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Research Article

Effect of *Aloe vera* Gel Supplementation as a Natural Source of Mannan Oligosaccharides on Productive Performance and Oxidative Status of Growing Rabbits

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Abstract:

This study was conducted to shed more light on the effect of *Aloe vera* gel (AVG) supplementation as a natural source of mannan oligosaccharides (MOS) on productive performance and oxidative status of growing rabbits. Sixty unsexed NZW rabbits, about 5 weeks of age with an initial body weight (625±5g) were divided randomly into five equal experimental groups with three replicates of 4 rabbits each. The 1st group (control) was fed a diet without any supplementation; the 2nd group fed the basal diet supplemented with 0.1% mannan oligosaccharides MOS, while AVG was added at levels of 0.05, 0.1, and 0.2%, respectively at the 3rd, 4th and 5th groups. Productive performance traits (body weight, weight gain, feed intake, feed conversion ratio, relative growth rate, and performance index) were determined during the entire experimental period. At the end of the experimental period, the oxidative status indices were measured in plasma. Results showed that, the best values of productive performance traits were achieved in groups fed a diet supplemented with 0.2% AVG compared to the control. Additionally, rabbits fed diet supplemented with AVG at the level of 0.2% possessed the highest activities of CAT, GPx and SOD followed by those received 0.1% and then those treated by 0.05% respectively, as compared to the negative control group. It could be recommended that, *Aloe vera* considered a natural source of mannan oligosaccharides and can be used in the diets of growing NZW rabbits as a feed additive up to a level of 0.2% to improve productive performance and oxidative status.

1. Introduction

Rabbit production holds global significance as a multifaceted industry contributing to both meat and fur production. As a lean and nutritious source of protein, rabbit meat plays a vital role in addressing global food security challenges, providing an alternative to traditional livestock. Additionally, the fur industry relies on rabbits for their soft and luxurious pelts, contributing to the fashion and textile sectors. The sustainability and efficiency of rabbit farming make it an important aspect of the global agricultural landscape, catering to diverse consumer demands while minimizing environmental impact compared to larger livestock industries. Understanding the interconnectedness of rabbit production with economic, dietary, and fashion considerations underscores its global importance (Elstiha et al., 2019).

In recent years, concerns over the widespread use of feed antibiotics have prompted numerous countries and organizations to develop various feeding strategies to eliminate their use and mitigate their adverse effects on animal, bird, and human health. As a consequence, researchers have embarked on a quest for natural alternatives to antibiotics, particularly herbal extracts, oils, and powders. The global quest was started for equivalently effective alternatives without detrimental effects on consumer health and animal welfare. Possible natural substitutes for antibiotics as growth promoters in poultry diets include herbal extracts, spices, and some of their constituents “phytogenic additives” (Alagawany et al., 2020; Abd El-Hack et al., 2022b; El Basuini et al., 2023; El-Rayes et al., 2023).

Plants play an important role in the identification of novel medicinal medicines. They have garnered a lot of interest lately for the isolation of biologically active compounds for improves productivity and illness treatment. Traditional remedies are used by around 80% of the inhabitants of poor nations to treat health issues. Biologically active chemicals originating from medicinal plants have been used to develop novel chemical leads for the pharmaceutical sector. Evidence suggests that just 1% of the 500,000 plant species found globally have been phytochemically examined, indicating that medicinal plants have a high potential for identifying novel bioactive chemicals. Phytogenic feed additives are plant-derived materials used in animal feeding to increase the performance of poultry. They may assuage customers' growing concerns because they are safe and effective. Phytogenics may also help to mitigate the severe environmental issue of bacterial resistance, which is produced by the usage of antibiotics as growth promoter chemicals (Perić et al., 2009).

Phytogenics can increase food consumption and conversion, as well as digestibility and weight growth in poultry. However, the mechanism of action of these compounds are yet unknown (Scheuermann et al., 2009). Studies on the use of phytogenics in rabbit feeding have yielded inconclusive - either good or negative - effects on poultry performance (Botsoglou et al., 2002; Ertas et al., 2005; Cross et al., 2007; Ocak et al., 2008).

In the poultry business, the usage of phytobiotic compounds in their diets became a widespread practice in order to support high performance by poultry. The good impact on feed intake, digestive secretions, immunological stimulation, antibacterial and coccidiostatical, antiviral, or anti-inflammatory activity of botanical supplements in poultry may be the source of these benefits (Reda et al., 2021; El Basuini et al., 2023; El-Rayes et al., 2023). To distinguish between the plant products used in veterinary medicine (prophylaxis and therapy of diagnosed health problems), Windisch and Kroismayr, (2006) redefined phytobiotic as plant-derived compounds added to feed to enhance the performance of poultry. In addition to their antimicrobial activity (Dorman and Deans, 2000), phytobiotic compounds exhibit antioxidants activities (Botsoglou et al., 2002) and can stimulate animal digestive systems (Ramakrishna et al., 2003) by increasing digestive enzymes secretion and improving the utilization of digestive products through enhanced liver functions (Hernandez et al., 2004).

In recent years, there has been a growing interest in sustainable and natural feed additives, with Mannan Oligosaccharides (MOS) emerging as a noteworthy example. MOS is a prebiotic derived from yeast cell walls, known for its ability to enhance the gut health of animals. As the agricultural industry seeks eco-friendly and health-conscious solutions, MOS has gained traction for its role in promoting beneficial gut microflora, improving nutrient absorption, and contributing to overall animal well-being. This shift towards sustainable feed additives reflects a broader commitment to environmentally friendly and ethically sourced practices in animal agriculture (Muhammad et al., 2020).

In this sense, *Aloe vera* is one of such plants, having a great medicinal potential (Rodrigues et al., 2018). *Aloe vera* is a succulent, stemless herb found widely in Egypt, having more than 70 biologically active compounds. Many studies have shown antibacterial, antiseptic, anti-inflammatory and immune-modulator effects of *Aloe vera* (El-Kholy et al., 2022). Many studies have also shown antioxidant and anti-cancerous properties of *Aloe vera* (Abo El-Azayem et al., 2023; Quaye et al., 2023).

Major ingredients of *Aloe vera* include anthraquinones, saccharides, vitamins, enzymes, and low-molecular-weight compounds (Choi and Chung, 2003) which give *Aloe vera* its anti-inflammatory, immunomodulatory, wound-healing, anti-viral, anti-fungal, anti-tumor, anti-diabetic, and anti-oxidant effects (Christaki and Florou-Paneri, 2010). Numerous studies suggest that many benefits of *Aloe vera* are attributable to polysaccharides contained in *Aloe vera* gel, which compose a large part of dry matter in this gel (Hamman, 2008). In other words, almost 60% of dry matter of *Aloe vera* gel is composed of polysaccharides (McAnalley, 1989). A compound often analyzed by researchers is the polysaccharide acemannan which has immunomodulatory, antimicrobial, and anti-tumor effects (Choi and Chung, 2003). Studies discovered different properties of *Aloe vera* gel, including wound healing, anti-parasitic, anti-viral, anti-fungal and anti-bacterial properties (Boudreau and Beland, 2006; Reynolds and Dweck, 1999). An important *Aloe vera* gel complex which has received attention from researchers is the polysaccharide acemannan – a mannose polymer (Reynolds and Dweck, 1999).

The research gap in Mannan Oligosaccharides (MOS) supplementation in rabbit diets sheds light on numerous unanswered questions. There is a need for further research to assess various natural alternatives as sources of Mannan Oligosaccharides, such as aloe vera gel, and their impact on production performance, physiological status, oxidative stress, and nutrient digestibility parameters. Additionally, exploring the influence of these alternatives on the microbial environment of the digestive tract is essential. This emphasizes the necessity for comprehensive investigations to enhance our understanding of the diverse aspects related to the utilization of different natural sources of Mannan Oligosaccharides in rabbit nutrition. So, the goal of the current study was to shed more light on the effect of *Aloe vera* gel (AVG) supplementation as a natural source of mannan oligosaccharides (MOS) on productive performance and oxidative status of growing rabbits.

2. Materials and Methods

2.1. Experimental Design

2.1.1. Collection and preparation of Aloe vera gel

For the extraction of *Aloe vera* gel, fresh *Aloe vera* leaves were collected from a private nursery for ornamental plants in Kafr El-Sheikh city. The leaves were cleaned with water and the *Aloe vera* gel was extracted from the leaf manually by making a cut. The latex of the leaf was removed, and the gel was collected in a beaker. The collected gel was shaken for 5-7 min to ensure thorough mixing and was then kept at -20ºC until use (Shokraneh et al., 2016).

2.1.2. Animals and management

Sixty (unsexed) NZW rabbits, about 5 weeks of age and with an initial body weight (625±5g), kept at the rabbitry farm of Animal Production Research Institute "Sakha Station". Rabbits divided randomly according to their weights into five equal experimental groups with three replicates of 4 rabbits each. The rabbits housed individually in one flat galvanized wire batteries (50×50×40 cm) with fodder and automatic nipple drinkers. The rabbits fed the experimental diets *ad libitum* for 8 weeks. All rabbits subjected to the same managerial, hygienic and environmental conditions.

2.1.3. Experimental diet

The basal diet formulated to cover all essential nutrient requirements for growing rabbits according to (De Blas and Mateos, 1998), as shown in (Table 1). The experimental groups of rabbits fed a basal diet supplemented with different levels of *Aloe vera* gel (AVG) or mannan oligosaccharides (MOS) as follow:

Control	Basal diet without any supplementation.
T1	Basal diet supplemented with 0.1% MOS.
T2	Basal diet supplemented with 0.05% AVG.
T3	Basal diet supplemented with 0.1% AVG.
T4	Basal diet supplemented with 0.2% AVG.

Table (1): The composition and calculated analysis of basal diet.

Ingredients	%
Yellow corn	18.00
Wheat bran	16.50
Barley grain	13.00
Soybean meal (44%)	15.00
Berseem hay	33.00
Molasses	2.00
Limestone	1.00
Dicalcium Phosphate	0.60
Premix[1]	0.20
DL-Methionine	0.10
Common salt	0.50
Anti-toxicants	0.10
Total	100
Calculated analysis	
Crude protein	16.70
Digestible energy, Kcal/kg[2]	2530
Crude fiber	13.52
Ether extract	2.31
DL-Methionine	0.40
Lysine	1.03
Calcium	0.94
Total phosphorus	0.45

[1] Each 3 kg of premix contained: Vitamin A 12000000 IU, V.D3 2200000 IU, V.E 10000 mg, V.K3 2000 mg, V.B1 1000 mg, V.B2 4000 mg, V.B6 1500 mg, V.B 12 10 mg, Pantothenic Acid 10000 mg, Niacin 20000 mg, Biotin 50 mg, Folic Acid 1000 mg, Coline chloride 500 gm, Selenium 100 mg, Copper 10000 mg, Iron 30000 mg, Manganese 55000 mg, Zinc 50000 mg, Iodine 1000 mg and carrier CaCoi to 3000 gm.

[2] Calculated according to Fekete and Gippert (1985), as follow:

DE (kcal/kg) = 4253 - 32.8(%CF) – 144.4 (% ash)

2.2. Measurements

2.2.1. Performance traits

Individual live body weight and feed intake recorded weekly from 5 weeks until 13 weeks of age. Relative growth rate (RGR), weight gain, feed conversion ratio (FCR) and performance index (PI) calculated on a group basis as follows:

$$\text{Relative growth rate} = \frac{w2 - w1}{1/2(w1 + w2)} \times 100$$

Wl = the initial weight, and W2 = the final body weight.

WG = Final LBW – Initial LBW

$$\text{FCR} = \frac{\text{Feed consumed (g) during a certain period}}{\text{Body weight gain (g) during the same period}}$$

$$\text{Performance index} = 100 * \frac{\text{Final live body weight (kg)}}{\text{Feed conversion ratio}}$$

2.2.2. Oxidative status:

At the end of the trial (13-week-old), three blood samples were collected from each treatment into heparinized tubes and centrifuged for 15 min at 3000 rpm to obtain plasma. Plasma samples were examined for the level of total antioxidants capacity (TAC), the activity of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GSH-Px) and the concentration of malondialdehyde (MDA) using a microplate spectrophotometer with a commercial detection kit (Bio-diagnostic, Egypt), following the manufacturer's instructions.

2.3. Statistical analysis

Data were statistically analyzed by one-way ANOVA, using the general linear model procedure (SAS, 1996). Tests of significance for differences among treatments were done according to Duncan (1955). The statistical model was used for the analysis of variance to estimate the effect of *Aloe vera* gel supplementation levels on NZW growing rabbits performance and oxidative status as follows:

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where:

Y_{ij} = the observations. μ = Overall mean.

T_i = effect treatments (i = 1, 2, 3, 4 and 5).

e_{ij} = residual effects (Random error).

3. Results

3.1. Productive performance traits

Data of growing NZW rabbits body weight, weight gain, feed intake, feed conversion ratio, growth rate and performance index as influenced by *Aloe vera* gel supplementation at different levels (0.05, 0.1 and 0.2%) from the 5th to 13th weeks of age are illustrated in Table (2). As influenced by AVG supplementation, the body weight of growing NZW rabbits was significantly ($P \leqslant 0.01$) improved at the end of the experimental period, compared to the negative control group. Whereas, rabbits fed a diet supplemented with AVG at the level of 0.2% had significantly ($p \leqslant 0.01$) the highest body weight followed by those received AVG at the level of 0.1%, and then those fed AVG at the level of 0.05% by (11.86, 9.85, and 7.73%), respectively as compared to the negative control group. Additionally, Weight gain of growing rabbits were significantly ($P \leqslant 0.01$) increased by increasing the supplementation level of AVG at the interval period from (5-9), (9-13), and (5-13) weeks of age.

Feed intake (FI) and feed conversion ratio (FCR) of growing NZW rabbits were improved ($P \leqslant 0.01$) by AVG supplementation levels at all the interval periods. Whereas, rabbits received diet supplemented with AVG at the level of 0.2% possessed significantly ($p \leqslant 0.01$) the best feed intake and feed conversion ratio at all the interval periods, compared with the negative control group.

During the interval period from (5-9) wks, the growth rate of growing NZW fed diet supplemented with AVG at levels of 0.05, 0.1 and 0.2% was significantly (P ≤0.01) improved by 17.00, 17.81, and 18.38%, those received AVG at 0.1% and then those treated by 0.05% by 9.75, 7.14, and 4.37% respectively, comparable with the negative control.

Table (2): Effect *Aloe vera* gel supplementation levels on performance traits of growing NZW rabbits

	Negative control	MOS 0.01% Positive control	% *Aloe vera* gel			SEM	Sig
			0.05	0.1	0.2		
No. of rabbits	12	12	12	12	12	--	--
Body weight:							
Initial (5 wks)	610.83	610.83	610.83	610.83	610.83	±48.57	NS
Final (13 wks)	2203.33^{c}	2299.17b^{c}	2373.75^{ab}	2420.42^{a}	2464.58^{a}	±35.74	**
Daily weight gain(g):							
5-9 wks	27.75^{c}	29.69^{bc}	36.41^{a}	36.88^{a}	37.20^{a}	±1.41	**
9-13 wks	29.12^{ab}	30.61^{a}	26.55^{b}	27.75^{b}	29.00^{ab}	±1.73	**
5-13 wks	28.44^{c}	30.15^{b}	31.48^{ab}	32.31^{a}	33.10^{a}	±1.52	**
Daily feed intake (g):							
5-9 wks	74.82^{a}	73.14^{a}	69.45^{ab}	66.16^{b}	68.16^{ab}	±1.69	**
9-13 wks	121.14^{a}	100.43^{c}	110.74^{b}	100.91^{c}	92.69^{d}	±2.54	**
5-13 wks	97.98^{a}	86.78^{bc}	91.57^{b}	83.53^{cd}	80.42^{d}	±1.91	**
Feed conversion rate:							
5-9 wks	2.69^{a}	2.46^{b}	1.91^{c}	1.79^{c}	1.83^{c}	±0.06	**
9-13 wks	4.16^{a}	3.28^{c}	4.17^{a}	3.63^{b}	3.19^{c}	±0.11	**
5-13 wks	3.45^{a}	2.87^{b}	2.91^{b}	2.58^{c}	2.43^{c}	±0.80	**
Growth rate (%):							
5-9 wks	77.76^{c}	80.98^{b}	90.98^{a}	91.61^{a}	92.05^{a}	±1.42	**
9-13 wks	45.41^{a}	45.82^{a}	37.13^{b}	38.24^{b}	39.45^{b}	±3.25	**
5-13 wks	113.18^{c}	116.04^{bc}	118.13^{b}	121.26^{ab}	124.22^{a}	±1.61	**
Performance index (%)	63.86^{d}	79.55^{c}	80.71^{c}	93.44^{b}	101.40^{a}	±2.13	**

-Means of each raw followed by the same letter are not significantly different at the 5% level according Duncan's Multiple Range Test.
-** indicate significance at P<0.01 - SEM indicate standard error of the mean

respectively, as comparable with the negative control group. The same direction was observed during the interval period from (5-13) wks, the growth rate of growing NZW fed diet supplemented with AVG at levels of 0.05, 0.1 and 0.2% was significantly (P≤0.01) improved by 4.37, 7.14, and 9.75%, respectively, as comparable with the negative control group. On the other hand, during the interval period from (5-9) wks, the growth rate of growing NZW fed diet supplemented with AVG at levels of 0.05, 0.1 and 0.2% was significantly (P≤0.01) decreased by 18.23, 15.79, and 13.12%, respectively, as compared to the negative control group.

Performance index (PI) of growing NZW rabbits was affected significantly (P≤0.01) by AVG supplementation levels. Growing NZW rabbits fed diet supplemented with AVG at the level of 0.2% had significantly (P≤0.01) the highest performance index followed by

3.2. Oxidative status

Data illustrated in Table (3) shows the effect of AVG supplementation levels on the oxidative status of NZW rabbits.

Results indicated a significant (P≤0.01) improvement in the antioxidant status; this is evident through a significant increase in the activities of catalase (CAT), glutathione peroxidase (GPx) and super oxide dismutase (SOD) and decreasing the amount of MDA in blood plasma. The Activity of CAT was significantly (P≤0.01) increased with increasing level of AVF. Rabbits fed diet supplemented with AVG at the level of 0.2% possessed the highest activities of CAT, GPx and SOD followed by those received 0.1% and then those treated by 0.05% respectively, as compared to the negative control group.

Table (3): Effect *Aloe vera* gel supplementation levels on oxidative status of growing NZW rabbits

Items	Negative control	MOS 0.01% Positive control	% *Aloe vera* gel			SEM	Sig.
			0.05	**0.1**	**0.2**		
CAT (µmol. min^{-1}.mg protein^{-1})	1752.33^{d}	1838^{c}	1870.33^{bc}	1909.67^{ab}	1936.67^{a}	±14.94	**
GPx (µmol. min^{-1}.mg protein^{-1})	1304.33^{d}	1345^{d}	1448.67^{c}	1547.67^{b}	1672^{a}	±26.08	**
SOD (µmol. min^{-1}.mg protein^{-1}	434.33^{e}	462^{d}	619.67^{c}	675.67^{b}	712.67^{a}	±7.56	**
MDA (nmol /ml)	6.40^{a}	5.44^{b}	4.51^{c}	4.54^{c}	3.97^{c}	±0.19	**

-Means of each raw followed by the same letter are not significantly different at the 5% level according Duncan's Multiple Range Test. -
** indicate significance at P<0.01 - SEM indicate standard error of the mean

4. Discussion

The improvement of productive performance of growing NZW rabbits may be due to the bioactive compounds found in Aloe vera. Whereas, the AVG is rich in flavonoids, terpenoids, lectins (Boudreauand Beland, 2006; Harlev et al., 2012) fatty acids, anthraquinones (Surjushe et al., 2008), mono- and polysaccharides (pectins, hemicelluloses, glucomannan), tannins, sterols (campesterol, β-sitosterol), enzymes, salicylic acid, minerals (calcium, chromium, copper, iron, magnesium, manganese, potassium, phosphorus, sodium and zinc) and vitamins (A, C, E, β-carotene, B1, B2, B3, B6, choline, B12, folic acid) (Sahu et al., 2013; Rodrigues et al., 2018). Moreover, Danhoff and McAnally (1988) reported that AV accelerated the growth of new cells, thereby resulting to increased body weight. Durrani et al. (2008) reported that the higher BWG and improved FCR values of the broilers given 10 mL of aqueous extract of AV gel/liter of drinking water could be due to better performance of the broilers and the diversified antimicrobial activities of Aloe gel. Furthermore, Aloe vera gel has several medicinal uses including the correction of digestive disorders. In addition, it has positive effects on gastrointestinal tract (GIT) microflora. In addition, Aloe vera contains growth hormones (auxins and gibberellins) and polysaccharide (glucomannan) which reduced the inflammation and haemorrhages in the intestines that led to better digestion and enhanced weight gain (Surjushe et al. 2008).

The positive effect of AVG on feed intake and feed conversion ratio may be due to the existence of phytogenic substance in aloe vera that may stimulate appetite, digestive enzymes and nutrient absorption causing improvement of performance. Herbs consuming results to increase appetite and endogenous secretions, which in turn, improve performance traits, therefore the present study results regarding of body weight and weight gain are in accordance to other scholar findings.

These findings were in agreement with the observations of Darabighane et al. (2011); Singh et al. (2013); Darabighane and Nahashon (2014); Singh (2014); Nassary and Waziri (2019); El-Kholy et al. (2022); Abo El-Azayem et al. (2023) who observed significant increase in body weight due to dietary supplementation of *Aloe vera*. On the contrary, Sinurat et al. (2002); Yadav, (2015) who also observed no significant difference in body weight due to supplementation of *Aloe vera* powder in broiler diet. Variation in results might be due to species/strain differences, agro-climatic differences, difference in levels of Aloe vera and seasons, etc.

Reactive oxygen species (ROS) is continuously produced during normal physiological events and they can easily initiate the per-oxidation of membrane lipids, leading to the accumulation of lipid peroxides. Under pathological conditions, ROS is overproduced and results in oxidative stress. Antioxidants are substances that delay or prevent the oxidation of cellular oxidizable substrates. They exert their effect by scavenging reactive oxygen species, activating a battery of detoxifying proteins or preventing the generation of reactive oxygen species (Ghiselli et al., 2000). Stress produces free radicals, which can damage cell membranes by causing the polyunsaturated fatty acids that surround the membranes to oxidize (Rehman et al., 2018). In recent years, there has been an increasing trend in finding natural antioxidants, which can protect the body from free radicals and retard the progress of many chronic diseases (Alagawany et al., 2020; Abd El-Hack et al., 2022b; El Basuini et al., 2023; El-Rayes et al., 2023).

Data presented in Table 3 revealed the different dietary levels of AVG leaves powder influences significantly the lipid peroxidation (LPO) by decreasing MDA values. Lipid peroxidation causes a negative effect on membrane fluidity, which, in turn, impacts immunological responses (Van der Paal et al., 2016). The SOD shields the cells from oxygen-free radicals by catalyzing the elimination of superoxide radicals, which harms the membrane and biological structures. The CAT was shown to be charged for the detoxification of H2O (Mahboob et al., 2005).

In this study, dietary AVG levels were observed in the activities of SOD and CAT, which were significantly higher in rabbits fed dietary AVG, especially at the highest levels (0.2%). Phenolic compounds in AVG leaves powder may play a crucial role in homeostatic animal antioxidant reactions and antioxidant activities (SOD, CAT, and GPx) which were significantly increased in treated rabbits with AVG. The observed increase in antioxidant activities and decline in the activities of MDA in AV leaves powder-treated rabbits suggest its potential anti-lipid peroxidative and antioxidant effects. Potent antioxidant effects of AVG, including the ability to scavenge superoxide anions have been attributed to the caffeoyl group of isorabaichromone, a derivative of aloesin (C-glycosylated 5- methylchromone) (Boudreau and Beland, 2006).

Many reports cited that, *Aloe vera* possess excellent antioxidant and free radical scavenging activity. This effect might be attributed to its polyphenolic content and other phytochemicals constituents (Patel et al., 2012; Rodrigues et al., 2018).

Our results of oxidative status are compatible with those observed by El-Kholy et al. (2022); Haritha et al., (2004) who found that the administration of AVG in growing rabbit or Wistar rats diet's improved the oxidative status.

5. Conclusions

In conclusion, It could be recommended that, Aloe vera considered a natural source of mannan oligosaccharides and can be used in the diets of growing NZW rabbits as a feed additive up to a level of 0.2% to improve productive performance and oxidative status.

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