Effects of Organic Fertilizer Mixed with Food Waste Dry Powder on the Growth of Chinese Cabbage Seedlings

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Abstract: Food waste is a common global threat to the environment, agriculture, and society. In the present study, we used 30% food waste, mixed with 70% bio-fertilizers, and evaluated their ability to affect the growth of Chinese cabbage. The experiment was conducted using different concentrations of food waste to investigate their effect on Chinese cabbage growth, chlorophyll content, and mineral content. Leaf length, root length, and fresh and dry weight were significantly increased in plants treated with control fertilizer (CF) and fertilizer mixed with food waste (MF). However, high concentrations of food waste decreased the growth and biomass of Chinese cabbage due to salt content. Furthermore, higher chlorophyll content, transpiration efficiency, and photosynthetic rate were observed in CF- and MF-treated plants, while higher chlorophyll fluorescence was observed in the MF × 2 and MF × 6 treatments. Inductively coupled plasma mass spectrometry (ICP-MS) results showed an increase in potassium (K), calcium (Ca), phosphorous (P), and magnesium (Mg) contents in the MF and MF × 2 treatments, while higher sodium (Na) content was observed in the MF × 4 and MF × 6 treatments due to the high salt content found in food waste. The analysis of abscisic acid (ABA) showed that increasing amounts of food waste increase the endogenous ABA content, compromising the survival of plants. In conclusion, optimal amounts of food waste—up to MF and MF × 2—increase plant growth and provide an ecofriendly approach to be employed in the agriculture production system.

Keywords: Brassica rapa; food waste; organic fertilizer; mineral content; abscisic acid

1. Introduction

It is estimated that about one-third to one-quarter of food is wasted. Food waste is also defined as food loss or un consumed food [1], and it represents the portion of food that is not eaten by anyone [1]. There are various causes of food waste, which may occur at any point of the food chain system, for example during production, processing, distribution, consumption, or at the retail stage. It is estimated that 30–40% of food is wasted within the food chain [2,3]. Kumar et al. [4] reported that 42% of waste derives from households, 39% from food industries, and 5% occurs during distribution. In order to achieve development goals for a sustainable environment, it is necessary to minimize these quantities. Food waste has a worse effect on the environment and agriculture industry because composting is not carried out properly [5]. Globally, 1.3 billion tons of food is wasted every year [6], an amount that is equivalent to more than half of the world’s annual cereal crop production (2.3 billion tons). Approximately 19 million tons of food waste is reported annually in Japan [7], 16.5 million tons in Taiwan [8] and 4.3 million tons in Korea [9]. According to the World Economic Forum, in South Korea 95% of the food waste is recycled in the effort to become a zero waste society [10,11].
Various strategies can be adopted to minimize wastage [12,13]. Nowadays, the biggest challenges in food waste recovery are animal feeding, reducing the volume of surplus food generation, combating world hunger, providing food waste to rendering industries, and using it for soil amendments or incineration. In recent years, anaerobic digestion (AD) technology for FW resource treatment has attracted considerable attention due to its advantages, as it allows for the obtaining of clean energy and low carbon emissions, and can be used in fertilization programs [14,15]. After the AD processes, the biogas obtained can be upgraded to bio-methane, which is considered an attractive renewable energy, as it is generally used as fuel in the transport sector, or is injected into gas grids, saving tons of CO₂-equivalent emissions [14,15]. On the other hand, AD technology allows for the recycling of nutrients when digestates are applied on agricultural lands, either used as food waste powder directly or mixed with other fertilizers [14,16]. For instance, anaerobic digestion for biogas production results in large amounts of liquid digestate, which contains high amounts of nutrients, such as nitrogen, potassium, and phosphorus, as well as micronutrients in plant-available forms [17]. The long-term application of food waste organic fertilizer was shown to improve soil quality, stimulate crop yields, and even have a positive influence on the growth of soil bacteria [17]. Furthermore, food waste can be used as organic fertilizer by composting natural biological degradation processes [2,18,19]. In accordance with the establishment of standards for fertilization processes and amendments to the designation notice, dry waste powder can be used as raw material for organic fertilizers [20,21]. In this case, it must include the standard values of less than 2% salt, less than 15% moisture, and less than 30% of all raw materials [20,22]. Soil salinity is a major abiotic stress that limits plant growth and development by affecting various physiological and biochemical processes. Previous reports have shown that there are many effective ways of improving salt-affected lands, such as water leaching, phytoremediation, and chemical remediation [23]. In particular, the remediation of salt affected soils through chemical agents includes the use of gypsum, organic matter (such as farmyard manure), green manure, or organic food waste [24]. It was also reported that the application of food waste improves the physical, chemical, and biological properties of soil and enhances plant growth and development in various crops such as rice [23], tomato [25], pakchoi [26], and common bean [27].

As it contains salt (and salinity is the biggest obstacle to food waste fertilization), food waste cannot be used directly as fertilizer, and therefore a method that includes mixed fertilization is being proposed as an alternative [22,28]. It is urgent to prepare a plan to effectively utilize food waste as raw material for fertilizers. This research contributes to the development of technology aimed at improving the stability and homogenization of dry powder to be used as a substitute for mixed organic fertilizers [29,30]. When food waste is used as raw material for fertilization, various problems—such as excessive salt content, mixing of impurities, and odors—arise [31–33]. Therefore, there is an urgent need to ensure stability and verify the procedures that are essential for safe fertilization processes. In present study, food waste was mixed with organic fertilizer in dry powder form, and its effects on Chinese cabbage seedlings and on the soil environment were analyzed [34,35]. Furthermore, the combined effect of food waste and organic fertilizer on the growth (shoot/root), biomass (fresh/dry weight), and chlorophyll content were also investigated, together with the effect on phytohormonal regulation and content of minerals, such as N, P, and K.

2. Materials and Methods

2.1. Physicochemical Properties of Soil

For the analysis of physicochemical properties, we collected soil samples and sieved them to remove impurities. The soil was air dried under normal temperature and sieved through a 2 mm mesh for further analysis. A quantity of 10 g of dry soil was mixed with 50 mL of water (1:5) by vortexing every 10 min for 1 min. After 1 h, the pH and electrical conductivity were measured through a pH meter and EC meter, respectively. Subsequently,
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Organic matter was quantified using the Mebius method. A quantity of 0.1–0.5 air-dried soil (0.5 mm) was mixed in 10 mL of 0.4 N dichromate solution, and 20 mL of concentrated sulfuric acid was added to a 250 mL Erlenmeyer flask and heated on an electric plate at 100 °C for 5 min. After cooling, 200 mL of distilled water, 10 mL of concentrated phosphoric acid, and 1 mL of diphenylamine (16%) were added [36].

2.2. Measurement of Available Phosphorous, Nitrate, and Cation Exchange Capacity

For the analysis of phosphorous content, we used the molybdenum blue method with 1-amino-2-naphtol-4-sulfonic acid, on the basis of the leaching method described in detail by Lancaster [37,38]. In this method, the modified molybdenum is used to determine the quantity of enzymatically hydrolyzable phosphorus in the soil. 1-Amino-2-naphtol-4-sulfonic acid by Lancaster leaching method was followed for available phosphorus estimation. For the analysis of nitrogen content, the method of Calazans et al. [39] was adopted. The NH$_4^+$ adsorbed on soil particles was replaced with potassium chloride solution, alkalinized with magnesium oxide and titrated to measure ammonia nitrogen. Then, Devarda’s alloy was added to convert NO$_3^-$ to NH$_4^+$ for distillation titration, and nitrate nitrogen was measured. A quantity of 10 g of air-dried soil (2 mm) was mixed with 0.1 g of a mixture containing sulfate—copper sulfate and selenium (10:10:1)—and 3 mL of concentrated sulfuric acid in a 100 mL polyethylene Erlenmeyer flask; then, the contents were shaken for 30 min. The mixture inside the glass tube was digested in a sealed vial and was heated until it turned white. Toward the end of the digestion process, 10 mL of distilled water was added, and the extract was distilled in the presence of sodium hydroxide (13 mol/L) in a Kjeldahl distiller. The extract was placed into a boric acid solution until a total of 50 mL of condensate in the flasks was obtained. The distillate samples were titrated with 0.01 N H$_2$SO$_4$, and the total nitrogen content in soil samples was calculated.

For the analysis of cation exchange capacity, 10 g of soil dried at 40 °C was weighted and put in centrifuged tubes, and 50 mL of 1 N NH$_4$OAc (pH 7) solution was added. The tubes were then kept in a shaking incubator for 3 h at 140 rpm and centrifuged at 600 rpm for 10 min. A 5.5 cm Buchner funnel with retentive filter paper was fit, the paper was moistened, and the soil extract was transferred. The decanted liquid was collected in a 100 mL volumetric flask and was added with 1 N NH$_4$OAc solution until reaching a volume of 100 mL. The exchangeable bases (K, Na, Mg$_2$, and Ca$_2$) were determined through the ICP measurement method.

3. Experimental Set Up

The Chinese cabbage needed for the experiment was purchased from Seoul-baechu, (Danong, Gyeonggi province, South Korea), and seedlings were grown for 3 weeks in a 72 Cells Seedling Trays (11 × 21.25 × 1.8 inches) containing horticultural soil (peat moss (10–15%), coco peat (45–50%), zeolite (6–8%), and perlite (35–40%), along with NH$^+$ (≈0.09 mg/g), NO$_3$ (≈0.205 mg/g), PO (≈0.35 mg/g), and KO (≈0.1 mg/g)) purchased from (Shinsung Mineral Co., Ltd., Goesan, Korea) [40,41]. The organic fertilizer mixed with food (MF) waste was incorporated into the upper 10 cm soil layer, and supplemented with various amounts of food waste, on the basis of the experimental design. The pots (17.0 × 12.0 × 13 cm) were labeled properly and divided into six groups (A) NT: not treated control; (B) CF: controlled fertilizers (3.78 g per pot), which were added only with high quality uniform compound fertilizer from a registered company (poweralchandeul, Farmhannong Co., Ltd., Yeongdeungpo-gu, Seoul, Korea, N-P2O5-K2O: 12-6-6); (C) MF: mixed fertilizer (10.61 g per pot) composed of castor oil cake/rapeseed oil/food waste powder (49:21:30), wherein a sub-group (MF × 2) was set up where the quantity of mixed fertilizer was double; (D) the amount of mixed fertilizer was increased four times; and (E) the amount of mixed fertilizer was increased six times.
4. Morphological Analysis

After 4 weeks of growth using organic fertilizers mixed with different concentrations of food waste, growth parameters, namely, root length, shoot length, fresh and dry weight, were measured. Furthermore, a LCpro T portable photosynthetic assay system (ADC Bioscientific Ltd., Herts, England) was used for the quantification of transpiration rate, stomatal conductance, and photosynthetic rate.

5. Endogenous Abscisic Acid (ABA) Quantification

For the quantification of endogenous ABA, we used the detailed method by Khan et al. [42,43]. A quantity of 0.3 g of freeze-dried sample was treated with 30 mL of extract solution (isopropanol, 95%, and glacial acetic acid, 10%), and 100 ng of ABA standard [(±)-3,5,5,7,7,7-d6] was added. The suspension was filtered, and the filtrate was concentrated using a rotary evaporator. The residue was suspended in 4 mL of 1 N NaOH solution and rinsed three times with methylene chloride (3 mL) in order to eliminate traces of lipophilic material. After decreasing the pH of the aqueous phase to 3.5 by adding 6 N HCl, we extracted it through solvent extraction with ethyl acetate three times. The ethyl acetate extract was then evaporated and the dry residue was re-suspended in phosphate buffer solution (pH 8), which was passed through a polyvinylpolypyrrolidone column. The eluted phosphate buffer solution was once again partitioned three times with EtOAc, after adjusting to pH 3.5 with 6 N HCl. All three aliquots extracted were pooled and evaporated using the rotary evaporator. Subsequently, the fractions were methylated with diazomethane to detect and quantify ABA using GC-MS/SIM equipment (6890N network gas chromatograph, Agilent Technologies) (Scheme 1). ThermoQuest Crop. (Manchester, UK) software was used to monitor signal ions (m/z 1162 and 190 for Me-ABA, and m/z 166 and 194 for Me-[2H6]-ABA).

| Equipment                  | Hewlett-Packard 6890, 5973N Mass Selective Detector |
|----------------------------|------------------------------------------------------|
| Column                     | HP-1 capillary column (30 m x 0.25 mm i.d. 0.25 μm film thickness) (J & W Scientific Co., Folsom, CA, USA) |
| Carrier gas                | He (40 mL/min.); head pressure of 30 kPa              |
| Source temp.               | 250 °C                                               |
| Oven conditions            | ABA: 60 °C (1 min.) 15 °C/min; 200 °C, 5 °C/min; 250 °C, 10 °C/min; 280 °C |
| Injector temp.             | 200 °C                                               |
| Ionizing voltage           | 70 ev                                                |

Scheme 1. GC/MS–SIM conditions used for analysis and quantification of the ABA.

6. ICP Analysis of the Uptake of Different Elements

For the quantification of different elements absorbed, such as sodium (Na), potassium (K), calcium (Ca), phosphorous (P), and magnesium (Mg), we followed the methods described in Khan et al. [44] and Sahile et al. [45], using 200 mg of freeze-dried powder. All the samples were digested with 5 mL of HNO3 and 3 mL of H2O2 in a microwave oven. Subsequently, 3% of HNO3 was added to digest the samples and was injected in an inductively coupled plasm mass spectrometry analyzer (Optima 7900DV, Perkin-Elmer, Waltham, MA, USA).

7. Statistical Analysis

The results of the current study were performed in a completely randomized design and were subjected to statistical analysis. The experiments were conducted as three parallels, and every replicate included 20 plants. Graphs were generated using Graph Pad Prism software (Version 6.01, San Diego, CA, USA), whereas means with standard error were compared using Duncan’s multiple range test (DMRT) in SAS (V9.1, Cary, NC, USA).
8. Results and Discussion

8.1. Physicochemical Properties of Soil

It is very important to monitor the physicochemical characteristics of soil because soil is a complex material that consists of various components, such as minerals, moisture, and organic matter [46]. These components greatly influence soil structure, texture, and porosity. Before the experiment, the electrical conductivity; organic matter content; and available phosphorus, nitrogen, calcium, potassium, magnesium, and sodium were analyzed (Table 1). Soil pH and electrical conductivity were 6.6 and 2.8 dS, respectively. Soil pH, whose optimal range is 5–7 for agricultural crops [47], plays a vital role in the regulation of nutrient availability [48], and is an indicator of the overall chemical status of soil [49]. Electrical conductivity is also an important indicator of soil health that affects plant nutrient availability and crop yield [50]. Moreover, soil is a major source of nutrients, and soil nutrient availability is essential in sustaining soil quality and plant productivity [51,52]. The nutrient contents obtained from soil analysis are the following: organic carbon, 20 g/kg; nitrate nitrogen, 141.8 mg/kg; available phosphorus, 330 mg/kg; K, 1.11 mg/kg; Mg, 3.65 mg/kg; and Na 0.6 mg/kg, as shown in Table 1. Crops plants need 16 essential elements that are vital for their normal growth [53]. The over-application of fertilizers, using water irrigation, can cause a micronutrient imbalance in soil and nutrients available to plants. Among the nutrients, organic matter increases the chemical and physical properties of soil that contribute to plant growth and development [54].

Table 1. Physicochemical properties of the soil used in the experiment.

| pH [1:5] | EC [1:5] (dS/m) | OM (g/kg) | NO₃-N (mg/kg) | AP (mg/kg) | K (cmol/kg) | Ca (cmol/kg) | Mg (cmol/kg) | Na (cmol/kg) |
|----------|-----------------|-----------|---------------|------------|-------------|--------------|--------------|--------------|
| 6.6      | 2.8             | 20        | 141.8         | 330        | 1.11        | 11.61        | 3.65         | 0.6          |

EC: electrical conductivity, AP: available phosphorus, OM: organic matter.

8.2. Effect of Food Waste on the Growth, Biomass, and Chlorophyll Content of Chinese Cabbage

Currently, food waste is one of the most urgent challenges facing the environment, society, and the economy because of the devastating effects it produces [55]. Different strategies are employed to manage/recycle wasted food, such as the use as fertilizer, and feeding [30]. However, using food waste as fertilizer presents a problem, namely, salinity stress. The excess sodium concentration of food waste may cause ionic imbalance, as it produces several morphological and physiological changes that inhibit crop growth and development [56–58]. However, optimal doses of food waste have been demonstrated to play a significant role in fertilization, as shown in Tables 2 and 3. In the current experiment, the growth characteristics of Chinese cabbage were investigated. After four weeks, the CF- and MF-treated plants showed an increase in leaf length (12.6–12.9 cm), root length (11.8–15.3 cm), fresh weight (14.7–16.5 g), and dry weight (3.4–3.9 g) compared to the control NT plants, where the same parameters were lower: leaf length (11.8 cm), root length (9.3 cm), fresh weight (13.4 g), and dry weight (2.5 g). However, leaf length decreased in the MF × 2-, MF × 4-, and MF × 6-treated plants (Table 2). In Table 3, results show that chlorophyll fluorescence, which measures the photosynthetic rate, was higher in the MF × 2- and MF × 6-treated groups than in the other treated groups (CF, MF, and MF × 4), and ranged between 0.76 and 0.79, which satisfactorily meets the needs of plants. In contrast, the transpiration rate decreased in the MF × 2- and MF × 6-treated plants (Table 3). The level of stomatal conductance, which measures the water relationships within a plant, decreased to 0.03 ± 0.0 mol/m² in the treated groups (except for CF), showing that the treated plants are not able to withstand drought or salinity stress. Similar results were also observed for the photosynthetic rates between treated and non-treated Chinese cabbage plants (Table 3).
Table 2. Growth characteristics of Chinese cabbage.

| Fertilizations | NT  | CF  | MF  | MF × 2 | MF × 4 | MF × 6 |
|----------------|-----|-----|-----|--------|--------|--------|
| Leaf Length (cm) | 11.8 ± 0.87 ab | 12.9 ± 2.19 a | 12.6 ± 0.95 a | 11.7 ± 0.76 ab | 10.9 ± 0.32 ab | 8.2 ± 0.46 b |
| Root Length (cm)  | 9.3 ± 0.77 a  | 11.8 ± 2.20 a  | 15.3 ± 7.98 a  | 11.1 ± 0.13 a  | 11.7 ± 2.39 a  | 9.8 ± 1.07 a  |
| Fresh Weight (g)  | 13.4 ± 4.33 ab | 14.7 ± 4.62 a  | 16.5 ± 3.69 ab | 9.5 ± 1.39 ab  | 7.9 ± 1.81 ab  | 3.3 ± 0.29 ab  |
| Dry Weight (g)    | 2.5 ± 0.48 bc | 3.9 ± 0.61 a  | 3.4 ± 0.13 ab  | 2.6 ± 0.39 bc  | 2.0 ± 0.35 cd  | 1.2 ± 0.29 d  |

NT: not treated; CF: controlled fertilizer; MF: castor oilcakes 49, rapeseed oil cake 21, food waste powder 30; MF × 2 (mixed expeller cake and food waste powder); MF × 4 (mixed expeller cake and food waste powder); MF × 6 (mixed expeller cake and food waste powder). Each value represents mean + SD of three replicates. Values with different letters in rows are significantly different from each other, as evaluated by DMRT.

Table 3. Photosynthetic characteristics of Chinese cabbage.

| Fertilizations | NT  | CF  | MF  | MF × 2 | MF × 4 | MF × 6 |
|----------------|-----|-----|-----|--------|--------|--------|
| Chlorophyll Contents (mg/m²) | 487.7 ± 27.28 b | 521.7 ± 13.13 a | 477.3 ± 34.89 b | 464.3 ± 38.52 b | 439.3 ± 40.91 c | 427.7 ± 37.53 c |
| Chlorophyll Fluorescence (Fv/Fm) | 0.80 ± 0.011 a | 0.79 ± 0.023 a | 0.79 ± 0.010 a | 0.81 ± 0.039 a | 0.76 ± 0.038 a | 0.82 ± 0.023 a |
| Transpiration Efficiencies (mmol/m²) | 2.0 ± 0.35 ab | 2.3 ± 0.31 a | 2.0 ± 0.37 ab | 1.6 ± 0.27 ab | 1.5 ± 0.24 ab | 1.3 ± 0.00 b |
| Stomatal Conductance (mol/m² s) | 0.08 ± 0.020 a | 0.09 ± 0.015 a | 0.07 ± 0.013 ab | 0.05 ± 0.007 ab | 0.04 ± 0.007 b | 0.03 ± 0.000 b |
| Photosynthetic Rate (µmol/m²/s) | 2.8 ± 0.04 a | 2.9 ± 1.14 a | 2.3 ± 0.81 ab | 2.0 ± 0.98 ab | 1.2 ± 0.33 ab | 0.2 ± 0.16 b |

Each value represents mean ± SD of three replicates. Values with different letters in rows are significantly different from each other, as evaluated by DMRT.

8.3. Effect of Food Waste on ABA and Mineral Uptake in Chinese Cabbage

Phyto-hormones, especially ABA, play a crucial role in plant responses to various environmental conditions [39,60]. In the present study, higher ABA contents were observed in the MF × 2- and MF × 4-treated plants than in NT and CF-treated plants (Figure 1). In contrast, a decrease in ABA content was observed in MF × 6-treated plants, possibly due to an overdose of food waste that led to the death of plants, a hypothesis that is also supported by the observed growth parameters. The increase in ABA content in MF × 2 and MF × 4-treated plants might have been due to the double and quadruple increase in the salt content of food waste. This result also supports the observation [61–63] that high salinity in maize, rice, and soybean enhances ABA contents. Furthermore, this increase in ABA content was also correlated with the ICP analysis of Na content in Chinese cabbage (Table 4). The concentrations of potassium, calcium, phosphorus, and magnesium were higher in the MF and MF × 2 groups than in the MF × 4 and MF × 6 groups. The amount of sodium increases with increasing concentrations of food waste, which is alarming, and induces salinity stress. Excess sodium ions cause ionic imbalance, as they compete with potassium ions, possibly resulting in necrosis and chlorosis [63]. Higher contents of K were observed in the MF- and MF × 2-treated plants, with significant effects on plant growth and photosynthetic rates. Previous studies showed that K plays a vital role in photosynthesis and in the translocation of nutrients in plants; it is considered a major nutrient, essential for plant growth and development [64]. Similarly, higher Mg concentrations also play an essential role in plant photosynthesis and growth, as this element is the central atom amid four nitrogen atoms in the chlorophyll molecule [65]. Another micronutrient is calcium (Ca), which provides structural support to the cell wall and acts as a secondary messenger when plants are stressed [66].
In summary, the combined application of organic and mineral fertilizers is the best approach to achieve multiple aims in terms of high yields, low-cost farming, and minimal negative environmental impacts. The good performance of this combination, together with the reduced expenses for mineral fertilizers, can encourage farmers to accept the use of organic fertilizers. Nabel et al. [67] reported that organic fertilization with digestates had a positive influence on soil properties (e.g., increased soil respiration and enhanced water-holding capacity), particularly in marginal sites. Organic fertilization via food waste would improve crop yields and soil fertility and should be considered as an effective strategy to manage compostable wastes. Mu et al. [68] reported the impacts of different compost treatments rates (10%, 30%, 50%, and 70% v/v), and synthetic fertilizers showed a positive effect on soil fertility, plant yield, and plant nutrient content in arugula and radish plants. Similarly, Dlamlnl et al. [69] also investigated the effect of organic food waste on soil conditions and on the yield of vegetables, and concluded that food waste fertilizers can be used as an alternative to synthetic fertilizers to increase crop yield and improve the physical properties of soil. At the same time, the negative impact of synthetic fertilizers on the environment would be reduced, as well as the impact of disposing of vegetable food waste in landfills.

9. Conclusions

In this study, Chinese cabbage was treated with different concentrations of food waste along with a 70% fertilizer component. An increase in Chinese cabbage growth parameters was observed up to the MF × 2 treatment. However, when the dose of food waste was further increased to four- and sixfold concentrations, plants showed inhibitory growth regulation. We demonstrated that the fertilization of seedlings with food waste within the
optimal MF to MF × 2 range provides a valuable, ecofriendly, and low-cost biotechnological approach for the improvement of sustainable crop production. Further on-field studies are needed to observe the effect of food waste on the soil micro- and macronutrient composition, as well as on the fertility rate in the long term. In addition, as the release of Na⁺ increases soil salinity, it is necessary to determine the proper application doses of these organic by-products in order to avoid negative impacts on soil, the environment, and human health. Moreover, different strategies are needed to avoid the negative impact of sodium on the growth of crops, as food waste contains more salt.

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