Life Table of *Rhodobium porosum* on Rose Cultivars in Subsurface 'Ebb and Flow' Irrigation System

Guilherme S. Avellar¹, Wellington G. Campos¹, Livia M. Carvalho²*, Elka F. A. Almeida³ and Márcia N. O. Ribeiro⁴

¹Department of Biosystems Engineering, Federal University of São João del Rei (UFSJ), São João del Rei, MG, Brazil.
²Agricultural Research Institute of Minas Gerais (EPAMIG), São João del-Rei, MG, Brazil.
³Institute of Agrarian Sciences, Federal University of Minas Gerais (UFMG), Montes Claros, MG, Brazil.
⁴Natural Science Department, Hostos Community College (CUNY HCC), Bronx, NY, US.

Authors’ contributions

This work was carried out in collaboration among all authors. Authors GSA, WGC, LMC and EFAA designed the study, performed the statistical analysis, and wrote the protocol. Authors EFAA and MNOR managed the literature searches. Author GSA and LMC wrote the first draft of the manuscript. Authors WGC and LMC handled different versions of the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Fertilization is a limiting factor in plant development and indirectly affects the population density of phytophagous insects. This study tested the performance of the aphid *Rhodobium porosum* (Hemiptera: Aphididae) on three rose cultivars exposed to two nutrient solutions in subsurface 'ebb and flow' irrigation systems in greenhouse. The performance of *R. porosum* was determined by means of the life table and fertility. Both cultivar and fertilization affected many life history and population traits of the aphid. The lower insect fertility and survival were observed in the nutrient solution usually recommended for fruit-type vegetables and on the 'Tineke' rose cultivar, because this combination caused lower net reproductive rate R₀ (0.52 female), finite rate of increase λ (0.9469 female/day), innate capacity to increase in number rₘ (-0.0516 female/female/day), as well

*Corresponding author: E-mail: livia@epamig.br;
as longer time interval between each generation T (16.62 days). The lowest performance of the \textit{R. porosum} on "Tineke" rose cultivated in the nutrient solution 1 suggests that this seems to be the best combination to reduce problems with aphid attack in soilless rose cultivation.

Keywords: Floriculture; fertirrigation; insect control; \textit{Rhodobium porosum}; \textit{Rosa sp.}

\section*{1. INTRODUCTION}

The rose is the most produced and commercialized cut flower in the world. It is a very demanding crop in terms of fertilization because constant harvest of its floral stems requires frequent replacement of nutrients \cite{1,2}. The amount of water and nutrients to be supplied to plants by means of fertirrigation, in a protected environment, has been object of research due to its economic and environmental importance \cite{2,3,4}. The subsurface irrigation system, also known worldwide as 'ebb and flow' or 'ebb and flood', is a type of irrigation in which plants are arranged on benches or floors that are periodically filled with nutrient solution and, after a specific period, the nutrient solution is drained and stored in tanks for recycling and reuse \cite{4,5,6}.

Soilless rose cultivation in a semi-hydroponic system with subsurface flooding is an alternative method to improve the production of this crop in a sustainable and innovative way. This technique has been used in Europe and North America and allows better water use efficiency and maintains production quality \cite{3,4,5,6,7}. The 'ebb and flow' irrigation has advantages over conventional cultivation, such as savings in water and fertilizers, higher production per area, reduction of contamination by leaf pathogens, since the plant canopy is always kept dry, and decreased spread of contaminants in the environment, since there is little or no leaching of nutrients \cite{6,7}.

The rose is highly susceptible to the attack of several pest insects, among them the aphids that feed on the sap and, in case of high infestation, cause bent stems, weak foliage, early leaf fall and excrete honeydew that promotes the growth of sooty mold on the surface of leaves and flowers, significantly damaging the plant \cite{2,8}. Among the aphids of the rose, we can highlight the \textit{Rhodobium porosum} \cite{Sanderson,1900} (Hemiptera: Aphididae), which mainly attacks seedlings, producing deformations and decreasing their growth, apart from being a vector for viruses \cite{2,9,10}. The use of chemicals is still the main strategy to control aphids and they can lead to selection of resistant populations, environmental contamination, and increased costs of flower production \cite{1,11}.

Both excess or lack of fertilization are limiting factors for the plant development, and they also indirectly affect the population density of phytophagous insects, therefore influencing crop production \cite{12,13}. The growth of insect populations can be estimated based on development, survival and reproduction data that are called fertility life tables. The life tables are crucial for understanding the population dynamics of pest insects, and these data are the basis for the development of pest management strategies \cite{8,14,15,16}. The life table parameters, particularly the intrinsic rate of population increase, are important for evaluating the insect fitness and the level of the plant resistance or tolerance to herbivory \cite{8,12,14,15,16}. Rose cultivars have shown differences in suitability as hosts for the aphid \textit{Macrosiphum rosae} \cite{L.,1758} \cite{(Hemiptera: Aphididae), which was measured in relation to its survival, development, and reproduction rates \cite{8}. In this context, the production of roses still needs investments in new advanced technologies, such as soilless cultivation systems, use of fertirrigation and pest management strategies. As there is very scanty information on pest management in soilless rose cultivation, this study on reproductive potential and survival of the aphid \textit{R. porosum} on three rose cultivars exposed to different nutrient solutions in subsurface 'ebb and flow' irrigation systems, and in greenhouse protected environment was conducted, and the results of which would be useful in developing management strategies of the pest.

\section*{2. MATERIALS AND METHODS}

\subsection*{2.1 Local of the Experiment}

The experiment was carried out in a greenhouse \cite{7 m x 21 m} located in São João del Rei (MG), Brazil, at 910 m of altitude, 21°06' South latitude and 44°15' West longitude. The regional climate is included in the Cwb group, in accordance with the Köppen classification, being characterized by humid summer and dry winter. The average annual temperature is 19.2°C, with a minimum of
2.2 Experimental Design

The experiment was set up with two causal factors, that is, nutrient solution and rose cultivar. Two types of nutrient solution were tested, one recommended for cultivation of fruit vegetables, in accordance with [17] and another one based on [5]. Each solution was made available to plants of three different cultivars (‘Carola’ - red, ‘Greta’ - pink and ‘Tineke’ - white), totaling 6 treatments, with 4 replications and 24 experimental plots, in a completely randomized layout. Each experimental unit consisted of two trays containing two plastic pots each, and each pot with two plants, that is, eight plants per unit.

Rose seedlings from the ‘Carola’, ‘Greta’ and ‘Tineke’ cultivars were transplanted to the plastic pots (11L) containing expanded clay at the bottom and hydrated coconut fiber (Golden Mix Granulated - Amafibra®). Plastic trays (70 cm long, 50 cm wide and 22 cm high) were arranged on metal worktops and, on each tray, two pots with the two roses per plot. Periodically, cultural management of the roses was performed, including the removal of undesirable buds in the floral stem, placement of little nets to cover the flower buds, pruning and lateral stem bending or arching technique.

The irrigations were performed using the system of ‘ebb and flow’ subsurface irrigation [5,6], which included the immersion of 40 to 50% of the volume of the pots, from the base, in water solution, for a period of half an hour, in the frequency of once a day. During the system’s operation, the water solution flowed from the tanks through the piping to the trays where the pots with plants were placed. As soon as the trays were filling up with the nutrient solution, it was returned to the water tank, thus avoiding the liquid overflow. After the automatic shutdown of the system, the solution in the tray returned by gravity to its respective water tank. The hydraulic circuit to move the solution was individualized for each of the two nutrient solution and for each of the three rose cultivars being therefore composed of six water tanks, motor pumps and timers to activate the solution circulation in each treatment at the same time.

The composition of the nutrient solution 1, recommended for cultivation of fruit vegetables, in accordance with [17] and solution 2 based on [5] are presented in Table 1. Each nutrient solution was made available for the ‘Greta’, ‘Carola’ and ‘Tineke’ cultivars. The following water-soluble fertilizers were used to compose the nutrient solution according to the treatments: monopotassium phosphate, magnesium nitrate, potassium chloride, calcium nitrate, sodium molybdate, iron EDTA 6%, zinc sulphate, manganese sulphate, boric acid, copper sulphate and sulfur molybdate. The control of the nutrient solution was performed by means of a portable digital conductivimeter. The pH of the nutrient solution was maintained between 5.2 and 6.0 and the electrical conductivity between 1.5 and 2.5 mS/cm.

2.3 Life Table of the aphid Rhodobium porosum

The survival and population growth of R. porosum was evaluated in the six experimental plant treatments and replicates. The insects were trapped on the leaves with clip-cages [18] that were made with transparent disposable plastic cups (2.5 cm diameter x 1.0 cm high). The clip-cage was attached to the foliage using a metal clip and a rigid plastic base (3.0 x 3.0 cm) that was placed on the abaxial surface of the leaf, which prevented any folding of the foliage surface and the aphid nymph from escaping.

The aphids were reared using an initial cohort of twenty adult females that were individually cloistered inside the clip-cages on the rose foliage of each replicate per treatment. After 24 hours, the adult female (grandmother) was removed, leaving only one mother nymph inside each clip-cage. Two clip-cages per plant were used.

The experimental design was completely randomized, and every mother nymph was considered a replicate (twenty nymphs per treatment). The aphids were observed daily until they died to follow their developmental time, reproduction period, longevity, and fecundity, as well as the mortality. The development period comprised the number of days from birth of the
mother nymph until the first reproduction. The number of daughter nymphs was counted along the reproduction period. The daughter nymphs were counted and removed daily from each clip cage.

2.4 Data Analysis

The biological variables of *R. porosum* on the three cultivars and the two nutrient solutions were used in the fertility life table. Using the values of age intervals (x), specific fertility (m0) and probability of survival (l0) of the fertility, the life tables were calculated the time interval between each generation (T), the net reproductive rate (R0), the innate capacity to increase in number (r0), the time required for the population to double in number of individuals (TD) by using the TWOSEX-MSChart programme [19]. This program is written in Visual BASIC for the Windows operating system and is available at http://140.120.197.173/Ecology (Chung Hsing University) and at http://nhsbig.inhs.uiuc.edu/wes/chi.html (Illinois Natural History Survey). The means and standard errors of the population parameters were estimated by using the Bootstrap procedure with 100,000 re-sampling and the life table parameters of *R. porosum* on different rose cultivars were compared by using a paired bootstrap test [16,20].

3. RESULTS

The age-specific survival rate (l0) showed that the development period of *R. porosum* was, on an average, 10 days. The reproductive period of *R. porosum* started on the same day that the aphid became adult, between the 9th and 13th days. However, the aphids maintained on the 'Tineke' cultivar in nutrient solution 1 had a development period of 12 days and a reproductive period of 5 days (Fig. 1).

The highest specific fertility (m0), that is, the total number of nymphs per female that originated females at stage x (mean time interval) was recorded for the nutrient solution 1 at day 13 for 'Greta', day 11 for 'Carola' and day 15 for 'Tineke'. For the nutrient solution 2 in the 'Greta', 'Carola' and 'Tineke' cultivars the highest specific fertility (m0) was at 13°, 10° and 10° days, respectively (Fig. 1). When the females of *R. porosum* reached the peak of nymph laying, later, the number of produced offspring was gradually falling as a function of the aging. By the fifth day of life of the females over 50% of the offspring had already been produced. Regardless of the cultivar, the average production of nymphs in the nutrient solution 1 was 0.5 offspring per female and in nutrient solution 2 was 0.86 offspring per female added to the population per day (Fig. 1).

The decrease in the survival curve (l0) of *R. porosum* was observed as a function of longevity in all cultivars and in both solutions, reducing gradually, as the number of surviving females (l0) was decreasing with age intervals x (Fig. 1). The average longevity of *R. porosum* was of about 7.75 days, and on the 'Tineke' cultivar exposed to nutrient solution 1 the longevity was 5 days. All females were dead after 17 days.

Population growth parameters of *R. porosum* were affected by rose cultivars and nutritional solutions (Table 2). The time interval for each generation (T) was greater on the 'Tineke' cultivar exposed to nutrient solution 1 of 16.62 days (P < 0.05), compared to the other cultivars and solutions (Table 2). The value of T represents the average duration of a generation, which indicates the time between the birth of the parents and the birth of their offspring.

### Table 1. Composition of nutrient solutions

<table>
<thead>
<tr>
<th>Nutrients</th>
<th>Quantity (mg L⁻¹)</th>
<th>Solution 1</th>
<th>Solution 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>N-NO₃</td>
<td>169</td>
<td>424.7*</td>
<td></td>
</tr>
<tr>
<td>P</td>
<td>62</td>
<td>152</td>
<td></td>
</tr>
<tr>
<td>K</td>
<td>311</td>
<td>265.2</td>
<td></td>
</tr>
<tr>
<td>Ca</td>
<td>153</td>
<td>150</td>
<td></td>
</tr>
<tr>
<td>Mg</td>
<td>43</td>
<td>21.6</td>
<td></td>
</tr>
<tr>
<td>S</td>
<td>50</td>
<td>76.6</td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>0.2</td>
<td>0.51</td>
<td></td>
</tr>
<tr>
<td>Cu</td>
<td>0.03</td>
<td>0.13</td>
<td></td>
</tr>
<tr>
<td>Fe</td>
<td>4.3</td>
<td>2.02</td>
<td></td>
</tr>
<tr>
<td>Mn</td>
<td>1.1</td>
<td>0.39</td>
<td></td>
</tr>
<tr>
<td>Mo</td>
<td>0.05</td>
<td>0.06</td>
<td></td>
</tr>
<tr>
<td>Zn</td>
<td>0.3</td>
<td>0.26</td>
<td></td>
</tr>
</tbody>
</table>

*50% of the recommended dose based on [5]
Table 2. Life table parameters of *Rhodobium porosum* on rose cultivars in subsurface 'ebb and flow' irrigation system

<table>
<thead>
<tr>
<th>Treatment</th>
<th>T (days)</th>
<th>Ro (female)</th>
<th>( \lambda ) (female/day)</th>
<th>( r_m ) (female/female/day)</th>
<th>TD (days)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Greta - S1</td>
<td>14.21 ± 0.06a</td>
<td>2.73 ± 1.37ab</td>
<td>1.066 ± 0.033a</td>
<td>0.0636 ± 0.0309a</td>
<td>17.68</td>
</tr>
<tr>
<td>Greta - S2</td>
<td>13.91 ± 0.46a</td>
<td>3.91 ± 1.75ab</td>
<td>1.094 ± 0.041a</td>
<td>0.0897 ± 0.039a</td>
<td>7.06</td>
</tr>
<tr>
<td>Carola - S1</td>
<td>15.13 ± 0.69ab</td>
<td>2.69 ± 1.34ab</td>
<td>1.058 ± 0.044ab</td>
<td>0.0557 ± 0.0433ab</td>
<td>10.48</td>
</tr>
<tr>
<td>Carola - S2</td>
<td>15.23 ± 2.16ab</td>
<td>2.55 ± 1.13ab</td>
<td>1.050 ± 0.037ab</td>
<td>0.049 ± 0.371ab</td>
<td>12.26</td>
</tr>
<tr>
<td>Tineke - S1</td>
<td>16.62 ± 0.32b</td>
<td>0.52 ± 0.32b</td>
<td>0.946 ± 0.040b</td>
<td>-0.055 ± 0.0433b</td>
<td>-13.43</td>
</tr>
<tr>
<td>Tineke - S2</td>
<td>14.27 ± 0.48ab</td>
<td>4.00 ± 1.52a</td>
<td>1.096 ± 0.034a</td>
<td>0.0918 ± 0.0321a</td>
<td>7.1</td>
</tr>
</tbody>
</table>

\( T \) = average duration of a generation, \( R_o \) = net reproduction rate, \( \lambda \) = finite rate of increase, \( r_m \) = innate capacity to increase in number, TD = time required for the population to double in number of individuals. Values are mean ± SE. Means within a column followed by the same letter are not significantly different (\( P < 0.05 \); paired bootstrap test). Standard errors were estimated with 100,000 bootstrap resamplings.
Fig. 1. Average number of nymphs/females ($m_x$) and survival percentage ($l_x$) of *R. porosum* on rose cultivars in subsurface 'ebb and flow' irrigation system with different nutritive solutions. (A) 'Greta' - Solution 1; (B) 'Carola' - Solution 1; (C) 'Tineke' - Solution 1; (D) 'Greta' - Solution 2; (E) 'Carola' - Solution 2; (F) 'Tineke' - Solution 2
The net reproductive rate ($R_0$) showed that *R. porosum* had lower increased population growth on 'Tineke' in the nutrient solution 1 of 0.52 females, added to the population from one generation to another, compared to the other cultivars and solutions. In nutrient solution 2, the $R_0$ for 'Tineke' was 4 females, showing a higher production of offspring per female ($P < 0.05$) (Table 2).

The value of the finite rate of increase ($\lambda$) of *R. porosum* was lower for 'Tineke', with 0.9469 nymphs/female/day maintained in the nutrient solution 1, indicating the number of individuals added to the population per female for one day ($P < 0.05$) (Table 2). The innate capacity to increase ($r_m$) showed again that the 'Tineke' rose cultivated in nutrient solution 1 was not favorable for the population growth of *R. porosum*, with lower value $r_m$ -0.0555 female/female/day ($P < 0.05$), compared to the other cultivars and solutions (Table 2). The population parameters of net reproductive rate ($R_0$), intrinsic rate of increase ($r_m$) and finite rate of increase ($\lambda$) were lower on 'Tineke' in nutrient solution 1, indicating that this host plant is more resistant to *R. porosum* than the other cultivars. The time needed for the population to double in number of individuals (TD) showed no significant difference among the nutrient solution 1 in the 'Greta', 'Carola' and 'Tineke' cultivars and to nutrient solution 2 ($P > 0.05$) (Table 2).

### 4. DISCUSSION

The performance of *R. porosum* was influenced by the quality of the host plant in subsurface 'ebb and flow' irrigation system. Present study demonstrated that rose cultivars were greatly different in suitability as hosts for *R. porosum* when measured in terms of survival, development, and reproduction rates. A faster rate of development in a specific rose cultivar may enable a shorter life cycle and fast population growth of the aphid, which could be translated into greater population density. Shorter development periods and increased reproduction of herbivorous insects are indicators of better susceptibility of the host plant [8, 12].

In this study, the variation in *R. porosum* performance on rose cultivars in different nutrient solutions may be attributed to differences in the host plant quality, i.e differences in the levels of secondary compounds and nutrients, as well as insect growth inhibitors in the plant sap. Plants are very different in suitability as hosts for specific insects, when measured in terms of their survival, development, and reproduction rates [21]. According to [8] chemical properties such as secondary metabolites and nutrient balance and physical characteristics such as pubescence and tissue resistance differ from one host plant to another and consequently influence the population levels of aphids in different ways. Moderate resistance of the host plant can significantly contribute to reducing the destructive effects of aphids on the roses, thus decreasing the amount of pesticide application. The biological and behavioral traits of insects are important for better understanding trophic interactions among them. In addition, it also helps to characterize the susceptibility or resistance of cultivars against herbivorous which is used in integrated pest management strategies [14].

In general, the aphid kept in the 'Tineke' rose cultivated in the nutrient solution 1, showed lower net reproductive rate $R_0$, finite rate of increase $\lambda$, and innate capacity to increase in number $r_m$, as well as longer time interval between each generation $T$. These values on 'Tineke' in nutrient solution 1 were related to longer development of immature stages, higher mortality of the nymphs and lower fecundity of the *R. porosum* on this cultivar than in the other treatments. Consequently, 'Tineke' provided to be less susceptible and relatively resistant to *R. porosum* attack. The development period, survival rates and reproduction potential of aphids may be affected by several factors, among them, the quality of the host plant [12]. Longer development periods and lower reproduction of herbivorous insects in a host are sublethal effects that suggest higher plant resistance [8, 21]. [8] demonstrated that the variation in survival rate of the aphid *M. rosae* in different rose cultivars was the result of different nutritional qualities or secondary plant substances of these plants. The longer development period, lower total fecundity, and lower survival rate of *M. rosae* in a Tea cultivar indicated a higher resistance of the plant.

The 'Tineke' cultivar exposed to nutrient solution 1 caused a longer development period, lower survival rate, and reduced fecundity of *R. porosum*. The different responses of *R. porosum* between rose cultivars may be attributed to the type and amount of nutrients that are ingested by these insects. The nutrient solution 1, recommended for fruit-type vegetables, has a lower quantity of nitrogen than the nutrient solution 2. Because nitrogen is highly required by phytophagous insects, possibly, this nutrient may
have determined the differences in suitability as hosts for *R. porosum*. Nitrogen is an essential nutrient for the formation of proteins and is one of the most required nutrients by phytophagous insects and, in most cases, can be a limiting factor in insect development [13]. Previous studies have shown correlations between N content of the crop and higher infection rates by aphids [22, 23]. In the present study, this was also shown for aphids, because the nutrient solution 2 had 60% more nitrogen (424.7 mg.L⁻¹ nitrate (N-NO₃⁻) compared to the nutrient solution 1 (169 mg.L⁻¹ nitrate (N-NO₂⁻)). [22] found a positive correlation between the reproduction of *Myzus persicae* (Sulzer, 1776) (Hemiptera: Aphididae) and the amount of soluble nitrogen in the plant. [23] verified that nitrogen fertilization enhances performance of the aphid *Aphis gossypii* Glover, 1877 (Hemiptera: Aphididae) providing higher nymph development pattern, increased fecundity, and longevity under higher nitrogen concentration.

Negative effects of the rose cultivars and the nutrient solution were assessed on many life-history traits of *R. porosum*, especially the rates R₀ and rₘ, which summarize the insect physiological quality and indicate the reproductive success and the potential for population growth, in the absence of other variables. The net reproduction rate (R₀) estimates the average number of females generated per female that will be produced in the next generation. Thus, if R₀<1, the female population are decreasing. If R₀= 1, the female population are stable. When R₀>1, the population is growing [24]. In the present study, the 'Tineke' cultivar maintained in the nutrient solution 1 was not favorable for the population growth of *R. porosum* and showed that the female population would be decreasing, because presented the R₀ < 1 and the innate capacity to increase in number with negative value (rₘ = -0.0555 female/female/day). Many life history traits, such as the development period, survival and fecundity rate may affect the intrinsic rate of increase; therefore, this variable adequately summarizes the physiological attributes of an insect and its population potential to increase [12, 14]. According to [15] the rₘ is the appropriate index to assess the performance of an insect in different host plants, as well as its susceptibility. [8] have reported that host plants with lower rₘ values are more resistant to aphid attack, when compared to plants with higher rₘ values. [23] verified a substantial difference in rₘ between aphid *A. gossypii* populations on host plant quality. [24] explained that several biotic factors such as fertility, survival, and generation time affect the intrinsic rate of increase, thus rendering this parameter adequate in describing the physiological qualities of insects and to evaluate performance of an insect on different host plants and host resistance. Life table analysis is a standard ecological method to estimate the demographic variables that are related to the population dynamics of a species. Estimating the intrinsic rate of increase, generation time, finite growth rate and doubling time help to explain the fluctuations in population density and provide a better understanding of the population dynamics of a species [25].

The fertility life table has shown that the cultivar and the quality of the nutritional solution affected the performance of the aphid *R. porosum*. The results showed that *R. porosum* has less reproductive potential on 'Tineke' cultivar and that this cultivar kept on the nutrient solution 1 was possibly the least adequate host than the other rose cultivars, that is, the aphid had lower survival and fecundity than in the nutrient solution 2. Thus, to reduce problems with *R. porosum* attacks, we recommend using the nutrient solution 1 in the 'Tineke' rose cultivars in subsurface ‘ebb and flow’ irrigation system. The use of resistant or less favorable crop cultivars is considered an important component of the integrated pest management (IPM). Our results can be used in the development of an integrated pest management program in soilless rose cultivation. However, the causes of differences in suitability to aphids between the cultivars that we tested, is not well known and deserves a more detailed study.

5. CONCLUSION

*Rhodobium porosum* was able to complete its life cycle on the three different roses cultivars and in both nutritional solutions, although life history trait values were significantly different. The highest performance of the *R. porosum* was found on 'Carola' and 'Greta' in both nutrient solutions. The lowest performance of the *R. porosum* was found on 'Tineke' rose cultivated in the nutrient solution 1, usually indicated for fruit-type vegetables nutrition. Therefore, this seems to be the best combination to reduce problems with aphid attack in soilless rose cultivation.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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