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Original article

The Effect of Different Gibberellic Acid (GA3) Doses on Seed Germination Properties of Some Soybean [*Glycine max* (L.) Merr.] Cultivars

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Abstract

In this study, the effect of different doses (0 (control), 50, 100, 200 ppm) gibberellic acid (GA3) treatment on the germination properties of Arısoy (C1), Crawford (C2) and Samsoy (C3) soybean cultivars were investigated. The experiments were factorial arranged in a completely randomized design with four replications. As a result of this study, final germination percentage (FGP), mean germination time (MGT), germination index (GI) value, germination energy (GE) value, seedling vigor index (SVI) value, seedling fresh weight (SFW), seedling dry weight (SDW) and seedling length (SL) were determined between 82-93%, 3-4.43 days, 27.8-37.3, 72-93%, 533-988, 0.57-0.71 g, 0.13-0.21 g, 6.5-10.6 cm, respectively. In terms of FGP, MGT, GI, GE, SVI, SL, SDW properties, a significant difference (P<0.01) was determined between cultivars, GA3 doses and cultivar x GA3 doses. Among the GA3 dose treatmens, FGP, GE, SVI, SL values with 100 ppm GA3 treatment and GI values with 150 ppm GA3 treatment reached the highest levels. MGT value was determined at the lowest 150 ppm GA3 treatment dose. Germination parameters were negatively affected with 200 ppm GA3 treatment dose. Responses of cultivars to GA3 doses were variable. In conclusion, seed priming with gibberellic acid (GA3) can be used to enhance germination properties of soybean cultivars.

Keywords: Soybean, Mean germination time, Seed vigor, Gibberellic acid (GA3), Seed priming.

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INTRODUCTION

Soybean [*Glycine max* (L.) Merr.] belongs to the legume family. Soybean, which has the highest cultivation area and production amount between legumes in the world, is rich in protein and oil proportion. Soybean is an industrial plant that can be used directly or as an additive in human and animal nutrition. Soybean plays an important role in providing the food supply and security of the increasing World population. Soybean has been produced in many countries such as the USA, Brazil, Argentina, and India for years.

The key component of cultivating plants sustainably is high-quality seed. Nowadays, different biotic and abiotic stress conditions negatively affect the germination and vigor of seeds. Therefore, crop yields also decrease significantly (Rhaman et al., 2020). Seed germination and viability depend on different conditions such as genetic structure (cultivar characteristic), sowing time, sowing depth, soil structure and content, irrigation, fertilization, plant diseases and pests, climate characteristics, moisture content of seeds at harvest time and storage. Priming (pre-treatment) is applied to reproduction materials such as seeds, seedlings, saplings, tubers, rhizomes, to improve the growth of plants under biotic and abiotic stress conditions. The history of this treatment dates back to the 4th century BC, and in the Ancient Greek period, Theophrastus soaked the seeds of cucumber in milk and water and observed that the seeds germinated earlier as a result of this practice (Evenari, 1984). It has been reported by various researchers that priming treatment increases seed germination, emergence, product yield, fertilizer use efficiency, water use efficiency and is an environmentally friendly treatment (Ghassemi-Golezani et al., 2012; Dalil, 2014; Bagheri, 2014; Dutta, 2018). Treatment of plant growth regulators to plant reproduction materials is one of the widely used priming methods. Gibberellins play an active role in many phases from the germination of the seed to plant improvement and they are widely used for priming in plant reproduction materials. It has been reported that gibberellic acid (GA3), one of the plant growth regulators, is both a natural chemical and has the ability to increase the properties of plants such as germination, vegetative growth and grain yield (Rahimi et al., 2011; Ma et al., 2018; Mangena, 2020). Seed germination is a complex process controlled by both physical and internal regulatory factors. Gibberellic acid plays a very important role in controlling and promoting of germination in plant species (Gupta and Chakrabarty, 2013). Gibberellins provide to break down of cell wall by increasing the endo-β-mannanase enzyme activity in the endosperm. Thus, germination ratio of seeds accelerates thanks to gibberellins (Karakurt et al., 2010).

The aim of the study was to determine the effects of four different gibberellic acid (GA3) doses, 0 (control) 100 ppm 150 ppm and 200 ppm, on the germination properties of Arısoy (C1) Crawford (C2) and Samsoy (C3) soybean cultivars.

MATERIALS and METHODS

The experiment materials were selected from cultivars prefered in the Black Sea Region Soybean Breeding Research Project conducted at the Samsun Black Sea Agricultural Research Institute. Arısoy (C1), Crawford (C2) and Samsoy (C3) soybean cultivars reach harvest maturity at a similar time. Thus, the seeds of these cultivars were preferred as trial materials. The research was carried out in the germination laboratory of Samsun Black Sea Agricultural Research Institute, in petri dishes with a diameter of 9 cm and under sterile conditions in 2021.

Experimental Design and Treatments

The experiments were factorial arranged in a completely randomized design with four replications. The first factor included four concentration of plant growth regulators i.e. GA3 i.e. (0, 100, 150, 200 ppm), the second factor included three soybean cultivars i.e. (Arısoy, Crawford, Samsoy).

For the sterilization of the seed samples in the study, the seeds were soaked in 10% sodium hypochlorite (NaClO) solution for 10 minutes, then washed 2-3 times with sterile water. Seed samples were subjected to pre-treatment (priming) by soaking them in gibberellic acid (GA3) solution at different doses (0 (control), 100 ppm, 150 ppm, 200 ppm). Seed weight to solution volume proportion was kept 1:5 (w/v) (Farooq et al., 2006). Priming was applied in a dark environment at 23±2°C for 12 hours. After the treatment, the seed samples were washed 2-3 times with sterile water and dried at room temperature (24 °C) until they had moisture content before the treatment. The primed seeds were sown as 25 pieces between filter papers (Whatman No. 2) in petri dishes with a diameter of 9 cm. After sowing, 10 ml of distilled water was added to each petri dish, and the outer surface of the petri dishes was covered with cling film to prevent evaporation. Prepared samples were germinated for 8 days at a temperature of 25± 1 °C in the dark. During the study, seeds with a radicle length of 2 mm and above and germinated were counted daily and recorded (ISTA, 2007).

Germination Properties

Final germination percentage (FGP)

It was calculated according to Equation (1) described by The International Seed Testing Association (ISTA) (2004).

$$FGP = \frac{\text{Number of germinated seeds after 8 days}}{\text{Number of germinated seeds}} x\ 100 \quad (1)$$

Mean germination time (MGT)

It was calculated according to Equation (2) described by Ellis and Roberts (1981).

$$MGT = \frac{\Sigma \text{Dn}}{\Sigma \text{n}} \quad (2)$$

where (n) is the number of seeds which were germinated on day (D), and (D) is the number of days counted from the beginning of germination.

Germination index (GI)

It was determined according to a notice reported by the Association of Official Seed Analysis (AOSA), (1983). It was calculated according to the Equation (3).

$$GI = \frac{No.of\ germinated\ seed}{Days\ of\ first\ count} + \cdots + \frac{No.of\ germinated\ seed}{Days\ of\ final\ count} \qquad (3)$$

Germination energy (GE)

It was the percentage of germinating seeds after 4 days from sowing relative to the number of tested seeds (Ruan et al. 2002).

Seedling vigor index (SVI)

It was calculated according to Equation (4) described by Abdul-Baki and Anderson. (1970).

$$SVI = \text{Seedling length (cm) x Final germination percent (\%)} \qquad (4)$$

In addition, at the end of the 8th day, the values of seedling length (SL), seedling fresh weight (SFW) and seedling dry weight (SDW) were determined in 10 seedlings randomly selected from each replication. Dry weight values were determined after the samples were dried in an oven at 70 °C for 72 hours.

Statistical Analysis

The statistical analysis of the research data was analyzed according to the factorial arrangements in the random plots in the SAS-JMP 16.0 package program, and results of variance analysis, the mean values of the treatments were compared with the LSD multiple comparison test at the $P < 0.05$ significance level.

RESULTS and DISCUSSION

As a result of the study, it was determined that 0 (control), 100 ppm, 150 ppm, and 200 ppm GA3 doses influenced the FGP, MGT, GI, GE, SVI, SL, SFW, SDW properties of C1, C2 and C3 soybean cultivars. Effects of GA3 doses on FGP and MGT properties and on GI and GE properties are illustrated in figure 1 and in figure 2 while effects of GA3 doses on SVI and SL properties and on SFW and SDW properties are illustrated in figure 3 and in figure 4, respectively.

The FGP values of soybean cultivars varied between 82% and 93%. The lowest FGP values were determined both at the 200 ppm GA3 dose treatment to the C1 cultivar and the 0 (control) dose applied to the C2 cultivar with 82% whereas the highest FGP values were detected both at 150 ppm GA3 dose applied to the C3 cultivar and 200 ppm GA3 dose applied to C2 cultivar with 93%. When the GA3 dose applied to C1 cultivar was increased from 150 ppm to 200 ppm, the FGP value decreased to 5.8%. The increase in GA3 dose applied to C2 cultivar triggered a boost the value of FGP. In the C3 cultivar, an increase till 150 ppm GA3 dose raised FGP value whereas 200 ppm GA3 dose caused a decrease in FGP

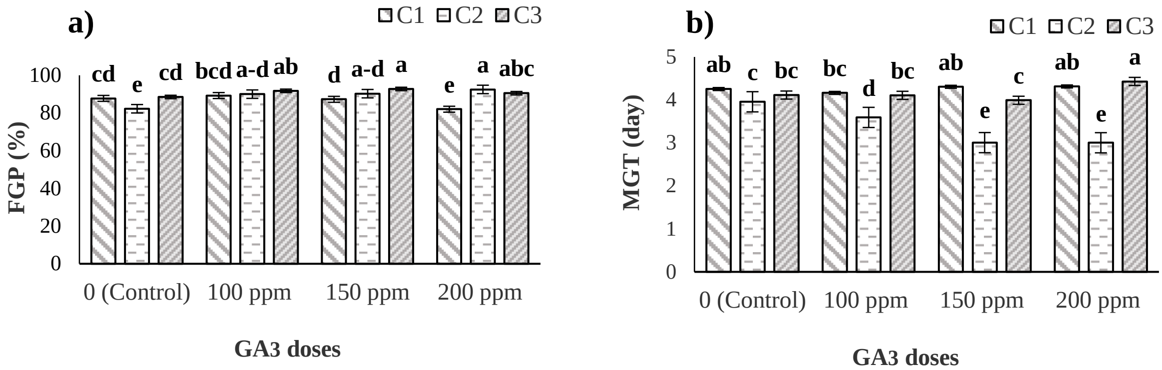

Fig. 1 Effects of different gibberellic acid (GA3) doses on **a** final emergence percentage (FGP) and **b** mean germination time (MGT). Bars represent the mean and ± standard error of the means of four replicates. Bars expressed with the same letters aren't significantly at *P*<0.05. Where **C1** Arısoy cultivar, **C2** Crawford cultivar, **C3** Samsoy cultivar.

value (Fig. 1a). It was determined that most appropriate doses of GA3 determined to shorten MGT were 100 ppm (4.16 days), 200 ppm (3 days), 150 ppm (3.99 days) doses in C1, C2, C3 cultivars, respectively. Applied 200 ppm GA3 dose caused an increase in MGT value of the C3 cultivar. The MGT value of the C2 cultivar was more decreased when compared to the MGT value of C1 and C3 cultivars at all applied GA3 doses (Fig. 1b). According to the results obtained, in soybean cultivars, seed priming with 100 ppm dose of gibberellic acid (GA3) increased FGP value while seed priming with 150 ppm and doses of gibberellic acid (GA3) shortened MGT value. Thus, the germination properties of C1, C2 and C3 soybean cultivars were improved compared to the control. Biró-Janka et al. (2019) reported that seed priming with gibberellic acid had a positive effect on the germination of basil varieties. In another study carried out, seed priming with 0.2 g/L GA3 in *Zea mays*, *Lathyrus sativus* and *Pisum sativum* rised the FGP value while shorten the MGT value compared to the control (unprimed) treatment (Tsegay and Andargie, 2018). In a similar way with our study, positive effects of seed priming with different gibberellic acid (GA3) doses were determined in sorghum (Azadi et al., 2013), sugar beet (Kandil et al., 2014), peanuts (Rouhi and Sepehri, 2020) and rapeseed (Zhu et al., 2021).

The GI value of the C1 cultivar is unaffected by the various GA3 treatment dosages. Depending on the increase in the applied GA3 dose, the GI value of the C2 cultivar increased. The GA3 dose increasing applied until 150 ppm in the C3 cultivar boosted the GI value but increasing of the applied

GA3 dose from 150 ppm to 200 ppm caused a decrease of 15.3% in the GI value. The highest GI value was determined in C3 cultivar at 0 (control), 100 ppm and 150 ppm doses of GA3 while that was

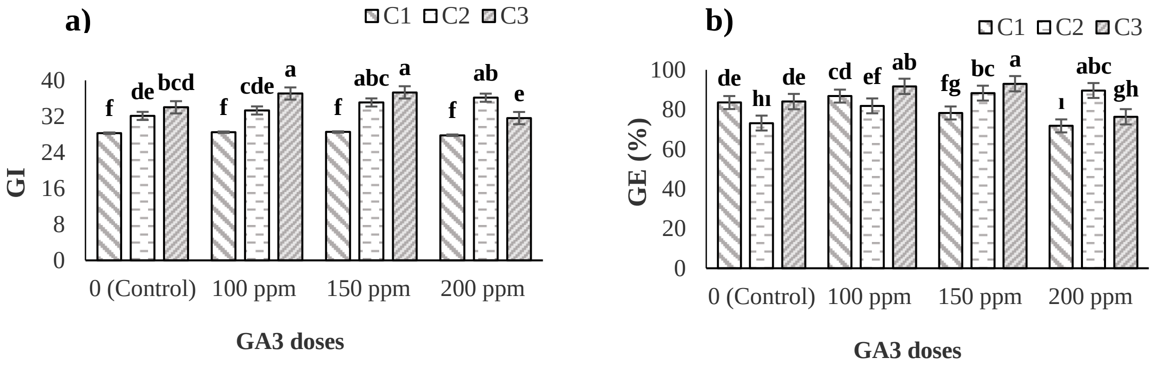

Fig. 2 Effects of different gibberellic acid (GA3) doses on **a** germination index (GI) and **b** germination energy (GE) using four replications. Bars represent the mean and ± standard error of the means of four replicates. Bars expressed with the same letters aren't significantly at $P<0.05$. Where **C1** Arısoy cultivar, **C2** Crawford cultivar, **C3** Samsoy cultivar.

determined in C2 cultivar at 200 ppm dose of GA3 (Fig. 2a). It was determined that The GE value of soybean cultivars vary between 72-93%. In C1 cultivar, GE value increased at 100 ppm GA3 treatment dose whereas GE value decreased at 150 ppm and 200 ppm GA3 treatment doses. As proportion of GA3 treatment dose increase, it was observed that GE value of C2 cultivar raised. GE value of C3 cultivar increased till 150 ppm GA3 treatment dose however that decreased at 200 ppm GA3 treatment dose. Treatment above 100 ppm GA3 dose level in C1 cultivar and 150 ppm GA3 dose level in C3 cultivar led to a decrease in GE value in both cultivars (Fig. 2b). Kumari et al. (2017) reported that 100 ppm GA3 treatment dose triggered to increase in GI and GE values of maize plant. In a study conducted in sunflower plant, it was detected that 150 mg/L GA3 treatment dose increased the GE value (Wahid et al., 2008). Researchers reported that 100 ppm GA3 dose in wheat (Mohaddes Ardebili et al., 2019), 300 mg/L GA3 dose in rapeseed (Zhu et al., 2021) and 50-100 ppm GA3 doses in soybean (Thongsri et al., 2021) increased GI values in three plant species. As a result of our examination, the most appropriate GA3 treatment dose was found 100 ppm for GE value, whereas the most appropriate GA3 treatment dose was found 150 ppm for GI value.

The highest SVI value between applied GA3 doses was determined at 150 ppm GA3 dose of C3 cultivar while the lowest SVI value was determined in 0 (control) treatment of C2 cultivar. In terms of SVI value of soybean, applied 150 ppm and 200 ppm GA3 treatment doses led to a decrease in C1 cultivar. In addition, applied 200 ppm GA3 treatment dose led to a decrease in C3 cultivar. According to SVI value of cultivars, it was determined that 100 ppm GA3 dose is the most ideal for C1 cultivar, 200 ppm GA3 dose is the most ideal dose for C2 cultivar and 150 ppm GA3 dose is the most ideal dose for C3 cultivar (Fig. 3a). Previous studies were demonstrated that 300 ppm GA3 doses in chickpeas (Mazed et al., 2015), 5 ppm GA3 dose in french beans (Barthwal and Prabha, 2018), and 100 mg/L GA3

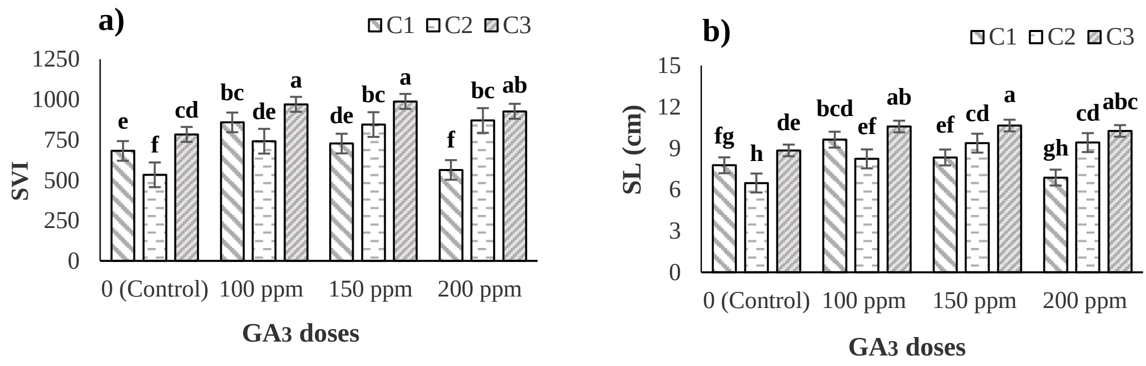

Fig. 3 Effects of different gibberellic acid (GA3) doses on **a** seedling vigor index (SVI) and **b** seedling length (SL) using four replications. Bars represent the mean and ± standard error of the means of four replicates. Bars expressed with the same letters aren't significantly at $P<0.05$. Where **C1** Arısoy cultivar, **C2** Crawford cultivar, **C3** Samsoy cultivar

dose in sorghum (Nimir et al., 2020) increased the SVI values of these plant species. When 100 ppm GA3 dose was applied, SL value reached the highest value with 9.6 cm in C1 cultivar but applied the GA3 dose more than 100 ppm caused a decrease in SL value of C1 cultivar. SL value increased significantly with 150 ppm GA3 treatment dose in C2 cultivar. In terms of SL value, there is a similarity in the effect of 200 ppm GA3 treatment dose and the effect of 150 ppm GA3 treatment dose in C2 cultivar. The highest SL value in the applied GA3 doses was determined in the C3 cultivar between examined cultivars. In C3 cultivar, the highest SL value was determined at 150 ppm GA3 dose with 10.6 cm, whereas the lowest SL value was determined in 0 (control) treatment with 8.8 cm (Fig. 3b). Seed priming with 100 ppm GA3 dose raised seedling length of cowpea plant (Arun et al., 2017). Seedling length of crop seeds primed was clearly greater than unprimed seeds (Nawaz et al., 2013). In C1, C2 and C3 soybean cultivars used in our study, priming with GA3 till a certain dose level increased the seedling length.

According to the applied GA3 treatment doses, the SFW values of C1, C2 and C3 cultivars varied between 0.57-0.71 g. The highest SFW value was determined at 200 ppm GA3 dose with 0.71 g in C3 cultivar, while the lowest SFW value was determined at 100 ppm GA3 dose with 0.57 g in C1 cultivar (Fig. 4a). The lowest SDW value was determined at 100 ppm GA3 treatment dose with 0.13 g whereas the highest SDW value was determined at 200 ppm GA3 treatment dose with 0.19 g in C1 cultivar. The increase of the ratio of applied GA3 doses led to a decrease in the SDW value of C2 cultivar. SDW value of C3 cultivar decreased when 100 ppm dose and 150 ppm GA3 doses apply compared to 0 (control) treatment. SDW value of C3 cultivar increased to 18.75% at 200 ppm GA3 treatment dose compared to 150 ppm GA3 treatment dose (Fig. 4b). Chauhan et al. (2019) reported that seed priming with GA3 increased the seedling fresh and dry weight values in three different oat cultivars, and the highest weight values were determined from 150 ppm GA3 treatment dose. Kumari et al. (2017) examined that the effects of halo-priming and hormonal-priming on seedling vigor and seed germination in maize.

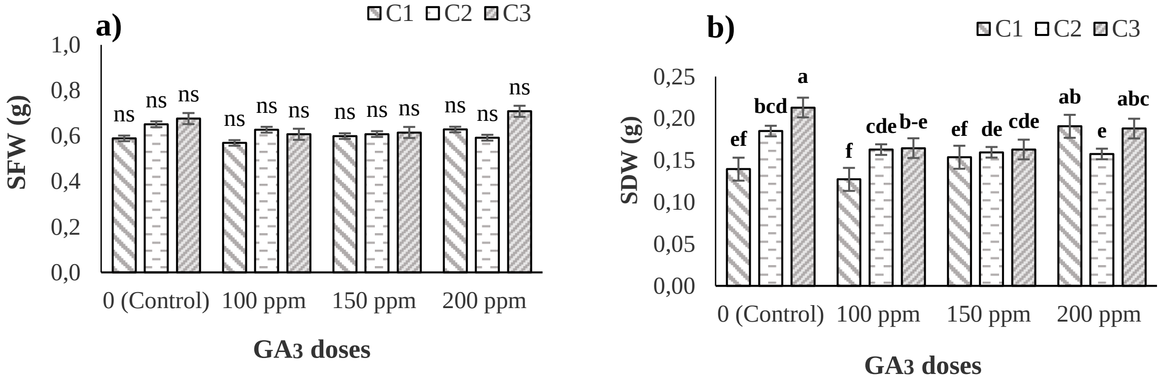

Fig. 4 Effects of different gibberellic acid (GA3) doses on **a** seedling fresh weight (SFW) and **b** seedling dry weight (SDW) using four replications Bars represent the mean and ± standard error of the means of four replicates. Bars expressed with the same letters aren't significantly at $P<0.05$. Where **C1** Arısoy cultivar, **C2** Crawford cultivar, **C3** Samsoy cultivar.

According to result of study, the highest SFW and SDW values were found at 100 ppm GA3 treatment dose. Seed priming with 0.2 g/L GA3 dose in maize, peas, and lathyrus (Tsegay and Andargie, 2018) and seed priming with 150 mg/L GA3 dose in rice (Banerjee and Roychoudhury, 2020) increased the SFW and SDW values of four plants species compared to the control (unprimed) treatments. The effect of seed priming with GA3 on the SFW of the C1, C2 and C3 cultivar was not observed in our study. This situation can be because the soybean plant is genetically different from other plant species.

As a result of the study, seed priming with 100 ppm GA3 increased the FGP, GE, SVI, SL values of C1, C2 and C3 soybean cultivars. According to statistical results, there were no statistically significant between the effect of GA3 treatment doses and the SFW value. According to mean value of cultivars, SDW value decreased at 100 and 150 ppm GA3 doses compared to 0 (control) dose. Moreover, the SDW value of 0 control dose and 200 ppm GA3 dose showed a similarity. Therefore, it was determined that there is no necessary to GA3 treatment for SFW and SDW properties in C1, C2 and C3 soybean cultivars. In terms of germination characteristics, the effect of GA3 doses showed different results between cultivars.

CONCLUSION

Seed germination is an important process that affects soybean production. The effects of hormonal priming can improve seedling development characteristic and field performance of soybean that is an important legume food. According to obtained results, it was clear that priming improves seed germination and seedling growth of soybean. Treatment of 100 ppm dose of gibberellic acid (GA3) in soybean can positively affect the germination of the seed. Treatment of 150 ppm dose of gibberellic acid (GA3) can shorten the mean germination time of seeds. However, this study is a laboratory study and more precise results should be obtained with greenhouse and field experiments.

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Additional Statement

Author contrubution rates: The authors contributed equally to the study.

The text confirming adherence to the ethical standards for research and publication: IJIAAR's research and publication ethics principles were followed throughout the article's process.

Conflict of interest declaration: There is no potential conflict of interest in this study.

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