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Assessment of Soft Wheat Genotypes *Triticum aestivum* L for Drought Tolerance at Germination and Early Seedling Growth Stages.

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A B S T R A C T

Drought has a harmful influence on the development and production of wheat crop. The response of soft wheat *Triticum aestivum* L plants to drought varies depending on the plant variety and growth stage. This study aimed to screen five genotypes of soft wheat to determine which genotypes are tolerant to drought stress at germination and seedling growth stages. Five genotypes of wheat plants were treated with two levels of drought in addition to the control. During the germination stage, wheat seeds were exposed to two levels 5 and 10% of Polyethylene Glycol 6000 solution. The study was planned according to a completely randomized design, with (45) experimental units, with (15) experimental units for each level of drought in addition to the control treatment, and with (3) replicates for each genotype. The results varied depending on the response of wheat genotype to drought. The findings revealed that there were very significant differences ($P<0.001$)) in both the seedling vigor index and the drought tolerance index traits, and a highly significant difference ($P<0.01$)) in the germination percentage trait under increasing severity of drought. In addition, the study revealed the presence of some drought-tolerant genotypes, and based on the drought tolerance index (%), the genotype (MEW22-1) is considered the most drought-tolerant at the germination stage. The study recommends further research and studies to assess the effect of drought on the growth and productivity of crops under field conditions and at different stages of plant life.

Introduction

Wheat is among the most significant crops in the world, providing more than (35%) of the world's food needs. It covers 3.42% of the world's food needs and constitutes a major food source for about 35% of the world's population, and covers 13% of the calories and protein in human nutrition, with a total production of about 778 million metric tons (Singh et al., 2023; Sharma and Sharma, 2025). The Mediterranean regions produce more than 50% of the world's wheat (Martínez-Moreno et al., 2022). Wheat is used in the global food industry and surpassed the list of crops in terms of cultivated area in 2018, with approximately 217 million hectares and a production of about 752 million tons (Erenstein et al., 2022). Wheat is one of the most important crops grown in Libya, cultivated under irrigated systems in some northern regions and non-irrigated systems in the eastern

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and mountainous regions. The cultivated area is estimated at 100,000–200,000 hectares (Shreidi et al., 2016). Like other crops, wheat is susceptible to various factors of damage and loss during its growth stages. Among the environmental stresses that have a significant impact on the growth and productivity of wheat plants is water stress, or drought.

Water stress (drought) refers to a deficiency in a plant's water requirements for a period of time adequate to cause injury. It can also be described as a decline in the quantity of available water in the soil, resulting in a deficiency in the plant's growth (Volaire, 2018). Drought is a widespread phenomenon that first affects plant establishment by decreasing water absorption and content, and then affects the seeds' germination, and increases the length of exposure to water stress, which causes the plant to suffer from numerous disturbances in various physiological functions, resulting in a complete termination of seedling growth (Da Silva et al., 2011; Yanagi, 2024). Drought is the main limiting factor in dry and semi-dry areas, as it is responsible for 50% of the weak production in the Mediterranean region. This phenomenon occurs during periods of low rainfall (UnNisa et al., 2022). Water stress affects all stages of growth from germination to seed production, and from the vegetative stage to the production stage (Ehtaiwesh, 2023). In general, the germination and seedling stage is less tolerant of environmental stresses compared to the adult plant (Khaeim et al., 2022). The germination stage is one of the most vital stages for agricultural crops. Often, wheat and barley grain crops are exposed to drought during the germination stage due to low rainfall during the planting season. Many studies have shown the importance of water absorption for seed germination, due to the role of water in metabolic processes during the germination stage. These studies also showed that the process of seed water absorption is affected by the availability of the water in the growth medium (Hassan and Ehtaiwesh, 2020) .Drought negatively affects many morphological, physiological, and biochemical characteristics, causing a decline in germination and seedlings growth (Khaeim et al., 2022). The results of some studies showed that drought causes a significant decrease in the rates of seed germination percentage, radicle length, dry weight, and seedling vigor index of barley plants (Ghosh et al., 2020). Some studies have also shown that drought has caused a decrease in germination rate, a decrease in seedling vigor index, as well as a decrease in the length of both the plumule and the rhizome and dry weight, and an increase in the proline content in wheat plants (Qadir, 2018).

Libya is located in dry and semi-dry areas; therefore, many crops face some ecological stresses such as water deficiency, warmness, salinity, etc. Because the response of plants to ecological stress varies according to plant genotypes, and according to plant diversities within the same species (Ehtaiwesh and Rashed, 2019), it has become necessary to identify varieties resistant to some environmental stresses such as drought. Thus, the purpose of the current research was to investigate the effect of drought on the gemination and seedling development of wheat crop, and to screen some varieties of soft wheat to identify drought-resistant varieties for inclusion in wheat breeding programs in Libya.

2. Materials and Methods

2.1 Plant material

The plant material used in this trial consisted of wheat *Triticum aestivum* L seeds obtained from the Misrata Agricultural Research Station. The varieties are: MEW22-1، MEW22-2، MEW22-3، MEW22-4 and Bohot 212).

2.2 Experimental and treatment conditions:

This laboratory trial was carried out in 2024 growing season, in the laboratory of the Botany Department, Faculty of Science, Gharyan University, using Petri dishes to evaluate the effect of three levels of drought control (0% concentration of Polyethylene Glycol 6000), moderate drought (5% concentration of Polyethylene Glycol 6000), and severe drought (10% concentration of Polyethylene Glycol 6000) on five varieties of soft wheat at the germination and seedling stages. The trial was planned based on a Completely Randomized Design (CRD) with three duplicates and two influences, where the first influence represented five varieties of soft wheat, and the second influence represented three levels of drought.

Initially, the seeds for all varieties under study were selected, washed thoroughly, and sterilized with a (5%) sodium hypochlorite solution (Ehtaiwesh, 2022). The seeds were cleaned thoroughly with distilled water several times, then the seeds were seeded in Petri dishes with filter paper, at a rate of (20) seeds per dish (Ehtaiwesh and Rashed, 2019). The dishes were separated into three sets, each set representing one of the drought treatments.

Water was added to the first group, which represents the control, and the seeds of the second group (moderate drought) were treated with a 5% concentration of Polyethylene Glycol (6000), while the seeds of the third group (severe drought) were treated with a 10% concentration of Polyethylene Glycol (6000) (Manoj, 2020).

After sowing, the plates were stored in a appropriate place at room temperature (25±2°C) and in a dark location. 24 hours after sowing, the number of germinated seeds (if any seed germinated) was counted daily, and the experiment continued for 10 days. During this period, the seedlings were monitored and water solution according to the required concentration were added as needed.

2-3 Data collection and calculations:

The germination percentage was calculated using the equation according to (Hassan and Ehtaiwesh, 2020).

$$\text{Germination (\%)} = \frac{Number\ of\ geminated\ seeds}{Total\ sown\ seeds} \times 100$$

The mean daily germination was calculated from the equation according to (Ehtaiwesh and Rashed, 2019.

$$mean\ daily\ germination = \frac{Number\ of\ geminated\ seeds}{Total\ number\ of\ days} \times 100$$

The germination speed was also calculated from the equation according to (Ehtaiwesh and Abuiflayjah, 2020).

$$Germiantion\ speed = \frac{n1}{d1} + \frac{n2}{d2} + \frac{n3}{d3} + \cdots$$

Where:
n = sum of seeds germinated on a given day.
d = the sum of days for seed germination.

The seedling vigor index was also calculated from the equation according to (Ehtaiwesh, 2019).

$$\text{seedling vigor index} = \frac{seedling\ length * \text{Germination \%}}{100}$$

The drought tolerance index was estimated following the formula (Ehtaiwesh, 2019)

$$\text{Drought tolerance index} = \frac{\text{Dry weight of seedling from drought treatment}}{\text{Dry weight of seedling from control treatment}} * 100$$

At the end of the research, on the tenth day, (3) plants were randomly chosen from each replicate for each treatment in order to record the length of the radicle (cm), the length of the plumule (cm), and the fresh weight (g) of the seedlings. Then, the plants were located in the air drier for drying at (60°C) for (48) hours, and then the dry weight (g) of the seedlings was measured (Ehtaiwesh, 2016).

2-4 Statistical Analysis of experimental data:

To statistically analyze the data, a two-way ANOVA was performed using SPSS 27 for germination and seedling growth traits with Duncan's Multiple Range test at a probability level of ($p \leq 0.05$).

3. Results:

The results in Table (1) show that drought had a highly significant outcome ($P<0.001$) on all studied germination traits, except for the germination percentage, where the consequence of drought was significant ($P<0.01$). The results also show that the plant genotype had a very high significant effect ($P<0.001$) on all studied traits except for fresh weight ($P<0.01$) and dry weight ($P<0.05$). In addition, the interaction between drought and plant genotype had a significant effect ($P < 0.05$) for all the traits involved in this research.

Table 1: The statistical probability (P value) of the effect of drought, plant genotypes and the interaction between them on some germination characteristics of five genotypes of soft wheat plants.

Traits	Drought (D)	Genotype (G)	(GxD)
Germination%	< 0.01	< 0.001	0.046
Mean daily germination	< 0.001	< 0.001	0.034
Germination speed	< 0.001	< 0.001	0.042
Seedling fresh weight (g)	< 0.001	< 0.01	0.046
Seedling dry weight (g)	< 0.001	< 0.05	0.004
Seedling length (cm)	< 0.001	< 0.001	0.032
Drought tolerance index	< 0.001	< 0.001	0.028
Seedling vigor index	< 0.001	< 0.001	0.034

Table (2) demonstrate the main effect of drought on the germination characters of five genotypes of soft wheat plant. The results indicate that drought led to a severe decrease in all the studied germination traits, and the decrease was highest under the severe drought treatment.

Table 2: The main effect of drought on some germination traits of five genotypes of soft wheat.

Traits	Concentration of Polyethylene Glycol		
	Control 0%	5%	10%
Germination%	96^{a*}	87.3^{b}	73c
Mean daily germination	4.6^{a}	3.5^{b}	2.2c
Germination speed	7.1^{a}	5.8^{b}	4.3c
Seedling fresh weight (g)	0.26^{a}	0.22^{b}	0.19c
Seedling dry weight (g)	0.056^{a}	0.047^{b}	0.036c
Seedling length (cm)	21.6^{a}	18.8^{b}	15.5c
Drought tolerance index	20.7^{a}	16.5^{b}	11.5c
Seedling vigor index	100^{a}	$^{b}82.9$	76.5c

*Values that share a common letter have no significant difference between them using multiple comparisons (Duncan)

Table (3) shows the main effect of the plant genotype on the germination traits. The results show that there is a difference in the response of the different wheat genotype to drought. In reference to the results, the genotype (MEW22-2) is more drought-tolerant based on the seedling length traits and the seedling vigor index traits, and based on the drought tolerance index, the genotype (MEW22-1) is the most tolerant among studies genotypes.

Table 3: The main effect of plant genotype on some germination traits of five genotypes of soft wheat.

Traits	MEW22-1	MEW22-2	MEW22-3	MEW22-4	Bo212
Germination%	^{de*}77.7	^{ab}90	^{a}91.6	^{c}88.8	^{d}78.8
Mean daily germination	2.9^{e}	3.52^{abc}	3.61^{a}	3.53^{ab}	3.02^{d}
Germination speed	^{de}5.2	^{b}6.1	^{a}6.2	^{abc}6.07	^{d}5.7
Seedling fresh weight (g)	0.21^{d}	0.24^{ab}	0.25^{a}	0.23^{abc}	0.19^{de}
Seedling dry weight (g)	0.045^{c}	0.048^{b}	0.055^{a}	0.043^{d}	0.040^{e}
Seedling length (cm)	15.6^{e}	21.08^{a}	19.6^{ab}	19.5^{abc}	17.4^{d}
Drought tolerance index	12.7^{e}	19.01^{a}	18.2^{b}	17.4^{c}	14.1^{d}
Seedlingvigor index	95.1^{a}	91.4^{abc}	64.1^{e}	92.7^{ab}	88.14^{d}

*Values that share a common letter have no significant difference between them using multiple comparisons (Duncan)

The results showed that drought has a significant effect on germination percentage, speed of germination and mean daily germination. however, different wheat genotypes have shown different responses to the effect of drought on these traits. Figure (1A) shows that different wheat genotypes exhibited varying responses to both moderate and severe drought, with the effect being most pronounced under extreme water stress. Figure (1A) also indicates that varieties MEW22-3 and MEW22-2 demonstrated the highest germination rates under severe drought compared to the other varieties. The results in Figure (1B) show that severe drought affected the germination speed, and the two varieties (MEW22-3) and (MEW22-2) recorded the highest value in germination speed in the severe drought treatment likened to the rest of the varieties comprised in the study. Also, the results in Figure (1C) show that the mean daily germination was clearly affected under the influence of severe drought, as the variety (MEW22-2) had the highest value in mean daily germination in the severe drought treatment compared to the rest of the studied varieties.

Figure (2) shows that drought affected some seedling characteristics, such as fresh weight, dry weight, and seedling length. Furthermore, the plant varieties exhibited differences under severe stress. Figure (2A) illustrates that drought had a significant impact on the fresh weight of different wheat varieties, with the MEW22-2 variety being the most drought-tolerant. Figure (2B) shows that drought had a negative effect on the dry weight of the different wheat varieties, with the MEW22-2 variety being the best in its drought tolerance, as it recorded the highest dry weight compared to the rest of the varieties under study. Figure (2C) shows the effect of drought on the seedling length of the different wheat varieties, as there was a difference in the tolerance of the varieties to drought, and the MEW22-2 variety recorded the highest value in terms of its tolerance to severe drought.

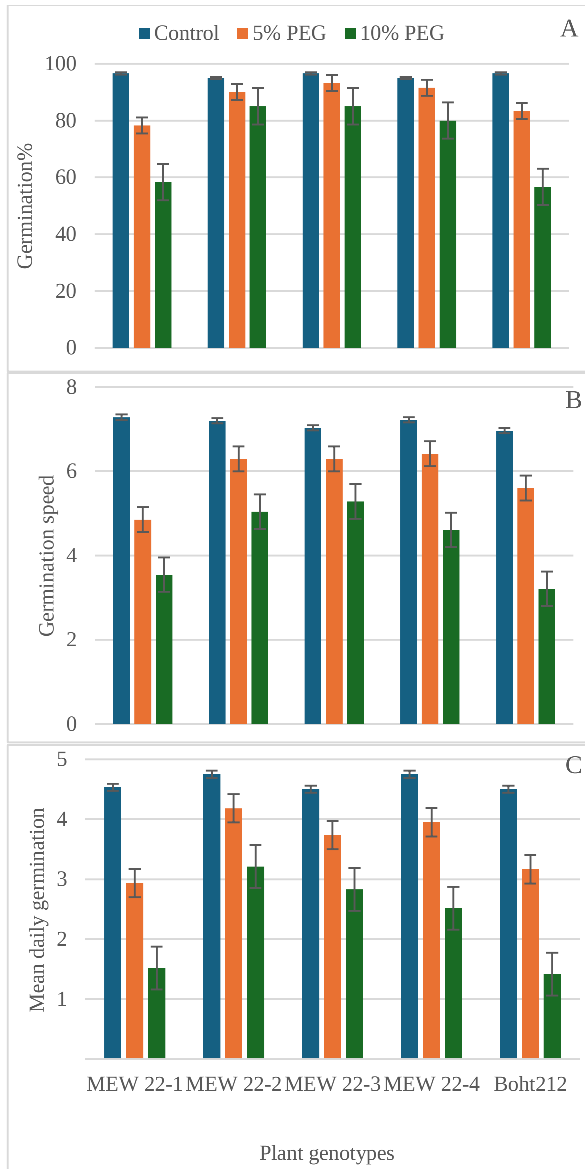

Figure 1: The effects of drought on (A) germination percentage, (B) germination speed and (C) mean daily germination of five soft wheat genotypes. Vertical lines on top of bars indicate standard error of means (n = 3).

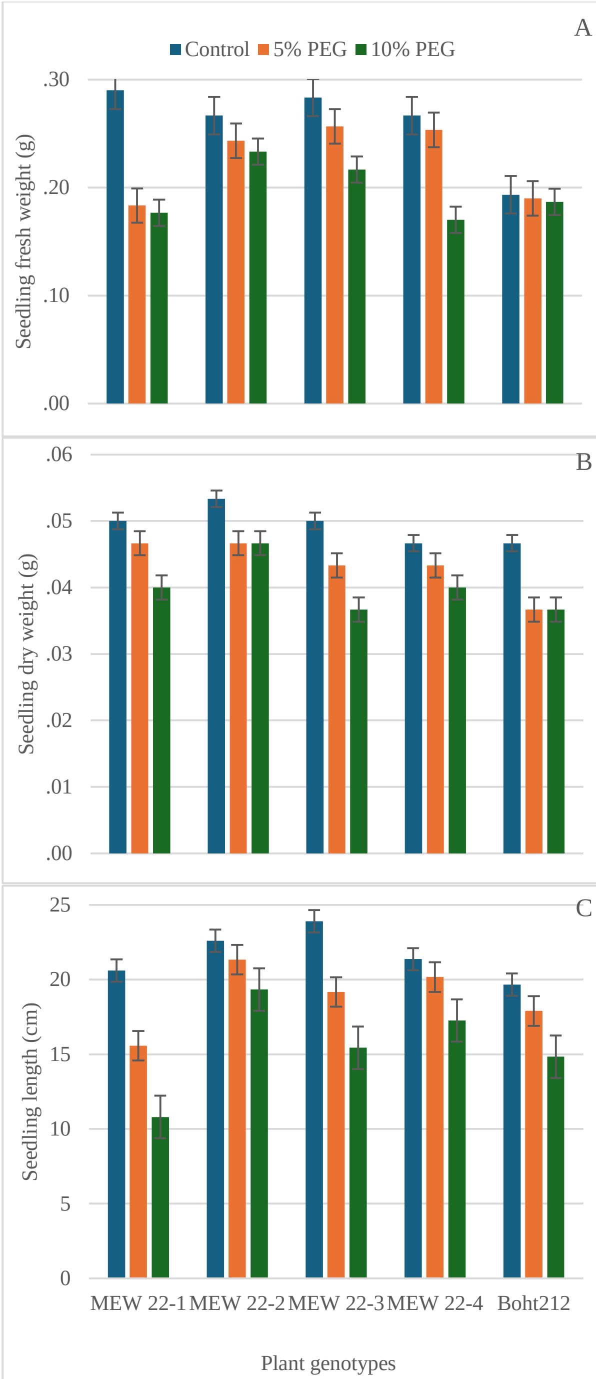

Figure 2: The effects of drought on (A) seedling fresh weight, (B) seedling dry weight and (C) seedling length of five soft wheat genotypes. Vertical lines on top of bars indicate standard error of means (n = 3).

Figure (3) clearly shows the effect of increasing water stress intensity on both the seedling vigor index and the drought tolerance index, as well as the different responses of the varieties to these drought tolerance traits. Figure (3A) shows that the seedling vigor index was best in the MEW22-2 variety. Figure (B3) shows that the drought tolerance index (%) was highest in the MEW22-2 variety under severe drought treatment compared to the other varieties.

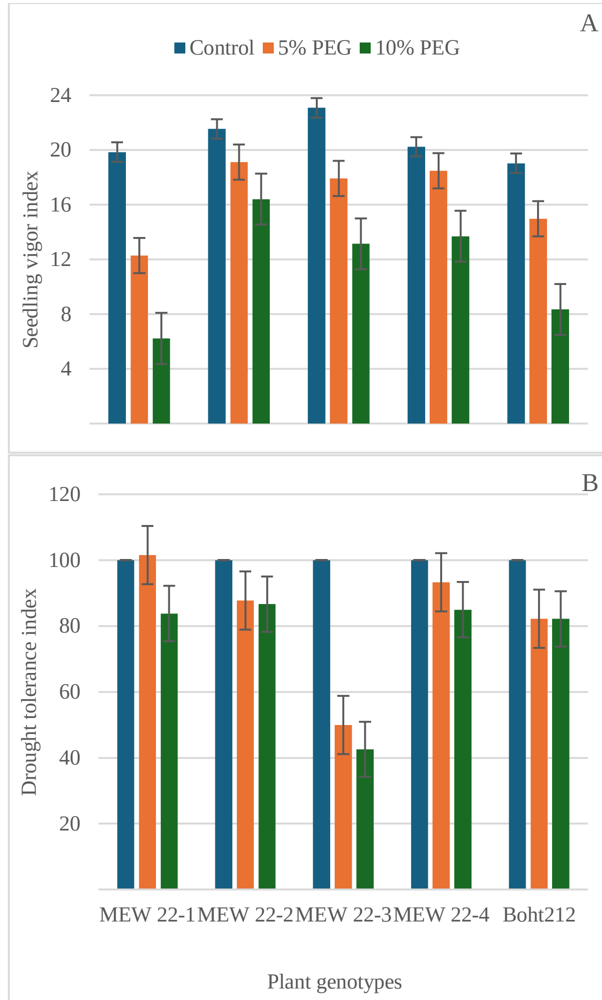

Figure 3: The effects of drought on (A) seedling vigor index and (B) drought tolerance index of five soft wheat genotypes. Vertical lines on top of bars indicate standard error of means (n = 3).

4. Discussion

The statistical outcomes presented in Tables 1 and 2 and Figures 1–3 showed that water stress made by polyethylene glycol (PEG-6000) had a highly significant effect ($P < 0.001–0.01$) on various wheat seed germination parameters. Germination percentage, mean daily germination rate, and germination speed all decreased, along with fresh and dry mass, seedling length, seedling vigor index, and drought tolerance index. This is linked to the reduction in the osmotic potential of the solution, which led to a reduction in water absorption during the imbibition stage, the stage most sensitive to any change in the moisture content of the surroundings. These outcomes are constant with what was recorded by Ghanifathi et al. (2011) and Qadir (2018), whose studies showed that water stress treatment using PEG leads to a clear decrease in germination rates, seedling length, and dry weight in several wheat varieties. It is likely that the reason is due to PEG's ability to mimic real water stress by drawing water from the seed and reducing the water potential in the medium, thus limiting basic physiological processes such as the activity of enzymes responsible for breaking down stored materials and releasing the energy needed to start growth. Increased drought intensity during the early growth stage of seedlings leads to slower engorgement and inhibition of cell division and expansion, resulting in a decrease in seedling rate and length. This is supported by the results of other studies such as Asaduzzaman et al., (2021) and Mahpara et al., (2022), which confirmed that water stress in the early stages of germination is a sensitive indicator used to assess seed vigor and the ability of genotypes to tolerate drought. The use of germination indicators is an effective tool in assessing the responses of genotypes to drought, as the results in Table (3) showed a clear difference between the tested varieties in the extent to which they were affected by the severity of the imposed drought, which indicates the existence of genetic variation in tolerance. Bowne et al., (2012) indicated that wheat varieties respond differently to water deficiency due to different physiological and molecular mechanisms associated with adaptation, such as the accumulation of soluble osmoles, increased antioxidant activity, and regulation of gene expression under stress. The findings of this research are also constant with what was reported in other research such as Bukhari et al., (2021) and Iyem et al., (2021), which showed that germination indicators under drought are a reliable means of identifying the varieties most able to continue growing in conditions of low water stress. Therefore, the differences recorded between varieties in the current experiment reflect the different levels of physiological efficiency and the ability to maintain water balance within the seed and seedling during the early stages of growth. Altogether, the findings of this investigation show that drought stress—even when applied only during the germination stage—has a substantial effect on the initial growth characteristics of wheat, making this stage a critical point in predicting the performance of varieties later under field drought. The results also indicate the importance of selecting varieties with the highest ability to maintain high germination rates and strong seedling indicators under drought conditions, which supports the role of breeding programs in exploiting this genetic variation to enhance wheat production in dry and semi-dry environ.

5. Conclusions:

This study concludes that drought has a significant effect on some germination characteristics such as germination percentage, mean daily germination, and germination speed. The results of this study also concluded that drought has a significant effect on seedling characteristics such as fresh and dry weight, seedling length, seedling vigor index, and drought tolerance index. The results also concluded that some varieties have shown an effective response in resisting different levels of drought. Hence, to obtain good germination and good seedling establishment of wheat plants under dry environmental conditions, planting drought-resistant varieties is the ideal solution. Understanding the impact of varieties on germination characteristics is crucial for improving agricultural productivity and ensuring good crop performance. However, further research and studies are still needed to delve deeper into this area.

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Conflict of interest: The authors declare that there are no conflicts of interest

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