Transylvanian Review

Vol 32, No. 3, 2024

Transylvanian Review
Centrul de Studii Transilvane| str. Mihail Kogalniceanu nr. 12-14, et.5, Cluj-Napoca

Email: transylvanianreview@gmail.com
Online Submission System: http://transylvanianreviewjournal.com/

Protective effects of pomegranate on mobile phone induced injury to spermatogenetic cells

Sibghatullah Sangi[1], Hadiya Sibghatullah[2], Shah Murad Mastoi[3], Sree Harsha Nagaraja [4]

[1]Faculty of Pharmacy, Northern Border University , Rafha, Saudi Arabia.
[2]Isra University, Hyderabad, Sindh, Pakistan.
[3]Department of Pharmacology, Quetta Institute of Medical Sciences, Quetta, Pakistan
[4]College of Clinical Pharmacy, King Faisal University, Al Ahsa, Saudi Arabia

ABSTRACT

This study examined the histopathological effects of EMR exposure on spermatogenesis and the antioxidant effects of Punica granatum (pomegranate). This study exposed 30 adults male Wistar rats to EMR (900 MHz) for one hour every day for eight weeks. A control group received standard filtered water, an EMR-only exposure group, and two treatment groups received Punicagranatum (0.4 ml/200g) and Punicagranatum (0.8 ml/200g). The last two groups received antioxidants during EMR exposure to see if they helped spermatogenic cells. After exposure, testicular tissues were stained with Hematoxylin and Eosin (H&E) to identify structural changes in the seminiferous tubules, including Sertoli cells, germinal epithelium, and spermatogenesis-supporting structures. There was significant difference in the sperm quantitative parameters sperm count 87 ± 13.36 x 106 and sperm motility 71 ± 15.97%, p=<0.05. A normal histological architecture with well-defined, closely packed seminiferous tubules, intact germinal epithelium, enough spermatogonia, spermatocytes, and supporting Sertoli cells indicated optimal spermatogenesis in the control group. In contrast, testicular sections from the EMR-only group showed severe structural disarray, including jumbled seminiferous tubules, Sertoli cell loss, and decreased spermatogonia and spermatocytes. When antioxidants were given, tissue preservation varied. The latter showed low Sertoli cell loss, maintained germinal epithelium, and reduced oxidative stress. Punica granatum's polyphenolic components, which scavenge free radicals, boost its efficiency. Finally, this study shows that spermatogenic cells are susceptible to EMR-induced oxidative damage and that antioxidant supplementation may mitigate such effects. Punica granatum protects testicular architecture and function, while the latter was more effective under oxidative stress. These findings suggest studying natural antioxidants to protect against EMR-induced reproductive abnormalities.

Keywords: Electromagnetic radiation, spermatogenesis, *Punicagranatum*, oxidative stress, infertility, testicular histopathology

Introduction

In the contemporary era, reproductive health faced numerous challenges, with a marked global rise in infertility rates attributed to both environmental and lifestyle factors. Traditional etiologies of infertility often overshadow the potential ramifications of modern lifestyle elements, including increased exposure to electromagnetic radiation (EMR) from mobile phones and Wi-Fi networks [1-3].

Infertility remains a global health issue, with increasing prevalence rates underscoring the need to explore non-traditional etiological factor s[4] [18]. Conventionally, infertility is attributed to genetic, physiological, or environmental causes, yet modern lifestyle choices—particularly the widespread use of mobile phones and Wi-Fi networks—introduce additional layers of risk that merit scrutiny[1, 3]. Electromagnetic radiation (EMR), specifically in the radiofrequency range emitted by mobile devices, has been implicated in a host of adverse biological outcomes, most notably oxidative stress and cellular damage within sensitive tissues [5]. Although the World Health Organization (WHO) maintains that mobile phone use has not been definitively linked to major health risks, studies suggest that prolonged exposure to EMR may induce subtle but significant physiological disturbances, particularly within the reproductive system [6]. EMR exposure from mobile devices and Wi-Fi networks can exacerbate oxidative stress, leading to cellular degeneration and potential carcinogenic effects, especially in tissues with high mitotic activity [5]. Similar findings have been reported in recent literature, emphasizing the damaging potential of EMR on brain and reproductive tissues due to reactive oxygen species (ROS)-induced oxidative imbalance [3, 7, 8].

Recent studies indicate that EMR exposure can disrupt spermatogenesis, the complex and delicate process by which spermatogenic cells in the testis develop into mature spermatozoa [9]. The spermatogenic cells are particularly vulnerable due to their high mitotic rate and reliance on oxidative balance, making them susceptible to oxidative damage—a process exacerbated by EMR exposure [10, 11]. Oxidative stress, defined as an imbalance between ROS production and antioxidant defenses, leads to cellular dysfunction, lipid peroxidation, and DNA damage, ultimately impairing fertility [12].

Studies by Sharma [13] and Dasdag et al. [7]demonstrate that EMR-induced ROS target testicular cells, disrupting the spermatogenic cycle and compromising reproductive function. Further, Zhao et al. emphasize that EMR exposure leads to cellular apoptosis, underscoring the need for intervention through antioxidants that counteract ROS effects [14, 15].In response to these findings, antioxidants have been proposed as protective interventions. Antioxidant, is widely studied for its neuroprotective and cytoprotective effects against oxidative damage [16]. By scavenging free radicals, antioxidants can mitigate the harmful effects of ROS on cellular structures, thereby preserving cellular integrity [17]. However, the antioxidant efficacy antioxidant varies, prompting exploration into other natural compounds with potentially superior antioxidative properties. Punica granatum (pomegranate) has emerged as a promising candidate due to its high content of polyphenols and flavonoids, which possess robust free radical scavenging and anti-inflammatory properties. This study examined the histopathological effects of EMR exposure on spermatogenesis and the antioxidant effects of Punica granatum (pomegranate). Its protective effects against EMR-induced cellular damage in reproductive tissues are thus a subject of significant research interest.

Materials and methods

Thirty adult male Wistar rats were selected for this study and exposed to EMR (900 MHz) for one hour daily over an eight-week period. The rats were divided into distinct groups: a control group receiving normal filtered water, an EMR-only exposure group, and two treatment groups supplemented with Punicagranatum. The latter two groups received antioxidants during the EMR exposure phase to assess any ameliorative effects on spermatogenic cells. Histopathological analysis was conducted on testicular tissues post-exposure, utilizing Hematoxylin and Eosin (H&E) staining to discern structural alterations within the seminiferous tubules, including any potential disruption to Sertoli cells, germinal epithelium, and spermatogenesis-supporting structures.

Results

The results of this study have shown promising results for Punica granatum. The weight of the study subjects among different groups ranging from 360.4 gm to 386.8 gm and the weight of the testis was ranging 0.47gm to 0.52 gm across the four groups. However, the difference among the groups was not statistically significant (Table 1). In addition, upon completion of the study there was a significant difference in the sperm count and motility in the EMR only group and Punica granatum group.

Table 1: Quantitate data across the groups

Parameters	Control (Normal Filtered Water)	EMR-Only Exposure (900 MHz, 1 hr/day)	EMR + Low-Dose Punicagranatum (0.4 ml/200g)	EMR + High-Dose Punicagranatum (0.8 ml/200g)	ANOVA p value
Body Wt in gm	375.0 ± 40.51	360.4 ± 45.93	377.0 ± 32.22	386.8 ± 59.81	0.311
Testis Wt in gm	0.52 ± 0.043	0.47 ± 0.073	0.52 ± 0.027	0.51 ± 0.026	0.245
% motile sperm	68 ± 8.50	45 ± 19.25	72 ± 18.78	71 ± 15.97	0.047*
Sperm count (x10^6 sperm/mL)	90 ± 12.83	54± 45.72	80 ± 20.39	87 ± 13.36	0.044*

P=<0.05 was considered statistically significant.

While assessing the histological aspect relevant to the outcome explored, it was observed that there was significant improvement in the groups with Punicagranatum supplementation.

The groups of description of results with the histopathological presentation of the preventive effect of Punicagranatum are described below.

Table 2: Overall outcomes and comparison of the effect among groups

Group	Testicular Architecture	Sertoli Cells	Germinal Epithelium	Spermatogonia & Spermatocytes	Oxidative Stress
Control (Normal Filtered Water	Normal	Abundant	Intact	Adequate numbers	Absent
EMR-Only Exposure (900 MHz, 1 hr/day)	Disorganized	Depleted	Disrupted	Reduced numbers	Present
EMR + Low-Dose Punicagranatum (0.4 ml/200g)	Partially preserved	Moderate reduction	Partially disrupted	Some reduction	Reduced
EMR + High-Dose Punicagranatum (0.8 ml/200g)	Substantially intact	Minimal loss	Preserved	Minimal reduction	Reduced

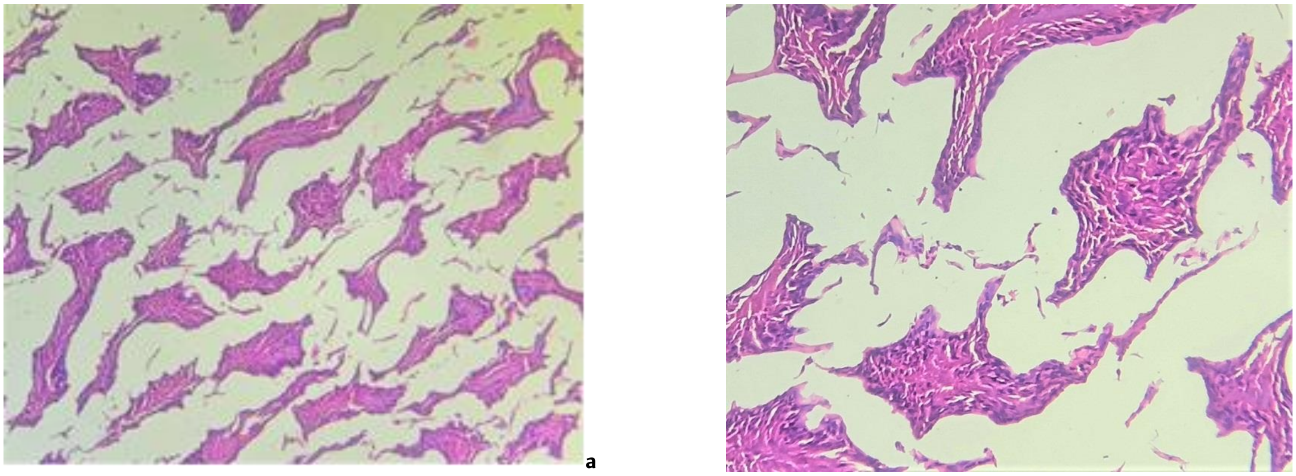
The testicular sections from the control group (Group 1) exhibited a well-preserved architecture characterized by closely packed seminiferous tubules with an intact germinal epithelium (Figure 1a). Supporting Sertoli cells appeared abundant and structurally intact, as indicated by their well-defined morphology (Figure 1b). The spermatogonia and spermatocytes were also present in adequate numbers, with visible spermatids and spermatozoa suggesting robust spermatogenic activity. Fibrovascular septae, consisting of fibroblasts, collagen fibers, and vascular spaces, maintained the structural integrity of the seminiferous tubules. Details are shown in Figure 1.

In Group 2, which was subjected to 900 MHz electromagnetic radiation for one hour daily, the testicular sections revealed significant architectural disruptions. There was a pronounced distortion in the germinal epithelium, with a notable loss of Sertoli cells (Figure 2a). This group showed a marked reduction in spermatogonia and spermatocytes (Figure 2b), accompanied by a visible depletion of spermatids and spermatozoa, suggesting impaired spermatogenic activity. The seminiferous tubules appeared separated by disorganized fibrovascular septae with irregular collagen fibers, indicating compromised structural support. Details are shown in Figure 2.

The testicular tissue in Group 3, exposed to EMR with concurrent low-dose Punicagranatum treatment, showed partial preservation of the seminiferous tubules. The architecture remained distorted, with some sections indicating loss of Sertoli cells and disruption in the germinal epithelium (Figure 3a). However, the spermatogonia, spermatocytes, and spermatids appeared more preserved compared to the EMR-only group, although a reduction in spermatozoa was still evident (Figure 3b) . The fibrovascular septae retained more structural organization, suggesting that

Punicagranatum provided a degree of protection against EMR-induced damage.

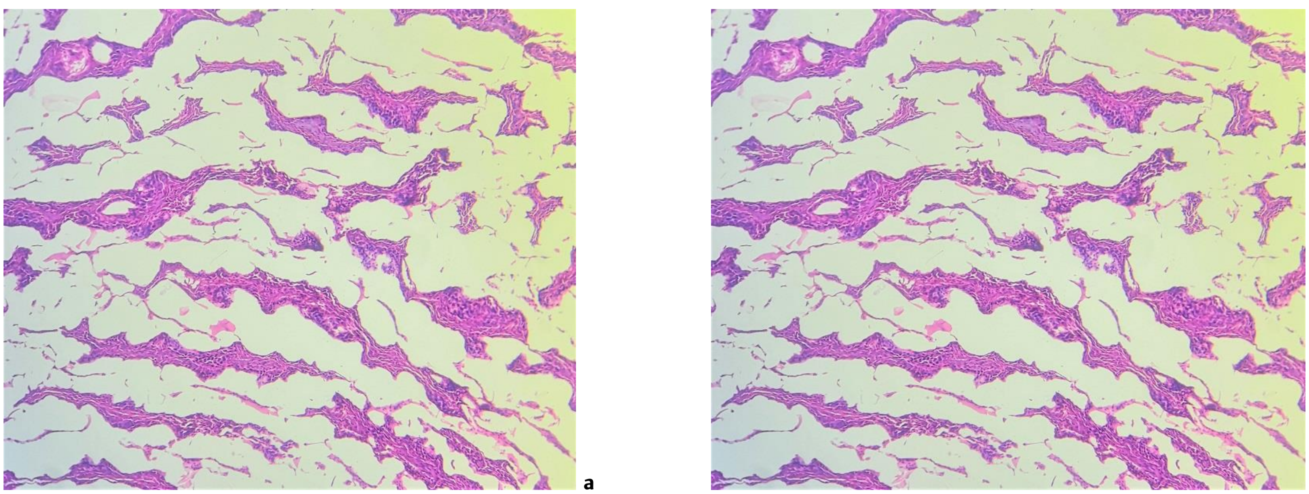
Details are shown in Figure 3.



a b

The section studied from the testis shows partially distorted architecture comprising of some closely packed seminiferous tubules with distortion of germinal epithelium and loss of supporting sertoli cells [Fig.1a]. The spermatogonia and spermatocytes appear adequate [Fig.1b]. The spermatids and spermatozoa appear reduced. The seminiferous tubules are separated by fibrovascular septae containing fibroblasts, collagen fibers and vascular spaces.

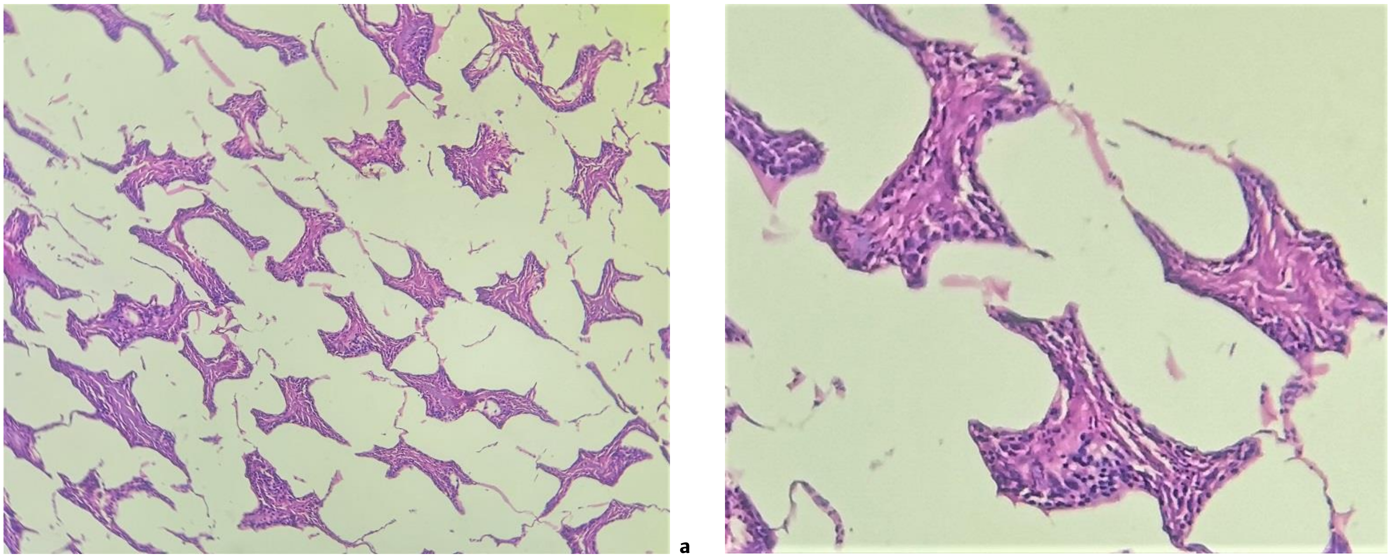
Figure 1: Group 1 Control (Normal Filtered Water)



a b

The section studied from the testis shows partially distorted architecture comprising of some closely packed seminiferous tubules with distortion of germinal epithelium and loss of supporting sertoli cells [Fig.2 a]. The spermatogonia and spermatocytes appear adequate [Fig.2 b]. The spermatids and spermatozoa appear reduced. The seminiferous tubules are separated by fibrovascular septae containing fibroblasts, collagen fibers and vascular spaces.

Figure 2: Group 2 EMR-Only Exposure (900 MHz, 1 hr/day)



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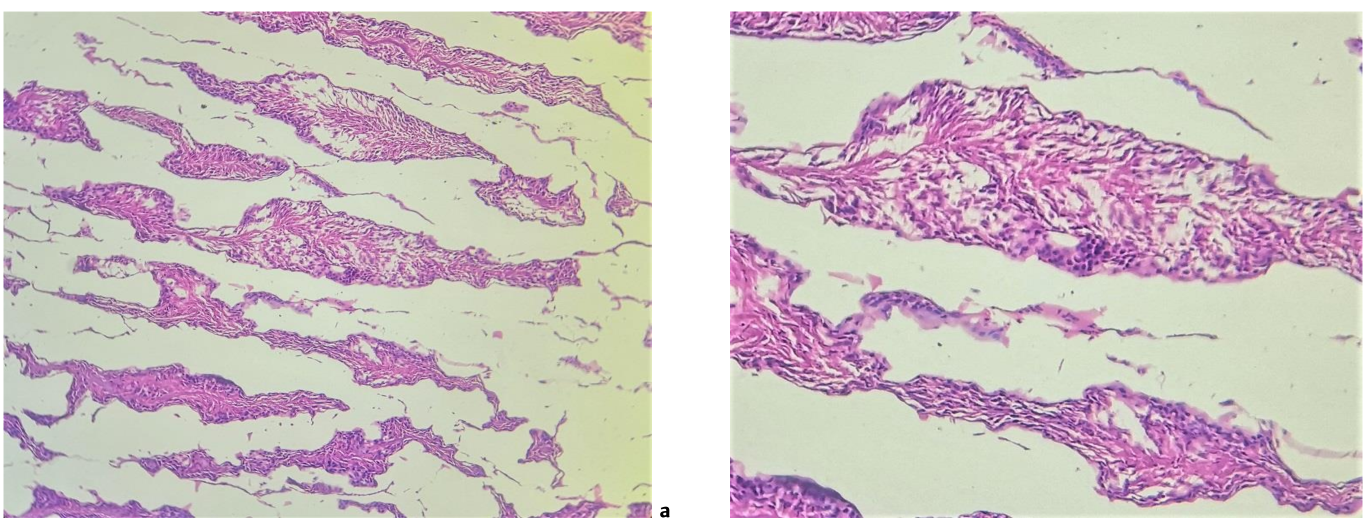
The section studied from the testis shows distorted architecture comprising of some closely packed seminiferous tubules with distortion of germinal epithelium and loss of supporting sertoli cells [Fig.3a]. The spermatogonia, spermatocytes and spermatids appear adequate [Fig.3b]. The spermatozoa appear reduced. The seminiferous tubules are separated by fibrovascular septae containing fibroblasts, collagen fibers and vascular spaces.

Figure 3: Group 3 EMR + Low-Dose Punicagranatum (0.4 ml/200g)

In Group 4, where rats were treated with a higher dose of Punicagranatum during EMR exposure, the testicular architecture showed remarkable preservation. The seminiferous tubules were closely packed, with minimal disruption in the germinal epithelium and abundant Sertoli cells (Figure 4a). Spermatogonia, spermatocytes, spermatids, and spermatozoa appeared intact and well-organized (Figure 4b), resembling the control group more closely. The fibrovascular septae were well-defined, with collagen fibers and vascular spaces maintaining a robust structural framework, indicating that Punicagranatum at a higher dose effectively mitigated EMR-induced testicular damage. Details are shown in Figure 3.



a b

Section studied from the testis shows intact architecture comprising of closely packed seminiferous tubules [Fig.4a] having intact germinal epithelium and supporting sertoli cells. The spermatogonia appear adequate in number and size. The spermatocytes, spermatids and spermatozoa appear adequate [Fig.4b]. The seminiferous tubules are bounded by fibrovascular septae containing fibroblasts, collagen fibers and vascular spaces.

Figure 4: Group 4 EMR + High-Dose Punicagranatum (0.8 ml/200g)

Discussion

The findings of this study align with an extensive body of literature indicating that EMR exposure induces oxidative stress and cellular damage, particularly in sensitive tissues such as those involved in spermatogenesis [9, 11, 17]. In the present study, the EMR-only group displayed significant histopathological alterations within the testicular architecture, including the distortion of seminiferous tubules, depletion of Sertoli cells, and reduced spermatogonia and spermatocytes. These structural disruptions mirror findings in prior studies, which reported similar morphological abnormalities in tissues exposed to EMR [4, 8]. These alterations are attributed to the oxidative stress resulting from EMR-induced ROS, which compromise cellular integrity and impede normal reproductive function[11, 18, 19].

The protective effects of *Punica granatum*, were pronounced than the EMR only group. *Punica granatum* antioxidative properties have been documented extensively, particularly in mitigating lipid peroxidation and preserving membrane stability[17]. However, in the context of this study, its protective efficacy was comparatively limited, as evidenced by the partial preservation of testicular architecture and reduction in oxidative markers, yet persistent structural disruptions were observed [7, 20]. This is consistent with findings from studies that noted that *Punica granatum*, while beneficial, may not fully counteract the oxidative effects of prolonged EMR exposure [10, 21].

In contrast, *Punica granatum* exhibited superior efficacy in preserving the structural integrity of the testicular microenvironment. Histopathological analysis revealed minimal disruption to the seminiferous tubules, intact Sertoli cells, and adequate spermatogonia and spermatocyte levels, suggesting robust antioxidative and anti-inflammatory effects. These results are supported by studies documenting the potent free radical scavenging properties of polyphenols and flavonoids in pomegranate [9]. The differential efficacy observed between P*unica granatum* aligns with emerging literature suggesting that polyphenolic compounds may offer enhanced protection against EMR-induced oxidative damage due to their ability to modulate intracellular signaling pathways and inhibit ROS production at a cellular level [3].

The results of this study are in agreement with the study conducted previously [22] The implications of these findings are substantial, given the ubiquity of EMR in daily life and the potential cumulative effects on reproductive health. This study reinforces the need for further investigation into the protective mechanisms of natural antioxidants and their application in mitigating EMR-induced reproductive damage, particularly in populations with high exposure levels [5].

Limitations

The results of this study are very basic now, which is mainly due to the lack of any funding for this project. Better methodological studies shall be planned, and the quantitative variables shall be explored in detail so that an extensive conclusion can be made regarding the supplementation of Punica Granatum.

Conclusion

The results of the study confirm that Cell phones as well as Wi-Fi directly damage the body cells including spermatogenic cells. The damage is proportional to the duration of exposure and strength of the radiation. Punica Granatum (Pomegranate) prevents and heals the damage caused by Cell phones and Wi-Fi, Punica Granatum comparatively works better. It's recommended to conduct further animal and human studies on the subject and some guidelines should be prepared to prevent male infertility

Study implications

The results of this study discuss a very important and neglected issue of our day-to-day lives. EMR are increasing day by day and it have negative impact on the male fertility. However, with appropriate supplementation of antioxidants these effects can be minimized. Clinical studies among human population shall be planned so that this effect can be investigated clinically, and appropriate clinical interventions can be identified.

Acknowledgement

The authors extend their appreciation to the Deanship of Scientific Research at Northern Border University, Arar, KSA for funding this research work through the project number "NBU-FFR-2024-1571-01."

Conflict of interest

There was no conflict of interest.

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