

## Role of ginger extract in microbial inhibition and sensory enhancement of stored plant-based drinks: A study on soymilk and Zobo

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### Abstract

Plant-based beverages like soymilk and zobo are increasingly popular but highly susceptible to microbial spoilage, particularly in Nigeria, where cold chain infrastructure is inconsistent. This study investigated the dual effects of ginger extract on microbial inhibition and sensory enhancement of these beverages during 6 h ambient storage. Fresh soymilk and zobo samples were collected from local vendors and treated with 2% and 5% ginger extract concentrations. Microbiological analysis, pH determination, and sensory evaluation using a 9-point hedonic scale were conducted on fresh and stored samples. Initial microbial counts revealed high total viable counts ( $4.20 \times 10^5$  to  $5.20 \times 10^5$  CFU/ml) and total coliform counts ( $8.40 \times 10^5$  to  $1.04 \times 10^6$  CFU/ml), indicating contamination during production. While 2% ginger showed inconsistent antimicrobial effects, 5% ginger significantly reduced microbial loads, with zobo sample ZBF showing the lowest counts ( $1.50 \times 10^5$  CFU/ml TVC). Biochemical characterization identified pathogenic organisms including *Escherichia coli*, *Staphylococcus aureus*, and *Pseudomonas aeruginosa*. Sensory evaluation revealed that 5% ginger enhanced zobo acceptability but compromised soymilk palatability due to overpowering flavor. The study demonstrates that ginger extract at 5% concentration provides effective antimicrobial activity, particularly in acidic matrices, offering a practical natural preservation strategy for resource-constrained environments where consistent cold storage is of concern.

**Keywords:** Plant-Based Beverages; Ginger-Natural Preservatives; Food Safety; Antimicrobial Activity; Sensory Evaluation.

### 1. Introduction

Plant-based beverages such as soymilk and zobo have gained increasing popularity due to their nutritional quality, affordability, and potential health benefits. Soymilk, made from soybeans (*Glycine max*), is a non-dairy alternative rich in plant-based protein and free of lactose and cholesterol, is particularly suitable for vegans and individuals with lactose intolerance or cardiovascular concerns [1, 2]. Similarly, zobo, an infusion of *Hibiscus sabdariffa* is widely consumed across West Africa for its vibrant red color, tangy flavour, and high antioxidant content, including anthocyanins and polyphenols [3, 4]. However, despite these advantages, neither beverage is readily available because they are readily susceptible to microbial spoilage. This is due to their high moisture content and absence of naturally occurring preservatives. This vulnerability not only reduces shelf-life but also poses a serious public health threat, especially when sold by informal vendors under non-refrigerated conditions.

In Nigeria, street-vended foods and beverages are a staple of daily consumption, especially among urban and low-income populations. Yet, several studies have revealed alarmingly high microbial counts in these products, often surpassing World Health Organization (WHO) permissible limits [5]. For example, local soymilk and zobo samples obtained from vendors in Enugu State showed high total viable counts (TVCs), suggesting inadequate hygiene and

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inadequate preservative measures. Nigeria records approximately 173 million episodes of diarrheal diseases annually, many of which are foodborne in origin [6]. In this context, incorporating natural preservatives like ginger (*Zingiber officinale*), which exhibits broad-spectrum antimicrobial properties, may offer a feasible and affordable strategy for enhancing the microbial safety of these widely consumed drinks [7, 8, 2]. Which will, in turn, reduce the harmful health effects of synthetic preservatives.

Ginger is rich in bioactive compounds such as gingerols, shogaols, zingiberene, and paradols, which have been shown to disrupt microbial cell membranes and inhibit metabolic pathways [1, 8]. A study demonstrated that a 5% aqueous ginger extract significantly suppressed bacterial and fungal growth in soymilk stored at room temperature for three days, maintaining microbial loads within safe limits [2]. Similarly another research improved pH stability, lower microbial counts, and enhanced sensory attributes in ginger-treated zobo samples [3]. Moreover, ginger does not just serve as a preservative; it also enhances flavour, aroma, and mouthfeel, aligning well with consumer expectations for clean-label, functional beverages [4]. These dual functional properties make ginger an especially promising additive for low-resource contexts.

The use of natural preservatives like ginger is particularly compelling in Nigeria, where electricity supply is inconsistent, and refrigeration is often unavailable. According to FAO and Carrier reports, over 50% of global food spoilage in developing nations stems from the lack of reliable cold chains [9]. Voltage fluctuations and frequent power outages undermine the efficacy of refrigerators in both household and vendor settings [10]. Consequently, short-term preservation strategies that do not rely on electricity are urgently needed. Ginger extract, being low-cost, accessible, and culturally familiar, offers a practical intervention to improve shelf-life and safety, even without cold storage infrastructure.

Past studies have largely focused on either microbial inhibition or sensory characteristics in isolation, and often within a single beverage matrix. For example, a study showed that 0.2% ginger extract delayed microbial spoilage in zobo for up to 10 days under ambient conditions [7], while another focused on microbial control in soymilk [8]. A recent study found that 5% ginger improved flavour, aroma, and overall acceptability in zobo sweetened with dates and sugar [4]. However, there is limited research on the simultaneous effects of ginger on both microbial quality and sensory appeal across different plant-based drinks, particularly under short-term storage durations relevant to street vending scenarios.

Therefore, the present study, titled “Role of Ginger Extract in Microbial Inhibition and Sensory Enhancement of Stored Plant-Based Drinks: A Study on Soymilk and Zobo,” investigates how 2% and 5% concentrations of ginger extract influence microbial loads, pH stability, and sensory parameters after six hours of ambient storage. The dual objective is to assess both food safety and consumer acceptability in real-world, low-resource contexts. By integrating microbiological analyses with sensory evaluation, this work aims to provide actionable insights for beverage vendors, policymakers, and food scientists interested in natural preservation strategies. Ultimately, encouraging the use of ginger among street vendors could significantly mitigate public health risks while supporting nutritional security and local entrepreneurship.

## 2. Materials and methods

### 2.1. Sample Preparation

#### 2.1.1. Sample collection

Fresh samples of soymilk and zobo (*hibiscus*) beverages were purchased from two different local vendors within and around Godfrey Okoye University, Enugu. Each sample was collected in clean, food-grade polythene bags and immediately transported to the Biological Sciences Laboratory for analysis within one hour of purchase.

#### 2.1.2. Preparation of ginger extract

Ginger rhizomes were obtained from Abakpa Market, Enugu. The ginger was cleaned, peeled, dried and ground into a fine powder using a sanitized commercial grinder. The extract was prepared by weighing 2 g and 5 g of the ginger powder and into 100 mL each of the soymilk and zobo beverages samples, corresponding to 2% and 5% concentrations, respectively.

### *2.1.3. Application of Ginger Extract to Beverage Samples*

The collected beverage samples were divided into 4 portions each for different treatments: The first portions were the samples on arrival without ginger; Second portions were samples without ginger stored for 6 h; third portions were samples with 2% ginger stored for 6 hours; and finally, the fourth portions were samples with 5% ginger stored for 6 hours. Each treatment group was stored at room temperature ( $28 \pm 2^\circ\text{C}$ ). The ginger extract was thoroughly mixed into the beverages using a sterile syringe and sealed to ensure proper contact and homogeneity. Microbial, sensory and pH analyses were conducted both on arrival and after storage.

## **2.2. Determination of pH**

The pH of each beverage sample was determined using a digital pH meter (Hanna Instruments). The pH meter was calibrated using standard buffer solutions (pH 4.0 and 7.0) prior to use. A 10 mL aliquot of each sample was poured into a beaker, the glass electrode was immersed, and pH values were measured [11].

## **2.3. Microbiological Analysis**

### *2.3.1. Serial dilution*

Samples were serially diluted up to  $10^5$  using sterile normal saline. Aliquots (1 mL) of appropriate dilutions were used for microbial plating.

### *2.3.2. Media preparation and inoculation*

The culture media used for this analysis are MacConkey agar, Nutrient Agar, Mannitol Salt Agar, Eosin Methylene Agar and Salmonella Shigella Agar. Each medium was prepared according to the specifications of the manufacturer, sterilized by autoclaving at  $121^\circ\text{C}$  for 15 minutes, and poured into sterile Petri dishes. Plates were inoculated using spread plate and streak plate methods, then incubated at  $37^\circ\text{C}$  for 24 – 48 h [12].

### *2.3.3. Enumeration of microbial loads*

Microbial colonies were counted and recorded as colony-forming units per milliliter (CFU/mL). Plates were examined visually to assess microbial load [13].

## **2.4. Biochemical Characterization and Identification of Isolates**

### *2.4.1. Gram staining*

Gram staining was used for preliminary classification of isolates. Smears were prepared, heat-fixed, stained with crystal violet, treated with iodine, decolorized with alcohol, and counterstained with safranin. Microscopic examination classified bacteria as Gram-positive (purple) or pink/red for Gram-negative [11].

### *2.4.2. Catalase test*

A drop of 3% hydrogen peroxide was placed on a clean slide. A loopful of isolate was added and observed for bubble formation, indicating catalase-positive organisms [14].

### *2.4.3. Oxidase test*

Filter paper strips were moistened with oxidase reagent. A smear of bacterial isolate was added. A dark purple coloration within 30 s indicated a positive result [15].

### *2.4.4. Coagulase test*

Isolates were emulsified on a sterile slide with distilled water. A drop of plasma was added. Clotting within 10 s indicated a coagulase-positive strain [11].

### *2.4.5. Indole test*

Isolates were inoculated into peptone broth and incubated at  $37^\circ\text{C}$  for 48 h. After incubation, Kovac's reagent was added. A red ring at the surface indicated a positive result for indole production [15].

## 2.5. Sensory Evaluation

A 9-point hedonic scale was employed to evaluate sensory attributes of the beverage samples, including colour, creaminess, flavour, sweetness, smoothness, viscosity/thickness and overall acceptability. The scale ranged from 1 = Dislike Extremely to 9 = Like Extremely. A total of 15 semi-trained panelists (students and staff aged 20–40) participated. Panelists rinsed their mouths between samples. Evaluations were conducted on all the samples [16].

## 3. Results and discussion

### 3.1. Microbial count and pH

The study evaluated the impact of ginger addition on the microbial quality and pH stability of soymilk and zobo beverages over a 6 h period (Table 1). Initial microbial assessments of the samples on arrival (SAA, SBA, ZAA, ZBA) revealed relatively high Total Viable Counts (TVC) and Total Coliform Counts (TCFU), with soymilk samples ranging from  $4.50 \times 10^5$  to  $5.20 \times 10^5$  CFU/ml (TVC) and  $9.00 \times 10^5$  to  $1.04 \times 10^5$  CFU/ml (TCFU), while zobo samples showed slightly lower TVC but higher TCFU, indicating possible contamination during production or handling. These findings align with previous research with similar levels of microbial presence in freshly prepared soymilk and zobo drinks from Nigerian vendors, attributing it to poor hygienic conditions and water quality used in processing [17].

After 6 hours of ambient storage without any ginger treatment (SAO, SBO, ZAO, ZBO), microbial counts either remained high or increased slightly. This trend reflects the high perishability of these beverages and their nutrient-rich compositions that support microbial growth. For instance, the microbial count in SAO increased to  $8.30 \times 10^5$  CFU/ml, suggesting progressive microbial activity, possibly due to residual enzymatic actions or post-processing contamination. However, the inclusion of ginger at 2% concentration (SAT, SBT, ZAT, ZBT) did not show consistent antimicrobial effects.

**Table 1** Microbial count and pH of the zobo and soymilk beverage samples

Samples	Treatment	Microbial count (CFU/ml)	Total Coliform Count (CFU/ml)	pH
SAA	On arrival (0 h)	$4.50 \times 10^5 \pm 0.045$	$9.00 \times 10^5 \pm 0.033$	$6.50 \pm 0.025$
SBA		$5.20 \times 10^5 \pm 0.150$	$1.04 \times 10^5 \pm 0.001$	$6.21 \pm 0.003$
ZAA		$5.00 \times 10^5 \pm 0.005$	$1.00 \times 10^5 \pm 0.005$	$4.55 \pm 0.050$
ZBA		$4.20 \times 10^5 \pm 0.075$	$8.40 \times 10^5 \pm 0.025$	$4.38 \pm 0.055$
SAO	Stored after 6 h without Ginger (control)	$8.30 \times 10^5 \pm 0.033$	$1.66 \times 10^5 \pm 0.055$	$6.74 \pm 0.110$
SBO		$4.40 \times 10^5 \pm 0.025$	$8.80 \times 10^5 \pm 0.005$	$6.46 \pm 0.025$
ZAO		$3.10 \times 10^5 \pm 0.067$	$6.20 \times 10^5 \pm 0.020$	$4.70 \pm 0.075$
ZBO		$2.60 \times 10^5 \pm 0.087$	$5.20 \times 10^5 \pm 0.001$	$4.54 \pm 0.067$
SAT	Stored after 6 h with 2% Ginger	$9.60 \times 10^5 \pm 0.050$	$1.92 \times 10^5 \pm 0.075$	$6.32 \pm 0.075$
SBT		$8.40 \times 10^5 \pm 0.035$	$1.68 \times 10^5 \pm 0.033$	$6.15 \pm 0.025$
ZAT		$4.90 \times 10^5 \pm 0.022$	$9.80 \times 10^5 \pm 0.045$	$5.30 \pm 0.050$
ZBT		$5.00 \times 10^5 \pm 0.001$	$1.00 \times 10^5 \pm 0.020$	$5.83 \pm 0.020$
SAF	Stored after 6 h with 5% Ginger	$2.90 \times 10^5 \pm 0.020$	$5.80 \times 10^5 \pm 0.025$	$6.55 \pm 0.075$
SBF		$4.70 \times 10^5 \pm 0.110$	$9.40 \times 10^5 \pm 0.050$	$6.38 \pm 0.020$
ZAF		$2.80 \times 10^5 \pm 0.075$	$5.60 \times 10^5 \pm 0.100$	$5.55 \pm 0.045$
ZBF		$1.50 \times 10^5 \pm 0.050$	$3.00 \times 10^5 \pm 0.083$	$6.07 \pm 0.055$

In some cases, microbial loads remained high or even increased, particularly in the soymilk samples. For example, SAT recorded a TVC of  $9.60 \times 10^5$  CFU/ml, higher than the initial and control values. This suggests that 2% ginger may have provided insufficient antimicrobial activity or contributed additional nutrients that promoted microbial growth in a

nutrient-rich medium like soymilk. Similarly, lower concentrations of ginger extract were observed that failed to suppress microbial activity in soymilk unless combined with other spices such as clove or subjected to mild heat treatment [18].

Where SAA and SBA represent soymilk samples from two different vendors without ginger on arrival, SAO and SBO represent soymilk samples from two different vendors without ginger stored after 6 hours; SAT and SBT represent soymilk samples from two different vendors with 2% ginger stored after 6 hours; SAF and SBF represent soymilk samples from two different vendors with 5% ginger stored after 6 hour; ZAA and ZBA represent zobo samples from two different vendors without ginger on arrival, ZAO and ZBO represent zobo samples from two different vendors without ginger stored after 6 hours; ZAT and ZBT represent zobo samples from two different vendors with 2% ginger stored after 6 hours; ZAF and ZBF represent zobo samples from two different vendors with 5% ginger stored after 6 hours.

Conversely, the application of 5% ginger (SAF, SBF, ZAF, ZBF) resulted in a marked reduction in microbial counts across both beverage types. ZBF, in particular, showed a dramatic decrease in microbial load to  $1.50 \times 10^5$  CFU/ml (TVC) and  $3.00 \times 10^5$  CFU/ml (TCFU), the lowest among all samples. This confirms the potent antimicrobial activity of ginger at higher concentrations, especially in an acidic matrix like zobo. These results are consistent with a demonstration that higher doses of ginger extract significantly inhibited common foodborne pathogens, including *E. coli*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus* [19]. The findings further agreed with another study that observed that spice-treated zobo drinks stored under ambient conditions showed significantly delayed spoilage when 5% or higher concentrations of natural preservatives were used [20].

The pH values of the samples also varied slightly across treatments. Soymilk maintained a relatively neutral to slightly acidic pH (6.15–6.74), while zobo samples, being naturally acidic, had pH values ranging from 4.38 (ZBA) to 6.07 (ZBF). The observed increase in pH among ginger-treated zobo samples suggests that ginger may buffer the acidity or reduce the metabolic acid production of surviving microbes. This finding is consistent with research that showed a gradual pH increase in spice-treated beverages during storage, attributing it to microbial inhibition and reduced organic acid accumulation [21]. The slight rise in pH may also be beneficial in extending the sensory acceptability of zobo, which is often limited by excessive sourness over time.

### 3.2. Microbiological Characterization

Table 2 presents the microbiological characterization, revealing the presence of multiple foodborne organisms, including *Bacillus cereus*, *Pseudomonas aeruginosa*, *Klebsiella* spp., *Enterobacter* spp., *Escherichia coli*, and *Staphylococcus aureus*. These organisms are frequently implicated in spoilage and foodborne illnesses, especially in unpasteurized beverages. The detection of *S. aureus* and *E. coli*, both pathogenic and indicators of poor handling hygiene, reinforces the need for improved food safety practices. Ginger has bioactive compounds like gingerols and shogaols, which can disrupt bacterial membranes and inhibit ATP production, making it effective against both Gram-positive and Gram-negative bacteria [22].

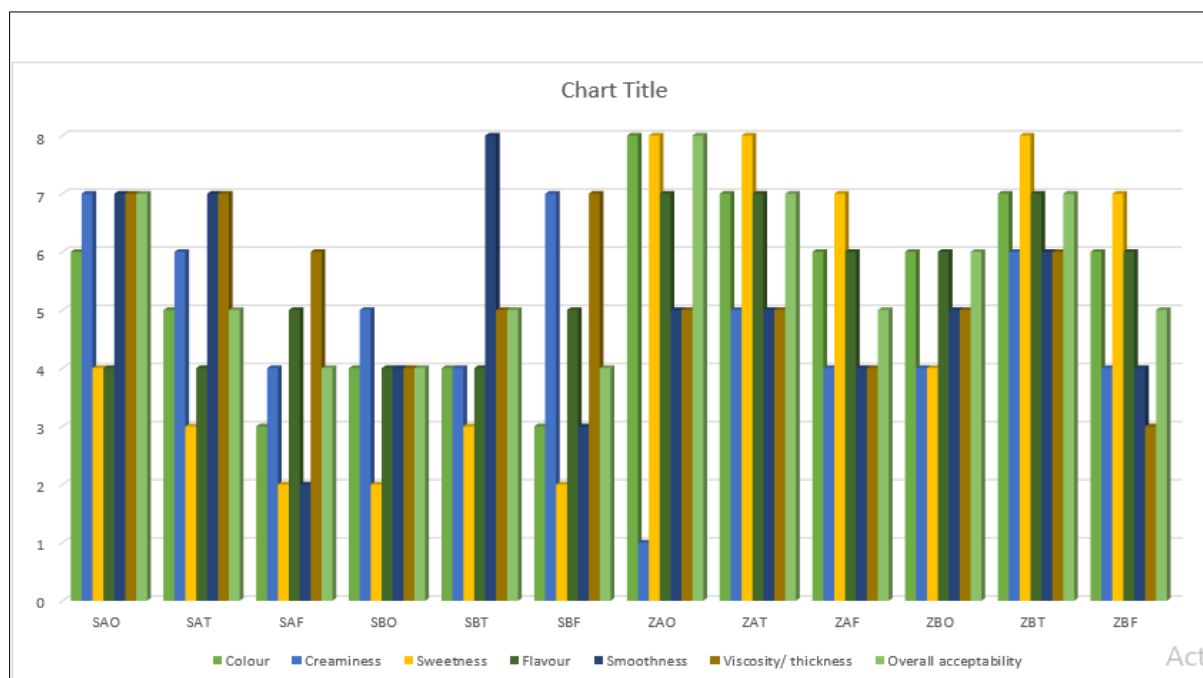
**Table 2** Morphological and Biochemical identification of the microorganisms isolated from the soymilk and zobo beverage Sample

TEST	Bac 1	Bac 2	Bac 3	Bac 4	Bac 5	Bac 6	Bac 7
Gram staining	+	-	-	+	-	-	+
Morphology	Rod	Rod	Rod	Cocci	Rod	Rod	Rod
Catalase	+	+	+	+	+	+	+
Coagulase	-	-	-	+	-	-	-
Oxidase	+	-	+	-	-	-	-
Indole	-	-	-	-	-	+	-
Probable organisms	<i>Bacillus cereus</i>	<i>Enterobacter</i> spp	<i>Pseudomonas Aeruginosa</i>	<i>Staphylococcus Aureus</i>	<i>Klebsiella</i> spp	<i>Escherichia coli</i>	<i>Bacillus</i> spp

### 3.3. Sensory Evaluation

The sensory evaluation of the soymilk and zobo beverage samples assessed key attributes including colour, creaminess, flavour, sweetness, smoothness, viscosity/thickness and overall acceptability as presented in Figure 1. Results indicated that the addition of ginger influenced the sensory properties of both beverages, with variations observed depending on the concentration of ginger and the type of beverage matrix.

For soymilk, panelists generally preferred the samples without ginger or with only 2% ginger addition. The fresh samples on arrival (SAA and SBA) scored highest in taste and mouthfeel, suggesting consumer preference for the mild, creamy flavour profile of plain soymilk. However, samples with 5% ginger (SAF, SBF) were rated lower in taste and overall acceptability. This could be attributed to the strong, pungent flavor of ginger, which tends to overpower the natural flavour of soymilk when used in higher concentrations. These findings are in line with a previous report that observed that while ginger enhances the functional properties of plant-based beverages, it can negatively impact sensory attributes when added beyond consumer tolerance levels [4].



**Figure 1** Sensory evaluation of the beverage samples

Where SAA and SBA represent soymilk samples from two different vendors without ginger on arrival, SAO and SBO represent soymilk samples from two different vendors without ginger stored after 6 hours; SAT and SBT represent soymilk samples from two different vendors with 2% ginger stored after 6 hours; SAF and SBF represent soymilk samples from two different vendors with 5% ginger stored after 6 hours; ZAA and ZBA represent zobo samples from two different vendors without ginger on arrival, ZAO and ZBO represent zobo samples from two different vendors without ginger stored after 6 hours; ZAT and ZBT represent zobo samples from two different vendors with 2% ginger stored after 6 hours; ZAF and ZBF represent zobo samples from two different vendors with 5% ginger stored after 6 hours.

In contrast, the zobo samples demonstrated improved sensory scores with increasing ginger concentration, particularly in aroma and overall acceptability. Panelists preferred the 5% ginger-treated samples (ZAF and ZBF) due to their perceived richness, spiciness, and refreshing flavor. The zobo matrix, which is naturally acidic and flavoured, appeared to complement its pungency, resulting in a harmonious blend. This is supported by another finding that reported ginger addition improved the sensory appeal and consumer acceptability of zobo drinks by enhancing both aroma and flavour complexity [23].

The aroma of both beverages improved with ginger addition, likely due to the volatile essential oils in ginger, such as zingiberene and citral, which contribute to a distinctive and pleasant fragrance. However, mouthfeel was negatively affected in some ginger-treated soymilk samples, possibly due to the fibrous nature of ginger particles or their interaction with soymilk proteins. Colour was not significantly affected in either beverage type, although a slightly darker hue was noted in ginger-enriched zobo samples, which was still acceptable to most panelists.

Overall, sensory evaluation suggests that while moderate levels of ginger (2%) may be acceptable in both beverages, higher concentrations (5%) enhance sensory attributes in zobo but may compromise acceptability in soymilk. These observations highlight the importance of optimizing spice concentration based on the specific beverage matrix and consumer preferences. A similar conclusion emphasized that balancing nutritional enhancement with sensory quality is critical in developing functional beverages using bioactive-rich ingredients like ginger [24].

#### 4. Conclusion

In summary, the study demonstrates that while ginger exhibits antimicrobial properties, its efficacy is concentration-dependent and influenced by the beverage matrix. The acidic environment of zobo enhances its activity, while the richer composition of soymilk may buffer its effectiveness. Overall, sensory evaluation suggests that while moderate levels of ginger (2%) may be acceptable in both beverages, higher concentrations (5%) enhance sensory attributes in zobo but may compromise acceptability in soymilk. These results support existing literature advocating for the combined use of natural antimicrobials and mild preservation techniques to enhance the safety and shelf life of traditional beverages.

#### Compliance with ethical standards

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##### *Disclosure of conflict of interest*

The authors declare no conflict of interest.

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