Live yeast supplementation during late gestation and lactation affects reproductive performance, colostrum and milk composition, blood biochemical and immunological parameters of sows

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A B S T R A C T

This study was conducted to evaluate the effects of dietary live yeast (LY) supplementation during late gestation and lactation on reproductive performance, colostrum and milk composition, blood biochemical and immunological parameters of sows. A total of 40 multiparous sows were randomly fed either the control (CON) diet or the CON diet supplemented with LY at 1 g/kg from d 90 of gestation to weaning. Results showed that the number of stillborn piglets and low BW piglets were significantly decreased in the LY-supplemented sows compared with sows in the CON group (P < 0.05). Moreover, the concentrations of protein, lactose and solids-not-fat were increased in the colostrum of LY-supplemented sows (P < 0.05). Interestingly, the plasma activities of aspartate aminotransferase (AST), alkaline phosphatase (ALP), gamma-glutamyl transpeptidase (γ-GGT) at d 1 of lactation and alanine aminotransferase (ALT) at weaning day were decreased by feeding LY diet (P < 0.05). Meanwhile, sows fed LY diet had higher plasma concentration of immunoglobulin G compared with sows fed CON diet at d 1 of lactation (P < 0.05). In conclusion, LY supplementation in maternal diets decreased the number of stillborn piglets and low BW piglets, improved colostrum quality and health status of sows.

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1. Introduction

Under the intensive production conditions, the sow is subjected to many stressors (repeat services, changes of housing during gestation and lactation, BW loss, etc.) (Kranendonk et al., 2007; Spoolder et al., 2009), which reduce their immune function and increase disease susceptibility (Sutherland et al., 2006). Besides, nutrient absorption by sows during gestation and lactation greatly influences litter characteristics at birth and at weaning (Kranendonk et al., 2007). Proper nutritional strategy to improve health status of sows and their reproduction performance would be of benefit to overall pig production, and probiotics have gained extensive attention with their beneficial effects by decreasing health problems and improving livestock performance (Alexopoulos et al., 2004; Chaucheyras-Durand and Durand, 2009).

Live yeast (LY) has been widely used as a live microbial feed supplement in livestock animals (Jang et al., 2013; Jiang et al., 2015; Wang et al., 2016). Previous studies have shown that LY can improve growth performance, immunological status, and intestinal structure of pigs (Jiang et al., 2015; Lemieux et al., 2010; Monroy-Salazar et al., 2012), and modulate intestinal microbial balance and attenuate intestinal inflammation in chickens (Haldar et al., 2011). In addition, trials have indicated that dietary supplementation of yeast products during gestation and lactation enhances litter weight gain and litter weight of weaned piglets (Kim et al., 2008; Shen et al., 2011), reduces weaning to estrus interval (Jang et al., 2013), and improves milk composition (Jurgens et al., 1997).
However, some researchers reported yeast supplementation did not improve reproductive performance (Jurgens et al., 1997) and milk composition of sows (Jang et al., 2013). Thus, the effects of yeast supplementation on sow performance and health status still need to be determined.

Therefore, the aim of this study was to investigate the effects of LY supplementation on reproductive performance, milk composition, blood biochemical and immunological parameters of sows during late gestation and lactation.

2. Materials and methods

The experimental protocol of this study was in accordance with the animal care and use committee of Sichuan Agricultural University (Sichuan, China), and followed the current laws of animal protection (Ethic Approval Code: SCAUAC201408-3).

2.1. Animals, diets and management

This study was conducted at Giastar Pig Experimental base, Chengdu, Sichuan, China. On d 90 of gestation, 40 crossbred sows (Landrace × Yorkshire, average parity 4.2) were selected and randomly assigned to 2 groups (20 sows/group). The dietary treatments included a basal diet (control, CON) and the diet supplemented with LY at 1 g/kg (treatments included a basal diet (control, CON) and the diet supplemented with LY at 1 g/kg (Saccharomyces cerevisiae, strain CNCM I-4407, 10¹⁰ CFU/g, Actisaf, Phileo Lesaffre Animal Care, Marcq-en-Belgium) (treatments included a basal diet (control, CON) and the diet supplemented with LY at 1 g/kg (Saccharomyces cerevisiae, strain CNCM I-4407, 10¹⁰ CFU/g, Actisaf, Phileo Lesaffre Animal Care, Marcq-en-Belgium, strain CNCM I-4407, 10¹⁰ CFU/g, Actisaf, Phileo Lesaffre Animal Care, Marcq-en-Belgium). The sow basal diet (Table 1) was formulated to meet or exceed the nutrient requirements for both gestating and lactating sows (NRC, 2012). Live yeast diet was formulated by substituting corn by LY. Live yeast diet was applied from d 90 of gestation to weaning (d 21 of lactation). Gestation diets were fed twice daily (07:00 and 17:30) at 3 kg/d from d 90 of gestation until farrowing day. From d 90 to 107 of gestation, the sows were housed in the same room with an individual pregnancy crate (2.0 m × 0.6 m). On d 108 of gestation, the sows were transferred to farrowing cages (2.0 m × 2.5 m) and individually fed. After farrowing, all sows were switched to lactation diets, which were supplied 4 times a day (07:00, 11:00, 14:00 and 18:00), starting at 2.0 kg/d and increased by 0.5 kg/d during the first 5 d, afterwards the sows had free access to diet until weaning. Cross-fostering was performed within 24 h post-farrowing and litters of piglets were adjusted to 12 to 13 piglets within the same treatment. The average daily feed intake (ADF) of the sows during lactation was recorded. Sows had ad libitum access to water during the experiment. The average temperature was maintained at 20 to 22 °C in the gestating room, and 24 to 26 °C in the farrowing room. Supplemental heat was provided to piglets with heat lamps (250 W). In this study, no milk replacer or creep feed was provided to the piglets.

2.2. Data and samples collection

After parturition, the total number of pigs born, born alive, mummified or stillborn, as well as the number of low BW piglets (BW < 1.0 kg) for each sow were recorded. The weight of the piglets was recorded individually at parturition and weaning. The backfat thickness of sows was measured at 65 mm to the left side of the dorsal midline at the last rib at d 90 of gestation, and farrowing and weaning days using an ultrasonic device (Renco Lean-Meatier; Renco Corporation, Minneapolis, MN, USA).

At d 1 and 21 of lactation, blood samples of sows (n = 10) were collected from ear veins. Approximately 10 mL of blood per sow was collected into heparinized tubes, followed by centrifugation at 3,000 × g and 4 °C for 15 min. Plasma was obtained from the supernatant and stored at −80 °C for later analysis.

The colostrum samples were collected within 1 h after onset of farrowing and milk was collected at d 21 of lactation after the injection of 1.5-mL oxytocin (Hangzhou Animal Medicine Factory, Hangzhou, China) via the ear vein of sows (n = 10). The samples were immediately stored at −20 °C until the further analysis.

2.3. Determination of milk composition

By referring our previous study (Wan et al., 2016), colostrum and milk samples were analyzed for the concentrations of protein, fat, lactose, and solids-not-fat by means of an automatic milk analyzer (Milk-Yway-CP2; Beijing, China). The results are expressed as the percentage in milk.

2.4. Determination of plasma metabolites and the related enzyme activities

The concentrations of glucose, triacylglycerol, and urea in plasma were determined using specific assay kits (Nanjing Jiancheng Bioengineering Institute, Nanjing, China) according to the manufacturer's instructions. Plasma activities of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) and gamma-glutamyl transpeptidase (γ-GGT) and total bilirubin (TBL) concentration were measured by corresponding commercial kits (Sichuan Maker Biotechnology Inc., Chengdu, China), using automatic biochemical analyzer (Hitachi 7020, Hitachi High-Technologies Corporation, Tokyo, Japan). The minimal detection limit was 0.02 mmol/L for glucose, 0.02 mmol/L for triacylglycerol, 0.10 mmol/L for urea, 4.00 U/L for ALT, 3.00 U/L for AST, 2.00 U/L for ALP, 0.70 μmol/L for TBL, and 2.00 U/L for γ-GGT. The intra-assay and inter-assay CV were <5% for each assay.

2.5. Determination of plasma immunological parameters

Plasma concentrations of immunoglobulin G (IgG), immunoglobulin M (IgM), complement 3 (C3), complement 4 (C4) and C-reactive protein (C- RP) were detected by corresponding commercial kits (Sichuan Maker Biotechnology Inc., Chengdu, China), using

Table 1 Composition and nutrient contents of basal diet (% as-fed basis).

| Item                    | Gestation | Lactation |
|-------------------------|-----------|-----------|
| Ingredients             |           |           |
| Corn                    | 59.23     | 62.21     |
| Soybean meal            | 12.89     | 23.29     |
| Wheat bran              | 22.18     | 5.00      |
| Fish meal               |           | 2.50      |
| Soybean oil             | 2.00      | 3.00      |
| i-Lysine HCl            | 0.14      | 0.28      |
| i-Threonine             | 0.05      |           |
| α-Methionine            |           | 0.14      |
| Calcium carbonate       | 0.96      | 0.98      |
| Dicalcium phosphate     | 1.45      | 1.50      |
| Salt                    | 0.40      | 0.40      |
| Chloride choline        | 0.15      | 0.15      |
| Vitamin and mineral premix† | 0.55 | 0.55 |
| Total                   | 100       | 100       |
| Calculated values       |           |           |
| Digestible energy, Mcal/kg| 3.11  | 3.35      |
| CP                      | 13.66     | 17.77     |
| Ca                      | 0.76      | 0.90      |
| Available P             | 0.40      | 0.47      |
| Total lysine            | 0.71      | 1.11      |

† The premix supplied the following vitamins and trace minerals per kilogram of diet: Cu, 20 mg; I, 0.3 mg; Mn, 40 mg; Se, 0.3 mg; Fe, 165 mg; Zn, 165 mg; Cr, 0.3 mg; 25,000 IU vitamin A; 5,000 IU vitamin D3; 12.5 IU vitamin E; 2.5 mg vitamin K; 1 mg vitamin B1; 8 mg vitamin B2; 3 mg vitamin B6; 0.015 mg vitamin B12; 17.5 mg niacin; 12.5 mg pantothenic acid; 0.25 mg folacin.
an automatic biochemical analyzer (Hitachi 7020, Hitachi High-
Technologies Corporation, Tokyo, Japan).

2.6. Statistical analysis

Data were analyzed by an independent-samples t-test using
SPSS 21.0 software (SPSS Inc., Chicago, IL, USA). Sows and their
litters were considered as the experimental units in the model,
which included diet (CON or LY) as the main effect. For the analysis
of reproductive performance, sows and their litters were used as
the experimental units. For the analysis of milk composition,
plasma immunological parameters, plasma metabolites and the
related enzyme activities, sows were used as the experimental unit.
Results were expressed as means ± standard error. Differences
were considered as significant when \( P < 0.05 \), whereas
\( 0.05 \leq P < 0.10 \) was considered a tendency.

3. Results

3.1. Reproductive performance

The results showed that feeding LY diet did not significantly
alter backfat thickness, total number of piglets born, born alive,
individual birth weight or litter birth weight, which were all similar
to the CON group (Table 2). However, sows fed the LY diet decreased
the number of stillborn piglets and low BW piglets (\( P < 0.05 \))
compared with sows fed the CON diet. The number of pigs weaned,
average daily gain for piglet, pre-weaning mortality, weaned litter
size, piglets/litter and individual weight were not significantly influenced
by dietary treatment. Besides, no significant treatment effect was
observed for sows’ ADFI during lactation.

3.2. Colostrum and milk composition

As shown in Table 3, the concentrations of protein, lactose and
solids-not-fat in colostrum were significantly higher (\( P < 0.05 \))
in LY-supplemented sows than those in CON group. There were no
significant differences in the concentrations of protein, fat, lactose
and solids no-fat in the milk at weaning between sows from the
CON and LY groups.

3.3. Plasma metabolites and the related enzyme activities

There were no significant differences in the plasma metabolites
of sows (glucose, triacylglycerol, urea) at d 1 of lactation or weaning
day between the 2 treatments (Table 4). However, the plasma ac-
tivities of AST, ALP and \( \gamma \)-GTP at d 1 of lactation, and ALT activity at
weaning day were lower (\( P < 0.05 \)) in sows fed LY diet than sows
fed CON diet.

3.4. Immunological and inflammatory response

The effects of dietary LY supplementation on immunological and
inflammatory response of sows at d 1 of lactation are presented in
Table 5. There were no significant differences for plasma concen-
trations of C3, C4, IgM, and C-RP between the 2 treatments. How-
ever, the plasma concentration of IgG was significantly increased by
dietary LY supplementation (\( P < 0.05 \)).

\[ \begin{array}{c|c|c}
\text{Item} & \text{Groups} & \\
\hline
\text{Birth, piglets/litter} & \text{CON} & \text{LY} \\
\hline
\text{Total born} & 14.22 ± 0.62 & 13.07 ± 0.40 \\
\text{Born alive} & 13.22 ± 0.53 & 12.88 ± 0.49 \\
\text{Stillborn} & 1.00 ± 0.36* & 0.19 ± 0.10n \\
\text{BW < 1.0 kg} & 3.22 ± 0.47* & 1.81 ± 0.36* \\
\text{Mean BW at birth, kg} & 1.29 ± 0.03 & 1.38 ± 0.04 \\
\text{Mean litter weight at birth, kg} & 16.82 ± 0.77 & 17.70 ± 0.74 \\
\hline
\text{Weaning day} & & \\
\text{Litter size, piglets/litter} & 11.61 ± 0.22 & 11.91 ± 0.33 \\
\text{Litter weight at weaning, kg} & 63.97 ± 4.79 & 68.24 ± 3.96 \\
\text{Average pig weight at weaning, kg} & 5.50 ± 0.07 & 5.71 ± 0.04 \\
\text{Average daily gain for piglet, g} & 205 ± 3.10 & 214 ± 2.29 \\
\text{Pre-weaning mortality, %} & 10.53 & 6.61 \\
\text{Sow backfat thickness, mm} & & \\
\text{Day 90 of gestation} & 15.20 ± 0.80 & 14.71 ± 0.60 \\
\text{Farrowing day} & 14.83 ± 0.59 & 13.22 ± 0.02 \\
\text{Weaning day} & 13.42 ± 1.32 & 12.12 ± 1.02 \\
\text{Decrease in backfat thickness during lactation} & 1.41 ± 0.28 & 1.10 ± 0.21 \\
\text{Lactation average daily feed intake, kg} & 5.34 ± 0.15 & 5.41 ± 0.22 \\
\end{array} \]

\( \text{CON} = \text{control; LY = live yeast.} \)

\( ^* \) Mean values within a row with different letter superscripts are significantly different (\( P < 0.05 \)).

\( ^n \) Values are expressed as means ± standard error, \( n = 10 \) for each group.

\[ \begin{array}{c|c|c}
\text{Colostrum} & \text{CON} & \text{LY} \\
\hline
\text{Fat} & 5.28 ± 1.06 & 5.57 ± 0.70 \\
\text{Solids-not-fat} & 13.58 ± 1.30* & 17.1 ± 1.35* \\
\text{Protein} & 5.12 ± 0.27* & 6.32 ± 0.50* \\
\text{Lactose} & 7.76 ± 0.45* & 9.46 ± 0.92* \\
\text{Milk at weaning} & & \\
\text{Fat} & 5.89 ± 0.41 & 6.25 ± 0.49 \\
\text{Solids-not-fat} & 9.32 ± 0.28 & 8.89 ± 0.16 \\
\text{Protein} & 3.50 ± 0.10 & 3.35 ± 0.06 \\
\text{Lactose} & 4.57 ± 0.15 & 4.72 ± 0.08 \\
\end{array} \]

CON = control; LY = live yeast.

\( ^* \) Values are expressed as means ± standard error, \( n = 10 \) for each group.

\[ \begin{array}{c|c|c}
\text{Plasma metabolites and the related enzyme activities} & \text{CON} & \text{LY} \\
\hline
\text{Lactation day} & & \\
\text{Urea, mmol/L} & 2.75 ± 0.24 & 2.52 ± 0.17 \\
\text{GLU, mmol/L} & 2.51 ± 0.36 & 2.40 ± 0.35 \\
\text{TG, mmol/L} & 0.28 ± 0.03 & 0.27 ± 0.05 \\
\text{ALT, U/L} & 27.39 ± 1.96 & 28.25 ± 2.71 \\
\text{AST, U/L} & 35.38 ± 3.64* & 24.45 ± 1.03* \\
\text{ALP, U/L} & 55.33 ± 7.60* & 33.33 ± 5.85* \\
\text{TBL, µmol/L} & 1.98 ± 0.29 & 2.07 ± 0.23 \\
\text{γ-GGT, U/L} & 64.33 ± 10.38* & 36.80 ± 6.07* \\
\hline
\text{Weaning day} & & \\
\text{Urea, mmol/L} & 5.34 ± 0.46 & 4.91 ± 0.52 \\
\text{GLU, mmol/L} & 3.50 ± 0.45 & 2.67 ± 0.29 \\
\text{TG, mmol/L} & 0.32 ± 0.04 & 0.25 ± 0.03 \\
\text{ALT, U/L} & 47.19 ± 5.31* & 34.06 ± 2.12* \\
\text{AST, U/L} & 36.60 ± 7.37 & 27.45 ± 3.25 \\
\text{ALP, U/L} & 36.00 ± 5.04 & 25.50 ± 2.82 \\
\text{TBL, µmol/L} & 0.29 ± 0.12 & 0.51 ± 0.16 \\
\text{γ-GGT, U/L} & 46.19 ± 4.11 & 37.60 ± 4.37 \\
\end{array} \]

CON = control; LY = live yeast; GLU = glucose; TG = triglyceride; ALT = alanine aminotransferase; AST = aspartate aminotransferase; ALP = alkaline phosphatase; TBL = total bilirubin; γ-GGT = gamma-glutamyl transpeptidase.

\( ^* \) Mean values within a row with different letter superscripts are significantly different (\( P < 0.05 \)).

\( ^n \) Values are expressed as means ± standard error, \( n = 10 \) for each group.
In the present study, sows supplemented with LY showed no significant effects on the number of total born piglets. The data was consistent with the results of previous studies (Jang et al., 2013; Jurgens et al., 1997). It was reported that the litter size in the pig was mainly determined by the fertilization rate and prenatal death occurring in the early pregnancy (Edwards et al., 2012), and is not likely to be influenced by diets during the late gestation. Interestingly, maternal LY supplementation decreased the number of stillborn piglets and low BW piglets, a possible explanation could be that the probiotic microorganisms can positively modulate the gut microbial communities, and thereby improve the health status of sows (Chaucheyras-Durand and Durand, 2009; Musa et al., 2009). A recent study reported that yeast supplementation to sows during gestation and lactation could improve the relative abundance of beneficial Bacilli (Paraprevotella, Roseburia, and Eubacterium) and inhibit the growth of pathogenic bacteria such as Proteobacteria in sows (Hasan et al., 2018). In addition, studies have shown that gut microbiota impact significantly on the relative composition and abundance of bile acids across multiple body compartments, and an elevation of serum bile acids shows a significantly increased risk for intrauterine fetal death (Geenes et al., 2014; Rezai et al., 2015; Swann et al., 2011). Tsai et al. (2015) also reported that LY could contribute to the regulation of bile acids metabolism in diet-induced obese mice. Additionally, LY supplementation showed no significant effects on the weight of litter, piglet at birth and weaning in this study. This result was in agreement with previous studies (Jurgens et al., 1997; Veum et al., 1995), which showed that sows supplemented with yeast during gestation and lactation had limited effect on litter performance. In contrast, beneficial effects of yeast culture supplementation to gestation and lactation diets have been reported by Kim et al. (2008) on litter weight gain. However, unlike LY, yeast culture contains cereal grain byproduct medium, which may have additional nutrients that may increase litter weight gain. Furthermore, these different results of LY affecting the reproductive performance of sows may be also derived from different strains and dose of LY, as well as the length of time LY was supplied.

Milk composition had an important influence on the growth and development of suckling piglets (Lewis et al., 1978; Wang et al., 2011). The current findings showed that LY supplementation during gestation and lactation increased the concentrations of colostral protein, lactose and solids-not-fat. Similar results were observed in the probiotics-supplemented sows during the pre- and post-farrowing period (Alexopoulos et al., 2001, 2004), the underlying mechanism may be associated with the optimization of bacterial profile in the gut, which would enhance the utilization of nutrients for colostrum composition (Kritas et al., 2006). In support of this possibility, Erasmus et al. (1992) showed supplementation of yeast culture tended to increase microbial protein synthesis in dairy cows and significantly altered the amino acid profile of the duodenal digesta. In addition, recent studies have also reported that LY enhanced fiber degradation in cows and dogs by increasing the fibrolytic bacteria and fungi colonization in the gastrointestinal tract (Chaucheyras-Durand et al., 2016; Stercova et al., 2016), thus more energy and nutrients could be available to improve colostrum components. In this study, however, the growth performance of suckling piglets did not significantly differ between groups. In fact, the amount of colostrum intake may be more important than the composition for the growth of piglets during the suckling period (Quesnel et al., 2012). Besides, Declerck et al. (2016) reported that colostrum intake was positively associated with weaning weight and negatively associated with mortality during the suckling and the nursery period. It is widely accepted that maternal metabolic burdens are greatly increased during late gestation and lactation, leading to an increased mobilization of body reserves (Bell, 1995). Besides, liver plays a crucial role in the control of whole body metabolism (Rui, 2011). Serum enzymes such as AST, ALP, ALT and γ-GGT are key biochemical parameters of liver health and function, and an excessive accumulation of these enzymes in the serum often prefigures liver injury (Bintvihok and Kositcharoenkul, 2006; Khanyile et al., 2016). A recent study has been shown that the plasma level of γ-GGT around farrowing was negatively related to litter body weight gain of sows (Loisel et al., 2014). In this study, the lower activities of plasma AST, ALP, ALT and γ-GGT in LY-supplemented sows indicated that LY plays an important role in improving liver function, which may partially explain the better milk composition and fetal development observed in this study.

Yeast and yeast products derived from S. cerevisiae are immunomodulating compounds that have positive effects both directly and indirectly on the immune system (Kogan and Kocher, 2007), thereby mitigating the potential negative effects associated with stress and disease. In the present study, a significant increase in plasma concentration of IgG at d 1 of lactation was observed in sows with LY supplementation during late gestation. Similarly, some other studies also showed that dietary supplementation with LY to sows during late gestation increased IgG concentration in the colostrum and milk (Jurgens et al., 1997; Zanello et al., 2016). As IgG constitutes the majority of total immunoglobulins in the periparturient period in the blood and mammary secretions (Klobasa et al., 1987), colostrum IgG is partly translocated from blood to the mammary gland probably by the neonatal Fc receptor (FcRn) receptor (Salmon et al., 2009), the increased IgG in colostrum and milk of sows fed LY may also suggest a higher blood concentration of IgG. The improvements on immune responses and animal health had been ascribed to the modulating role of LY on bacterial profile, and the α-mannan or β-glucan contained on the yeast cell wall (Jiang et al., 2015; Kogan and Kocher, 2007; Volman et al., 2008). Our recent study also showed that dietary supplementation with β-glucans can improve growth performance and partially alleviated the inflammation response of pigs after lipopolysaccharide challenge (Wu et al., 2018).

5. Conclusion
Collectively, the results of present study indicated that dietary supplementation of LY to sows in late gestation and lactation decreased the number of stillborn piglets and low BW piglets, improved colostrum composition and parameters related to liver function and immunity.

Conflict of interest
We declare that we have no financial and personal relationships with other people or organizations that can inappropriately

### Table 5
Effects of dietary live yeast supplementation on immunological and inflammatory parameters in plasma of sows at d 1 of lactation.

| Item          | Groups | CON            | LY              |
|--------------|--------|----------------|-----------------|
| C3, mg/L     |        | 28.13 ± 0.74   | 30.86 ± 1.45    |
| C4, mg/L     |        | 17.29 ± 2.35   | 17.67 ± 2.58    |
| IgG, g/L     |        | 3.33 ± 0.42a   | 4.91 ± 0.37a    |
| IgM, g/L     |        | 0.97 ± 0.09    | 1.00 ± 0.09     |
| C-RP, mg/L   |        | 25.63 ± 0.82   | 25.24 ± 0.26    |

CON = control; LY = live yeast; C3 = complement 3; C4 = complement 4; IgG = immunoglobulin G; IgM = immunoglobulin M; C-RP = C-reactive protein.

a Mean values within a row with different letter superscripts are significantly different (P < 0.05).

1 Values are expressed as means ± standard error, n = 10 for each group.
influence our work, there is no professional or other personal interest of any nature or kind in any product, service and/or company that could be construed as influencing the content of this paper.

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