic drug therapy administration. Several drugs are known to be ototoxic agents, such as aminoglycoside, macrolides, vancomycin, loop diuretics, quinine, nonsteroidal anti-inflammatory drugs (NSAIDs), antiretrovirals, (1,2) and platinum-based chemotherapy agents (3).

Cisplatin is a platinum chemotherapeutic agent which is widely used in treatment of various malignant tumours (4). Cisplatin ototoxicity causes progressive hearing impairment in high frequencies and leads to inner ear damage when administered in high, cumulative doses (5).

Increased hearing threshold was found in 75-100% patients treated with cisplatin (6). The prevalence of cisplatin ototoxicity is around 80% with a decreased hearing threshold of at least 20 dB (5). The prevalence of hearing loss due to cisplatin in children ranges from 22-77% (7).

From *in vitro* animal studies, cisplatin was found to interact with cochlear tissue such as outer hair cells in the organ of Corti, stria vascularis, spiral ligament, and spiral ganglionic cells (6). Cisplatin will undergo hydrolysis in blood to form cisdiamine-aquachloro-platinum (II), which is the main metabolite that plays a role in cytotoxic activity (6). The entry of cisplatin into cells is facilitated by several different transporters such as mammalian copper transporter 1 (CTR 1), while copper-activated P-type ATPases (ATP7 A and ATP7B) function to regulate the release of cisplatin from within cell (6). Exposure of cisplatin to outer hair cells will produce excessive amount of reactive oxygen species (ROS) (8). The ROS are reactive chemical molecules containing oxygen that can be expressed higher as a result of stress due to toxin exposure, radiation, and a disease leading to formation of local oxidative stress and cellular responses (9). Cochlear closed structure makes toxins trapped inside it. This will lead to ROS accumulation and antioxidant system decrease that will cause cell injury and also apoptosis (6).

ROS will also produce signal transducer and activator of transcription 1 (STAT1), which will induce the inflammatory process. The activation of STAT 1 is also associated with the activation of p53 enzyme which causes apoptosis of cochlear cells (7). The increase in vascular endothelial growth factor (VEGF) can be triggered by the presence of cell hypoxia both *in vitro* and *in vivo*. The increase in VEGF is also thought to occur due to an increase in ROS (10).

A study using experimental animals found that cisplatin causes the destruction to the outer hair cells starting from cochlear basal turn (4). The destruction starts from the first row of outer hair cells, followed by inner hair cells to stria vascularis (4). This indicates that ototoxicity starts at high frequencies and can progress to low frequencies (1).

Otoacoustic emission examination (OAE) is an objective measurement to assess cochlear function, because OAE is an acoustic response produced by outer hair cells of cochlea either spontaneously or with sound stimulation (11).

The aim of this study was to investigate a correlation between STAT 1, VEGF, and SNR value in OAE examination on organ of Corti cochlea due to cisplatin. This study is intended to prove the role of STAT 1 and VEGF in the prevalence of cisplatin ototoxicity.

MATERIAL AND METHODS

Materials and study design

This random, double blind, *ex vivo* experimental laboratory study with posttest control group study was done in a standardized laboratory at the Animal House Laboratory of the Faculty of Mathematics and Natural Sciences, Universitas Sumatera Utara in the period between September 2020 and February 2021. Immunohistochemical examination was done at the Anatomical Pathology Department, Universitas Sumatera Utara.

Before any procedures, we submitted ethical feasibility assessment and the approval was obtained from the Research Ethics Commission of Universitas Sumatera Utara (No:223/KEP/USU/2020).

The sample had a similar population strain and was homogeneous in gender and age. The rats used were *Rattus norvegicus* Wistar male strain, healthy condition, aged 2-3 months, weighing 200-250 grams. Dropout criteria were applied if the subject become sick or died thus could not fulfil the research procedure.

Methods

Cisplatin was injected to experimental animals intraperitoneally at the dose of 8 mg/kgBW. Twenty-four rats were divided into 4 treatment groups. Group 1- control group did not get any treatment of cisplatin, samples injected with NaCl 0.9% at the dose 8 mg/kgBW, and OAE examination was performed before the treatment and 7 days after; Group 2: OAE examination was performed before the treatment and on the third day after the treatment; Group 3: OAE examination was performed before the treatment and on the fourth day after the treatment; Group 4: OAE examination was performed before the treatment and on the seventh day after the treatment. All samples had to meet criteria where the SNR value before the treatment was categorized as a normal value (SNR≥6 dB) to guarantee the outer hair cells function was not damaged before.

Samples were terminated and stained immunohistochemically, followed by assessment of the cochlear organ of Corti damage based on VEGF and STAT1 expression.

Prior to the OAE examination, experimental animals were anesthetized with ketamine hydrochloride 50 mg/kgBW combined with xylazine 7.5 mg/kgBW intraperitoneally (12). After the experimental animals became more calmed, ear canal and tympanic membrane were inspected using an otoscope. It was ensure that the ear canal was clean, no abnormalities in the ear canal or tympanic membrane. If any abnormalities were found, animals were excluded. OAE examination (Grason-Stadler Corti, USA) was performed on both ears using a probe that matched the ear canal of an experimental animal. Signal to noise ratio (SNR) was assessed at frequency of 1.5-12 kHz. The results of OAE examination were considered pass if acoustic emission value was ≥6 dB from background noise at the same frequency.

Experimental animals were terminated using carbon dioxide (CO2) inhalation. The administration of CO2 gas can rapidly cause loss of consciousness before inducing nociceptive distress and respiratory distress (13). Following the termination, the temporal bone tissue was taken, and fixed with formalin buffer solution, decalcified with 10% ethylenediaminetetraacetic acid (EDTA) for 4 weeks. The tissue was washed with phosphate buffer saline (PBS) and fixed with formalin buffer solution. The tissue was dehydrated with graded alcohol, cleared with xylene twice and infiltrated with soft paraffin for 60 minutes at 48 °C and blocked-in hard paraffin in the embedding cassette, and left for 24 hours. The tissue was cut with a microtome into 4µm thickness subsequently after the block was cooled using a cold plate to facilitate cutting. The tissue was put in warm water at 47 °C and heated on a warm plate at 65 °C after being placed on an object glass. Furthermore, the object glass that contained sample tissue was immersed in xylene to remove the remaining paraffin in the tissue, and rehydrated with graded alcohol then rinsed with distilled water (dH2O) and hematoxylin-eosin staining was done on the tissue.

The cochlear tissue was examined immunohistochemically. The object glass was put in xylol, rehydrated with graded alcohol (absolute, 96%, 80%, 70%, 50% and 30%), and rinsed with buffer solution. Blocking nonspecific protein at room temperature and washed with buffer solution, and incubated with STAT1 polyclonal antibody (no. E-AB-32977; Elabscience, USA), after that washed with buffer solution. Samples were incubated with secondary antibody (Ultratek Anti-polyvalent), washed with buffer solution, incubated with Ultra Tek HRP at room temperature, washed with buffer solution and dH_2O. Diamino benzidine (DAB) was dripped on the sample and incubated in the humidified chamber at room temperature, counterstained using Mayer's Hematoxylin solution then washed with dH_2O. The sample was dripped with bluing reagent and washed with dH_2O, dehydrated with xylene and dried. The sample was covered with a cover glass, then observed using a light microscope (Olympus CX 21, Tokyo, Japan).

The distribution assessment was carried out by calculating the percentage of brown stained cells in the entire field of view using 10x magnification. To assess STAT 1 and VEGF expression, the IRS (immunoreactive score) formula was used by multiplying the percentage of brown stained cells by the intensity of the stain. The distribution of brown stained cells was divided into 4 categories: 0 = no brown stained cells, 1 = <10% cells stained brown, 2 = 11-50% cells stained brown, 3 = 51-80 % cells stained brown, 4 = >81% cells

stained brown. The intensity of the brown stained cells was assessed by the following categories: 0 = no stain, 1 = weak intensity, 2 = moderate intensity, 3 = strong intensity. The calculation results could show a minimum score of 0 and a maximum score of 12 (14). The calculation results were done by two people (author and anatomical pathology specialist).

Statistical Analysis

Mean and median values of SNR, STAT 1 and VEGF expression were analysed to shaw the difference among groups. Multivariate test (ANOVA and Post-hoc Bonferonni test for SNR variable, Kruskal Wallis and Post-Hoc Mann-Whitney test for STAT 1 and VEGF variable) was performed to see which treatment groups had significant differences followed by the bivariate Spearman test to analyse a correlation between SNR values with STAT 1 and VEGF expression for each group.

RESULTS

The largest decrease in SNR values was found in Group 4 where experimental animals were exposed to cisplatin and terminated on the 7th day. This is inversely proportional to the increase in STAT 1 and VEGF expression values, which showed the highest expression in group 4 followed by group 3 and group 2 (Table 1).

Table 1. Differences in the mean and median values of signal to noise ratio (SNR), vascular endothelial growth factor (VEGF), and signal transducer and activator of transcription 1 (STAT1) among four groups

	Group*	Mean ± SD	Median (Min-Max)
SNR	Group 1	16.84±0.98	16.50 (15.83-18.33)
	Group 2	2.70±0.40	2.75 (2-3.17)
	Group 3	1.54±0.43	1.38 (1.17-2.33)
	Group 4	1.44±0.61	1.62 (0.58-2.17)
VEGF	Group 1	1.50±0.54	1.5 (1-2)
	Group 2	2.67±0.816	2.5 (2-4)
	Group 3	5.17±1.83	5 (3-8)
	Group 4	5.67±2.42	5 (3-9)
STAT1	Group 1	1.50±0.55	1.5 (1-2)
	Group 2	2.67±0.82	2.5 (2-4)
	Group 3	4.67±1.50	5 (3-6)
	Group 4	6.33±2.25	6 (4-9)

*Group 1: Control group; Group 2: otoacoustic emission (OAE) and immunohistochemistry examination on day 3 after treatment; Group 3: OAE and immunohistochemistry examination on day 4 after treatment; Group 4 OAE and immunohistochemistry examination on day 7 after treatment

The increase in STAT 1, also VEGF expression along with the length of cisplatin exposure in experimental animals was noticed. The intensity of brown colour was the strongest and broadest in group 4 compared to other groups (Figures 1, 2).

The results of the multivariate test showed significant differences between SNR values also STAT 1 and VEGF expression of each group (Table 2). After Post Hoc test, a significant difference between SNR values between groups ($p<0.05$) except between group 3 and group 4 was found ($p>0.05$). This result in accordance with STAT 1 also VEGF.

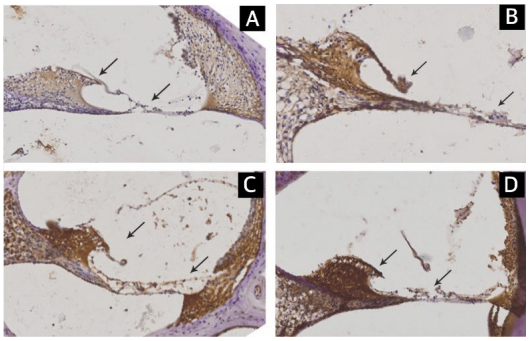

Figure 1. Signal transducer and activator of transcription 1 (STAT1) expression marked by brown color in organ of Corti (arrows) according to the duration of cisplatin exposure. A) Group 1 (control); B) group 2; C) group 3; D) group 4

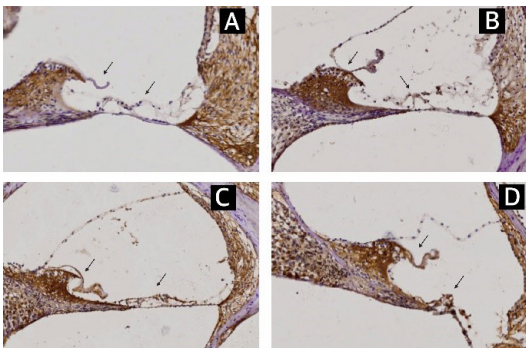

Figure 2. Vascular endothelial growth factor (VEGF) expression (stained brown cells) increased in treatment group 2 (B), 3 (C), and 4 (D). Group 1 (A) was a control group.

A negative correlation between SNR values and STAT1 and VEGF expression from bivariate test was found, as well as very a strong positive correlation between STAT 1 and VEGF with $p<0.05$ (Table 3).

The decrease in SNR value was inversely proportional to the increase in VEGF and STAT1 expression as well; the lower SNR value, the higher the VEGF and STAT1 expression.

Table 2. Multivariate test results of signal to noise ratio (SNR), and signal transducer and activator of transcription 1 (STAT1) and vascular endothelial growth factor (VEGF) among four groups

Variable	Group		p
SNR	1		0.000
	2		
	3		
	4		
STAT 1	1		0.000
	2		
	3		
	4		
VEGF	1		0.001
	2		
	3		
	4		
Post-Hoc Test			
SNR	1	2	0.000
		3	0.000
		4	0.000
	2	3	0.034
		4	0.019
	3	4	1.00
STAT 1	1	2	0.020
		3	0.003
		4	0.003
	2	3	0.025
		4	0.006
	3	4	0.179
VEGF	1	2	0.020
		3	0.003
		4	0.002
	2	3	0.014
		4	0.014
	3	4	0.742

Table 3. Spearman correlation between signal to noise ratio (SNR) values and vascular endothelial growth factor (VEGF) expression among four groups

Variable		Correlation r	p
SNR	STAT 1	-0.959	0.000
	VEGF	-0.980	0.000
STAT 1	VEGF	0.950	0.000

STAT 1, signal transducer and activator of transcription 1

DISCUSSION

High-frequency hearing loss due to cisplatin administration is reported to start 3-4 days after the cisplatin administration. Hearing loss manifests in various symptoms, such as severe and profound sensorineural hearing loss, depend on high doses of chemotherapy (15). Cakil et al. (15) found hearing loss at 15 dB or more in 75% of patients who received two doses of cisplatin at frequency of 4-8 kHz; hearing loss was found at frequency of 3-8 kHz on 10th and 17th days (15). Gündoğdu et al. (16) found that hearing loss at frequency of 2-8 kHz occurred three days after the cisplatin administration (16). In our study a significant decrease in SNR values was found in all study groups, from day 3 to day 7 and was lowest on the 7th day.

As in previous studies, ototoxicity due to cisplatin administration was found at the cellular level starting on the 3rd day after cisplatin administration and reached the maximum level on the 7th to 10th day (15). Outer hair cells damage in samples that were treated with cisplatin was proven by the decrease of SNR values in OAE examination. In Somdas et al. study (17), a significant SNR decrease was found on the 7th day after cisplatin administration (17).

Based on a study conducted by Perse et al. (18), the most frequently injected non-lethal dose of cisplatin in rats ranged between 1 mg/kgBW and 8 mg/kgBW. Cisplatin administration 3-8 mg/kgBW in rats will cause damage to kidney morphology in 3-4 days after injection (18). In this study, 8 mg/kgBW dose was used because there had been a decrease in hearing since the 3rd day after cisplatin administration with the stated dose, and all samples survived.

STAT1 is a protein that acts as a transcription factor and will increase as a response to inflammatory stimuli and oxidative stress (19). Inflammation is one of the responses that often results from ototoxicity (20). Mahmoud et al. (19) found increased expression of STAT 1 in the lungs of rats experiencing inflammation and oxidative stress (19). STAT1 is activated by cisplatin through the generation of ROS via transient receptor potential cation channels 1 (TRPV1) and NADPH Oxidase 3 (NOX3) pathway and widely distributed in the cochlea especially in the organ of Corti and spiral ganglion (21). Generation of ROS cause mitochondrial damage that leads to cell death by apoptotic reaction (22). In an animal model a study conducted by Jiang et al. (23), ototoxicity due to aminoglycosides caused a decrease in the number of outer hair cells and an increase in STAT 1 level. This was in line with ototoxicity caused by cisplatin (23). Inactivation of STAT1 in other tissues has also shown protective effects, such as increased autophagy in the heart or increased muscle regeneration (24).

The increase of STAT1 expression in all study groups with a higher immunoreactive score with longer exposure was found in our study. The highest

expression of STAT1 was seen in Group 4 where most cells stained brown on cochlear organ of Corti. We can ensure that there is an inflammation induced apoptotic mechanism on cochlear organ of Corti induced by cisplatin ototoxicity.

Cisplatin-induced nephrotoxicity increases VEGF expression as assessed with ELISA examination in a study conducted by Lin et al. (25). The role of VEGF in the cochlea is currently still unknown. VEGF expression is found in the normal cochlea, and its expression increases in response to hypoxia or oxidative stress in the cochlea. VEGF expression decreases in response to aging (26). VEGF expression increased in all the groups from our study, and the highest expression was found in Group 4. The increase of VEGF expression was in accordance with the cochlea organ of Corti damage. VEGF expression increased in noisy model rat cochlear cells on the 7-day group (10). Our study found that the highest increase in VEGF expression value was found in Group 4. Our study has proven that longer duration of Cisplatin exposure leads to more apoptotic hair cells in cochlea.

In this study, a negative correlation was found suggesting that there was a decrease in SNR value and increased STAT 1 also VEGF expression. This showed that there is an indication of hearing loss in experimental animals due to inflammation and apoptosis resulted from upregulation of STAT 1 and VEGF causing destruction to outer hair cells which manifested in decreased SNR values.

This study strengthens the evidence that STAT 1 and VEGF are important parameters in cochlear damage due to cisplatin administration, as evidenced by the correlation between increased STAT 1 and VEGF expression with decreased SNR values. A further study is needed to obtain therapeutic agents capable of inactivating STAT 1 and VEGF thus cochlear ototoxicity can be prevented or treated in the future.

In conclusion, cisplatin ototoxicity leads to damage to cell death in the organ of Corti in the cochlea through inflammation and apoptosis mechanism as evidenced by increased STAT 1 and VEGF expression also decreasing SNR values. This study found an association between STAT 1 and VEGF expression with SNR values.

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Conflict of interest: None to declare.

REFFERENCES

1. Paken J, Govender CD, Pillay M, Sewram V. A review of cisplatin-associated ototoxicity. Semin Hear 2019; 40:108-21
2. Rybak LP, Ramkumar V, Mukherjea D. Ototoxicity of non-aminoglycoside antibiotics. Front Neurol 2021; 12:652674.
3. Ganesan P, Schmiedge J, Manchaiah V, Swapna S, Dhandayutham S, Kothandaraman PP. Ototoxicity: a challenge in diagnosis and treatment. J Audiol Otol 2018; 22:59-65.
4. Ramkumar V, Mukherjea D, Dhukhwa A, Rybak LP. Oxidative stress and inflammation caused by cisplatin ototoxicity. Antioxidants 2021; 10:1919.
5. Rybak LP, Mukherjea D, Ramkumar V. Mechanisms of cisplatin-induced ototoxicity and prevention. Semin Hear 2019; 40:197-204
6. Rybak LP, Mukherjea D, Jajooo S, Ramkumar V. Cisplatin ototoxicity and protection: clinical and experimental studies. Tohoku J Exp Med 2009; 219:177-86.
7. Sheth S, Mukherjea D, Rybak LP, Ramkumar V. Mechanisms of cisplatin-induced ototoxicity and otoprotection. Front Cell Neurosci 2017; 11:338.
8. Zhang W, Xiong H, Pang J, Su Z, Lai L, Lin H, Jian B, He W, Yang H, Zheng Y. Nrf2 activation protects auditory hair cells from cisplatin-induced ototoxicity independent on mitochondrial ROS production. Toxicol Lett 2020; 331:1-10.
9. Deavall DG, Martin EA, Horner JM, Roberts R. Drug-induced oxidative stress and toxicity. J Toxicol 2012; 2012:645460.
10. Piciotti PM, Fetoni AR, Paludetti G, Wolf FI. Vascular endothelial growth factor (VEGF) expression in noise-induced hearing loss. J Heares 2006; 214:76-83.
11. Shetty S, Bhandary SK, Bhat V, Aroor R, Shety J, Dattatreya T. Role of otoacoustic emission in early detection of cisplatin induced ototoxicity. Indian J Otolaryngol Head Neck Surg 2022; 74:164-69.

12. Sanli A, Aydin S, Sarisoy ZA, Paksoy M, Ayduran E, **Çelebi** Erdivanli Ö. The protective effect of dexamethasone and lactate against cisplatin-induced ototoxicity. Turk J Med Sci 2011; 41:467-74.
13. Shomer NH, Allen-worthington KH, Hickman DL, Jonnalagadda M, Newsome JT, Slate AR, Valentine H, Williams AM, Wilkinson M. Review of rodent euthanasia methods. J Am Assoc Lab Anim Sci 2020; 59:242-53.
14. Czogalla B, Kahaly M, Mayr D, Schmoekel E, Niesler B, Kolben T, Burges A, Mahner S, Jeschke U, Trillsch F. Interaction of ERα and NRF2 impacts survival in ovarian cancer patients. Int J Mol Sci 2019; 20:112.
15. Cakil B, Basar FS, Atmaca S, Cengel SK, Tekat A, Tanyeri Y. The protective effect of *ginkgo biloba* extract against experimental cisplatin ototoxicity: animal research using distortion product otoacoustic emissions. J Laryngol Otol 2012; 126:1097-101.
16. Gündoğdu R, Erkan M, Aydin M, Sönmez MF, Vural A, Kökoğlu K, Karabulut D, Şahin MI. Assessment of the effectiveness of querctin on cisplatin-induced ototoxicity in rats. J int Adv Otol 2019; 15:229-36.
17. Somdaş MA, Güntürk I, Balcioğlu E, Avci D, Yazici C, **Özdamar** S. Protective effect of N-acetylcysteine against cisplatin ototoxicity in rats: a study with hearing tests and scanning electron microscopy. Braz J Otorhinolaryngol 2020; 86:30-7.
18. Perše M, Večerić-Haler Ž. Cisplatin-induced rodent model of kidney injury: characteristics and challenges. Biomed Res Int 2018; 2018:1462802.
19. Mahmoud AA, Abdel-Aziz HO, Elbadr M, Elbadre H. Effect of nicotine on stat 1 pathway and oxidative stress in rat lungs. Rep Biochem Mol Biol 2021; 10:429-34.
20. Bodmer D, Kern P, Bachinger D, Naldi AM, Huaman SL. STAT1 Deficiency predisposes to spontaneous otitis media. PLoS One 2020; 15:1-15.
21. Kros CJ, Steyger PS. Aminoglycoside and cisplatin-induced ototoxicity: mechanisms and otoprotective strategies. Cold Spring Harb Perspect Med 2019; 9:a033548.
22. Baghal-Sadriforoush S, Bagheri M, Abdi Rad I, Sotoodeh Nejadnematalahi F. PI3K inhibition sensitize the cisplatin-resistant human ovarian cancer cell OVCAR3 by induction of oxidative stress. Rep Biochem Mol Biol 2022; 10:675-85.
23. Jiang P, Ray A, Rybak LP, Brenner M. Role of STAT 1 and oxidative stress in gentamicin-induced hair cell death in organ of corti. Otol Neurotol 2016; 37:1449-56
24. Levano S, Bodmer D. Loss of STAT 1 protects hair cells from ototoxicity through modulation of STAT 3, c-Jun, Akt, and autophagy factors. Cell Death Dis 2015; 6:e2019.
25. Lin MT, Ko JL, Liu TC, Chao PT, Ou CC. Protective effect of D-Methionine on body weight loss, anorexia, and nephrotoxicity in cisplatin-induced chronic toxicity in rats. Integr Cancer Ther 2018; 17:813-24.
26. Clinkard D, Amoodi H, Kandasamy T, Grewal AS, Chen S, Qian W, Chen JM, Harrison RV, Lin VYW. Changes in the cochlear vasculature and vascular endothelial growth factor and its receptors in the aging c57 mouse cochlea. ISRN Otolaryngol 2013; 2013:4306.