

Research Article

# Influence of rice husk ash-derived silica nanoparticles on sweetcorn (*Zea mays* L. *sachharata*) seed germination

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# Abstract

In agriculture, the utilization of nanomaterials has garnered significant global attention. This research adopts a pioneering approach to investigate the influence of nanosilica on the germination dynamics of sweetcorn seeds. The present study aimed to synthesize and analyze an amorphous nano-silica material using rice husk ash (RHA) and its impact on the germination of sweetcorn seeds (*Zea mays* L. *sachharata*). The extracted nano-silica particles dispersed into six rates of suspensions (0, 100, 200, 300, 400 and 500 ppm) were used to study their effects on seed germination. The synthesized amorphous nano-silica was determined for size, shape, and elemental content. The amorphous nature of the silica sample was confirmed by transmission electron microscopy-selected area electron diffraction (ED) patterns and X-ray diffraction (XRD), whereas siloxane and silanol groups were mainly detected by Fourier-transform infrared (FT-IR) spectroscopy. Image obtained using scanning electron microscopy (SEM) revealed the presence of original nanoparticles alongside secondary microparticles, probably due to agglomeration. Particles in the extracted amorphous silica had an average diameter of 35 nm. Nano-silica powder was amorphous, according to XRD. As per the EDS analysis, the extracted silica sample is 96.87 % pure. The amorphous nano-silica significantly boosted germination metrics such as germination percentage, germination index, vigour index, and mean germination time of sweetcorn. With the addition of 300 ppm nano-silica, the germination percentage increased by 40.1%, the germination index by 96%, and the vigor index by 120% over control seeds. The improvement of seed germination by amorphous nano-silica in seed germination.

Keywords: Amorphous nano-silica, Synthesis, Rice husk ash, Seed germination, Sweetcorn

# INTRODUCTION

Corn (*Zea mays* L.), commonly referred to as maize, is the world's principal cereal crop and plays an important part in human nutrition. It is the most important cereal crop, with rice and wheat close behind. During the financial year 2021, India produced over 31 million metric tons of maize. According to the fourth advance estimate for 2022-23, production has grown, reaching 32.62 million metric tons (Ministry of Agriculture and

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Farmers Welfare, 2022, https://pib.gov.in). Sweetcorn, also known as sugar corn, is a specially bred maize hybrid designed to increase its sugar content. Its introduction to India can be traced back to its origin in the United States. Sweetcorn is valued in the human diet for its health-promoting nutritional properties. Its optimal nutritional quality is achieved when it contains 72.7% moisture and 22.3% total solids, including 81% carbohydrates, 13% proteins, and 3.5% lipids. At the optimal market maturity stage, sweet corn typically contains around 5-6% sugar, 10-11% starch, 3% water-soluble polysaccharides, and 70% water content. Sweetcorn has a significant oil content, with linolenic acid and oleic acid being the predominant fatty acids at 50% and 30%, respectively. It also provides moderate amounts of potassium, protein, and A-vitamin. Farmers cultivating maize are increasingly transitioning to sweetcorn cultivation, enticed by its higher income, as sweet corn demand and market value continue to rise (Swapna et al., 2020).

Seeds are the consequence of plant survival mechanisms and serve as the material base for production (Sun et al., 2021). Plant growth is directly affected by the growth condition during seed germination (Li et al., 2008). After sowing, rapid germination of corn seeds results in early-stage, homogenous, and vigorous seedlings, establishing the foundation for high-quality, highyield harvests (Epstein, 1994). The worldwide system for classifying soil places silicon (Si) after nitrogen (N), phosphorus (P), and potassium (K) as the fourth element. Si is abundant in soils, but most of it cannot be properly absorbed by plants (Zhu and Gong, 2013). Si is a crucial element for humans and animals, but is not obligatory for higher plants (Rahman et al., 2017). The significance of Silica to plants has not been established however, laboratory and field studies have suggested that it positively fosters plant growth (Yan et al., 2018). In crops, Si has been noticed to increase drought tolerance, resistance to pests and diseases (Ghareeb et al., 2011; Kurabachew and Wydra, 2014), yield (Rambo et al., 2011), and the negative effects of some toxic elements in the soil (Rizwan et al., 2012 and Marmiroli et al., 2014). Si can improve seed vigor, boost seedling dry weight, enhance seedling relative growth rates, and encourage seedling cotyledon growth, providing the prerequisites for improving yield and enhancing quality (Artyszak, 2018; Shi, 2014; Dong et al., 2017 and Biju et al., 2017).

Utilizing RHA, known to have a high silica content, is one of the most often used sources of silicon (Amutha *et al.*, 2010). By exploiting an abundant, low-value agricultural byproduct that can help with trash disposal issues, rice husk ash (RHA) positively impact the environment and the economy. As established by numerous researchers (Sun, 2020; Bassiouni *et al.*, 2020; Patil *et al.*, 2018; Anand *et al.*, 2018), Si treatment can improve rice growth, but its effects on sweetcorn seed germination and growth are barely understood. The investigation aimed to assess the impact of various nano-silica concentrations on the seed germination metrics of sweetcorn. This work was made to synthesize and characterize nanosilica powder from RHA and assess the possibilities of using this powder as a Silica source for seed germination of sweetcorn plants.

# MATERIALS AND METHODS

# Preparation and characterization of amorphous nanosilica

The present work was conducted in the Centre for Agricultural Nanotechnology and Department of Agronomy at Tamil Nadu Agricultural University, Coimbatore with the objective to synthesize and characterize the amorphous nano-silica from rice hush ash (RHA) and aimed to examine the influence of amorphous nano-silica synthesized from RHA on sweetcorn seed germination. An et al. (2011) successfully synthesized amorphous silicon dioxide nanoparticles (SiO<sub>2</sub>-NPs) from acid-treated RHA using a two-step acid-alkali reaction method. This technique was modified to be less harmful to the environment. The rice husk was washed twice with running tap water and distilled water before being leached for 30 minutes with a 1:5 ratio of one normal hydrochloric acid, followed by an additional two-washing operation with double-distilled water to remove any remaining acid, flour, dust, or pollutants. The residual rice husk was shade dried at room temperature for a day before being gathered on a clean glass petriplate and dried uniformly in a hot air oven at 105 °C. The cleaned rice husk was then extracted from the liquid using a filter mesh with a cell size of five millimeters.

Prior to producing RHA, rice husk was first leached with 1 N HCl and then heated to 700°C for six hours. At a ratio of 1:10, ten grams of RHA have been mixed in one hundred milliliters of double-distilled water. Using a magnetic stirrer, the mixture was stirred properly for 30 minutes at 500 rpm at room temperature. After that, to ensure that the RHA particles were evenly dispersed throughout the water, the combination was ultrasonically treated using an ultrasonic probe for 15 minutes at a frequency of 24 kHz. It was then mixed with an equivalent volume of regular sodium carbonate, heated for two hours at 90 °C while stirring occasionally, and the resulting solution was cooled to room temperature before being centrifuged three times at 5000 rpm for 10 minutes each to remove the carbonic acid supernatant (H<sub>2</sub>CO<sub>3</sub>). The sample was aged for 24 hours before being repeatedly cleansed with double-distilled water to eliminate the contaminants. The remaining sodium silicate (Na<sub>2</sub>SiO<sub>3</sub>) was brought to neutral pH by adding one drop of regular hydrochloric acid (HCI) at a time to the residue. After that, the sample was dried in a vacuum oven at 650 mm Hg pressure for two hours to eliminate water.

The functional groups of the SiO<sub>2</sub> nanoparticles were analyzed using the Fourier-transform infrared spectroscopy (FT-IR) method. Spectral-grade KBr powder and nanosilica were mixed in an agate mortar at a weight ratio of 2 mg SiO<sub>2</sub> to 200 mg KBr. The powder mixture was shaped into pellets 13 mm diameter and a 0.5 mm thickness. Type FT-IR 6800 The infrared (IR) spectra of nanosilica across the 4000 to 400 cm wavenumber range were measured using spectroscopy. The amorphous phase of nanosilica was identified using the RIGAKU's ULTIMATE-IV X-ray diffractometer (XRD). The XRD pattern was obtained using CuK as the radiation source ( $\lambda$  = 1.5405) while operating at a constant voltage and current of 30 mA at 40 kV with a diffraction angle of 20 scan range of 5 to 80°. To assess the morphology and particle size of the nanosilica, a Tescan Mira3 Field Emission Scanning Electron Microscope (FESEM) was employed. The size of the silica particles were determined from the FEGSEM image using the "Image J" software. The chemical composition of nanosilica was assessed using Energy dispersive X-ray spectroscopy (EDS) with a JEOL 6610 LA model.

# Methodology of seed germination of sweetcorn

To compare the effects of five different concentrations of nano-silica (ns) suspensions (0, 100, 200, 300, 400 and 500 ppm as ns0, ns100, ns200, ns300, ns400 and ns500 respectively) on the germination of sweetcorn seeds, a completely randomized design with three replicates and distilled water was used as a control. Prior to the experiment, 30 sweetcorn seeds per replicate were carefully cleaned with distilled water and then submerged in a 10% hypochlorite solution for 5 minutes. To allow seeds to germinate, seeds were then evenly spread on doubled filter paper in sterile petri dishes with a 15 cm diameter. Before transferring, seeds from each treatment were first immersed in a suspension of nano-silica at the same concentration as that used in the treatment's nano-silica. Each copy received 15 mL of the nano-silica treatment that was tested on it. Throughout the seven days experiment, all petri dishes were kept moist and damp by tightly sealing them to prevent drying. All petri dishes were kept at 25 °C in a dark environment. From the time of germination until seven days later, a daily count of emergent radicles with a minimum length of 2 mm was performed. The following germination indicators were calculated:

Final germination percentage (FGP) was computed as percentage (Ranal and Santana, 2006) using the equation Eq 1:

Number of seeds germinated	(Fa 1)
Total number of seeds subjected to germination	(Eq. 1)

b) Germination speed (GS) was computed as described by (Czabator, 1962) using the equation 2 presented below:

$\overline{}$	Number of seeds germinated on day ti	(Fa 2)
$\square$	number of days during germination period	(Eq. 2)

c) Coefficient of velocity of germination (CVG) was calculated after Jones and Handreck (1967) by the following equation 3:

$\Sigma$ Number of seeds germinated on day ti	(Fa 3)
$\Sigma$ Number of seeds germinated on day ti * number of days * 100	(Eq.5)
during germination period	

 d) Mean germination time (MGT) was calculated by equation 4 cited by Mauromicale and Licandro (2002) given below:

 $\sum \begin{pmatrix} \text{Number of seeds germinated on day ti *} \\ \frac{number of \ days \ during \ germination \ period}{\text{Total number of seeds subjected to germination}} \end{pmatrix}$ (Eq. 4)

Where, ti = (between 0 and 7 days)

e) Vigor index (VI) was calculated using the equation 5 of Amooaghaie *et al.*, (2015), as follows:

# (mean length of the seedling (cm) \* germination percent

f) Germination index (GI) was computed as in the Association of Official Seed Analysts (AOSA, 1983) by following equation 6:

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Number of seedlings emerging on day "d"
(Eq. 6)

Days after planting
(Eq. 6)
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Where, d = days after planting

# Statistical analysis

Data collected from the experiment was analyzed using Microsoft Excel 2010 (mean values and standard deviation), and statistical analysis was performed using the XLSTAT software suite. When there was a significant difference between treatments, the Fisher's test was adopted to perform repeated comparisons. Significant differences were recognized at the p<0.05 criterion. The categorized treatments show how significantly distinct they are from those that do not share a letter label.

# **RESULTS AND DISCUSSION**

#### Nanosilica extracted from rice husk ash

The FTIR spectra depicting the functional groups of nanosilica is shown in Fig. 1, which stated that the bending vibration of the H<sub>2</sub>O molecule in the Si-OH group is responsible for the band position at 1635.03 cm<sup>-1</sup> in the silica sample, the stretching vibration of the O-H group is responsible for the band position at 3495.14 cm<sup>-1</sup> in the RHA-Silica. Si-O-Si is said to have an asymmetric stretching vibration in the 1054.72 cm<sup>-1</sup> band. The symmetric stretching vibration of the Si-O bond produced the wavenumber at 800.97 cm<sup>-1</sup>, while the Si-O bending of the siloxane group for RHA-Silica produced the band at 450.13 cm<sup>-1</sup>. The outcomes closely mirror the findings of Hossain *et al.* (2019), indicating the prevalence of silanol (Si-O-H) and siloxane (Si-O-Si) groups within the range of 1700 to 2750 cm<sup>-1</sup>



**Fig.1.** *FT-IR* spectra depicting the functional groups of nanosilica

in RHA-Silica. This alignment is further corroborated by Costa and Paranhos (2018), who noted a broad absorption band around 3420 cm<sup>-1</sup>, attributing it to the stretching of hydrogen bonds and the bending of hydroxyl (OH) groups linked to the cellulose structure found in the rice husk. Azat *et al.* (2019) also lend support, stating that the broader peaks at 1095, 796, and 471 cm<sup>-1</sup> correspond to the stretching vibrations of the siloxane groups and the distinct absorption peak at 1055 cm<sup>-1</sup> can be attributed to the vibration modes of the siloxane network, indicative of a highly condensed silica network.

Fig. 2 depicts the XRD pattern of RHA-Silica. A large hump at a 20 angle of 22.56 degree was seen in the silica sample, showing the sample's amorphous nature. Nayak and Datta (2021), Ma et al., (2012), Yalc and Sevinc (2001) extracted silica nanoparticles from rice husk, they detected a significant peak orientated at a 20 angle of 22.259° and 22° respectively. Other scanning angles ranging from 10° to 80° revealed no strong peaks, indicating that no specific structure was present in the crystalline form. Furthermore, the Gaussian peak fitting of the Bragg diffraction peaks was performed. After fitting Gaussian functions to the whole Bragg diffraction peak collection, broader de-convoluted peaks were obtained (Fig. 2). This analysis showed that the sample contained typical amorphous elements despite the absence of any structured crystalline phases. Sampath et al. (2012) discovered that a broader Gaussian peak signals the presence of an amorphous constituent in an X-ray diffraction examination of amorphous and nanocrystalline structures.

The Field emission scanning electron microscopy (FESEM) images of  $SiO_2$ -NPs sample using FESEM micrograph are shown in Fig. 3. The spongy structure of synthesised nanosilica was demonstrated using FESEM with high enlargement, and the particle size



**Fig. 2.** XRD spectra of nanosilica extracted from rice husk ash

and shape of synthesised silica were examined.  $SiO_2$ -NPs were round in shape and had particles ranging in size from 19.79 to 36.51 nm. Agglomeration and spherical SiO<sub>2</sub>-NPs were seen in RHA-Silica. According to Hossain *et al.* (2019), strong cohesive intramolecular forces may dominate the aggregation process rather than gravity force.

The results of the elemental composition analysis of the rice husk sample that was extracted using EDS research, which were represented graphically by EDS peaks in Fig. 4. Only two distinct peaks of O K and Si K as well as tiny peaks of Na K, Mg K, and Fe K could be found in the RHA-Silica sample. The extracted silica sample is 96.87% pure, according the EDS analysis. The results align with prior research on mesoporous nano-silica in Pumice, a silica-rich rock utilized as a raw material, as demonstrated by Mourhly *et al.* (2019).

# Seed germination indices

The variances for the growth characteristics of sweetcorn seeds under the effect of six nano-silica levels are presented in Figs 5-10. The results demonstrated significant variations in final germination percentage (FGP), germination speed (GS), coefficient of velocity of germination (CVG), mean germination time (MGT), vigour index (VI), and germination index (GI) related to the application of nano-silica at p<0.05. These results are consistent with findings of Sun et al. (2021), indicating that an appropriate concentration of silicon (Si) can aid in breaking dormancy in maize seeds, accelerating seed vigour restoration, and facilitating recovery from damage during seed drying. Additionally, Si is crucial in supporting complete germination and emergence processes while providing favourable physiological conditions for subsequent seedling growth. Similarly, Sabaghnia and Janmohammadi (2014) found that applying 1 mM nano-silicon dioxide positively influenced root and shoot length, seedling weight, mean germination time, and seedling vigour index in lentil seeds.

#### Germination percentage

After being treated with various nano-silica concentrations, the data in Fig. 5 indicates the computed values of sweetcorn seeds' final germination percentage (FGP,%). Significantly higher FGP was obtained with all nano-silica treatments than with nano-silica-free seeds (control). The control seeds germinated with 69.8% after 7 days, treatments with 300 and 400 ppm nano-silica doses achieved 97.8% and 95.3% FGP. The respective FGP of seeds exposed to 100 and 500 ppm nano-silica was 78.6% and 82.6%. Compared to untreated seedlings, the rate of 300 ppm improved ultimate germination by 40.1%. Crop productivity is determined by various genetic and environmental factors, with seed germination regarded as the fundamental process that ensures and determines if subsequent agricultural practices and ideal production will attain the desired yield (Bhattacharjee, 2008; Al-Mudaris, 1998). The recently acquired results are consistent with those reported by Lu et al. (2015), who stated that FGP of tomato increased, by 70.3% after treatment of nanosilica suspension with 5 g L<sup>-1</sup> against control seeds. Similarly, after treating maize seeds with nano-silica, seed germination increased by 2-11% (Yuvakkumar et al., 2011). Jiang et al. (2022) and Nair et al. (2011) utilized nano-silica to promote rice seed germination, and their findings indicate that Si-based materials exhibit varying effects on germination depending on concentration levels. Specifically, the range of 10-200 mg L<sup>-1</sup> of nanosilica (nSiO<sub>2</sub>) was found to support rice seed germination without adverse consequences, potentially due to the enhancement of water permeability in seed coats, cell walls, or organelle membranes by these nanomaterials.



Fig. 3. FESEM image of nanosilica using MIRA3 TESCAN

#### Rate of germination

The dynamic of germination speed is described in detail by germination speed (GS, %/day), which indicates the number of seeds germinating each day. Fig. 6 displays the values of GS. The daily germination rate of control sweetcorn seeds was 23.6%, but treated seeds with nano-silica had a modest reported value of 32.3% in the case of treatment with 100 ppm nano-silica. But when seeds were treated with 300 ppm nano-silica, the highest GS (39.6%) against control treatment was achieved. These findings showed that nano-silica significantly and favorably induced sweetcorn seed germination compared to control seeds. The present results are supported by Sun et al., (2021) and Khalaki, et al. (2016), who reported that GS of Zea mays and Thymus kotschyanus was improved by the application of nanosilica under in vitro conditions respectively.

# Coefficient of velocity of germination

The Coefficient of Velocity of Germination (CVG) measures how quickly seeds germinate. The quantity of seeds that germinate determines its value and the quantity of seeds that germinate increases as CVG and germination time decrease. Theoretically, CVG can have a value of 100 (Al-Mudaris, 1998). Fig. 7 illustrates the considerable variations in CVG values between control sweetcorn seeds and seeds treated with different concentrations of nano-silica in the current experiment. The CVG of untreated seeds was 26.4, while that of treated seeds with various doses of nanosilica was greater. The most notable CVG reported was 38.3, which was 45.07% higher than the control treatment when 300 ppm of nano-silica was used. However, the CVG escalated compared to the control recording 34.3 and 32.6 at 400 and 500 ppm of nano-silica, respectively. Since the optimum value of CVG is 100, higher CVG values indicate that the treatments employed have a positive effect.



Fig.4. Elemental composition of nanosilica through EDS

#### Mean germination time

Mean germination time (MGT) refers to how long a seed takes to begin and finish germinating. MGT is expressed as a unit of day or, more simply, as the total number of days required for seed germination. Shorter germination times are preferable to longer ones, therefore any treatment that shortens the MGT will be significant and impact on the vigor of seedlings and, ultimately, productivity. The data of MGT of sweetcorn seeds during the germination experiment are shown in Fig 8. When substantial variations were assessed, the application of nanosilica was found to have a significant impact on the MGT of seeds. Treatment with 300 ppm nano-silica reduced the time needed for full germination from 5.6 days for control seedlings to 3.3 days. Furthermore, compared to control seeds, other nano-silica treatments at 100, 200, 400, and 500 ppm were observed to lower MGT. The results of the present study support the experiment conducted by Alsaeedi et al. (2019), which stated that the application of nano-silica demonstrated a significant effect on the mean germination time of Cucumber seeds, leading to notable differences between the control and treated groups. Control seeds exhibited a mean germination time of approximately 4.1 days, while seeds treated with 200 mg L-1



**Fig. 5.** Effect of different concentrations of nanosilica on germination percentage of sweetcorn





nano-silica showed a substantially reduced MGT of only 2.8 days. Similar to this, Suriyaprabha *et al.* (2012) discovered that nano-silica positively decreased the MGT of maize seeds during germination, while Na<sub>2</sub>SiO<sub>3</sub> extended the MGT and micro-SiO<sub>2</sub> and H<sub>4</sub>SiO<sub>4</sub> had no impact. These results further support an experiment conducted by Azimi *et al.* (2014), who showed that add-ing nano-silica by 40 mg L<sup>-1</sup> reduced the MGT of tall wheatgrass seeds to the quickest germination period observed.

# Seed vigour index

Fig. 9 shows the results of the seed vigour index (SVI) of sweetcorn seeds treated with and without nanosilica. The stronger the seedling, the higher the vigor index. It is evident that germinating sweetcorn seeds in nano-silica produced more robust seedlings than the control. In comparison to control seeds, all nano silica treatments showed higher VI. The seeds that encountered 300 ppm of nano-silica, on parity to 400 ppm, had the highest measured (SVI) of 1623. However, it was discovered that VI of 934, 1119, and 1380 were considerably better than control seeds at 100, 200, and 500 ppm of nano-silica, respectively. The ability of SVI to predict and specify whether seedlings can continue



**Fig. 6.** Effect of different concentrations of nanosilica on germination speed of sweetcorn



**Fig. 8.** Effect of different concentrations of nanosilica on mean germination time (MGT) on sweetcorn





**Fig. 9.** Effect of different concentrations of nanosilica on seed vigour index of sweetcorn

growing well after germination makes VI an essential characteristic of seed germination. According to Alsaeedi *et al.* (2019), the inclusion of 200 mg L<sup>-1</sup> nanosilica resulted in a significant 40% enhancement in the vigour index (VI) of Cucumber seeds compared to the untreated control seeds and Azimi *et al.* (2014) stated that adding nano-silica by 40 mg L<sup>-1</sup> improved VI of tall wheatgrass seeds by 120% compared to control seeds, confirm present findings. Similar findings were made by Lu *et al.* (2015), who discovered that providing tomato seeds 7 g L<sup>-1</sup> of nano-silica boosted the SVI.

# **Germination index**

The germination index (GI) depends on the germination and percentage. Low values indicate that most seeds germinate late, whereas high values indicate that majority seeds germinate early in a short time (Al-Mudaris, 1998). The results revealed that nano-silica with a concentration of 300 ppm in the germination medium boosted GI to reach 19.5 as the highest recorded value among all treatments (Fig. 10). The absence of nanosilica from the germination medium as in the control lowered GI values recorded 9.9. All nano-silica treatments, however, had higher and more significant GI than the control group. GI was 11.2, 15.4, 19 and 13.7 for 100, 200, 400 and 500 ppm nano-silica treatments, Alsaeedi et al. (2019), reported comparable findings with Cucumber seeds, noting that the inclusion of nanosilica at a concentration of 200 mg L<sup>-1</sup> in the germination medium led to a significant rise in germination index (GI) by 68.75%, surpassing the control group where nano-silica was not present in the germination medium.

# Conclusion

The present study demonstrated significance of nanosilica for sweetcorn seed germination. For all germination measures and indices, all seeds treated with nanosilica had better and greater outcomes. Regarding all



Fig. 10. Effect of different concentrations of nanosilica on germination index of sweetcorn

germination indices, treatment with 300 ppm of nanosilica outperformed 100, 200, 400, and 500 ppm treatments. Applying 300 ppm of nano-silica resulted in the highest final germination percentage, germination speed, coefficient of germination velocity, vigour index, germination index and lowest mean germination time. Based on the findings, it was concluded that using 300 ppm nano-silica is critical for sweetcorn germination and growth to enhance productivity. This desirable impact of nano-silica may be attributable to its small size, which allows it to swiftly penetrate the cell wall and trigger several physiochemical processes that speed up germination and growth.

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#### **Conflict of interest**

The authors declare that they have no conflict of interest.

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