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The Interleukin -2 and Interleukin -4 Response Profiles in Pediatric BCG Vaccinee

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Introduction: BCG childhood vaccination is still in use all over the world.BCG within the continuum of vaccinee childs activate immune cells to produceTH1 and TH2 cytokines The present work was aimed at investigating IL2 and IL4 cytokine responses among BCG pediatric vaccinee and non-vaccinee control in ALDiwanyah child and maternaty hospital-ALQadysiah province/Iraq.During the period Oct 2021-July 2021.

Materials And Methods: The study population was 60 BCG vaccinee child subject and 30 non-vaccinated healthy control subjects. Thirty out of the 60 vaccinated were scar bearing and the other 30 were non-scar bearing.The ELIZA determination of IL2 and IL4 in vaccinated and non-vaccinated child subjects sera were done in accordance with the manufacturer instructions.

Results: The cytokine responses were with marked heterogeneity in both groups of vaccinee. Though the mean concentration of vaccinated child groups were higher than that of control child subjects. There were fractions of both vaccinated groups that shown concentrations lower than the

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means of vaccinated and mean of control. This finding could be attributed to either of the followings; immune waning effect of age progression, minimal toleragenic potential of BCG epitopes, presence of an immunosuppressive tissue microenvironment. Population IL2 and IL4 responses include; low, moderate and high cytokine responders.

Conclusion: The IL2 and IL4 cytokine responses of BCG vaccinee are being heterogeneic and divergent. The immune herd plots were of Gaussian distribution plot types. A finding is being in line with cytokine herd responses of other microbial diseases in this area.

Keywords: BCG vaccinee; cytokine; Inteluekine -2; heterogeneity; Interluekine -4.

1. NTRODUCTION

BCG is still in use standard biologics that possess specific and non-specific immune potentials as; vaccine, vaccine adjuvant, nonspecific immune stimulant, child mortality reducer and protectant against heterogeneous pathogens [1]. BCG vaccination in childhood may activated immune cells to synthesize and secret an array of TH1 and TH2 cytokines profiles. Since the antigenic makeup of the whole BCG vaccine is complex multi-epitopic constitution. It does contain THi, TH2, toleragenic, anergic, immunosuppressive and/or allergenic epitopes [1-5]. The present work was aimed at determination of IL2 & IL4 cytokine response profiles among BCG vaccinee child subjects as compared to no-vaccinated child subjects.

2. MATERIALS AND METHODS

A population of 90 clinically proven healthy child subjects with an age range of 1/2 year to 10 years were the study group in Diwanyah Child andMaternaty Hospital-ALQadysiah province/Iraq during the period Oct to July 2021. Of which 60 were BCG vaccinated and 30 non-vaccinated as controls.Blood samples were collected,sera were saved at -18C freezer till use. Simple random sampling technique were followed to choose 15 random sample from each of; scar bearing, no-scar bearing and control. ELIZA kets of IL2 and IL4 were used following manufacturer instruction for determination of IL2 and IL4 cytokine responses in vaccinee groups and control.SSBS computer program for checking paired t test and F test to the differences between vaccine groups and control.

3. RESULTS

3.1 Interleukin -2 Response

The lower limits of IL2 concentration ,179.5 pg/ml. among no-scar bearing vaccinee child subjects was lower than that of healthy control subjects 198.6 pg/ml. The mean concentration values of scar bearing vaccinated child 887.2 pg/ml. and non-scar bearing vaccinated child subjects 985.4 pg/ml. were higher than the control mean concentration 198.2pg/ml. Though the non-scar bearing child subject mean concentration value was higher than scar bearing child subjects. Tables 1 and 2. Paired t test and F test for the difference between scar bearing and non-scar bearing vaccinee were statistically non-significant at P 0.05 levels. Both scar and non-scar bearing vaccinated subjects have shown low, moderate and high concentration individuals. Three immune herd responder fractions were found in both vaccinee groups. The immune herd plots were of Gaussian distribution plot types. Table 3, Fig. 1.

3.2 Interleukin -4 Responses

The lower limits of both vaccinated groups were higher than the lower limit of normal healthy control. Both of the scar bearing IL4 concentration mean 333.375pg/ml.and non-scar bearing IL4 concentration mean 380.083 pg/ml. vaccinee child subjects were higher than that of control subjects 51.875pg/ml. While the non-scar bearing vaccinated were higher than scar bearing vaccinated child subjects, Tables 2 and 3. Paired T test and F test for the difference between IL4 concentration between scar and non-scar bearing child subjects were statistically no-significant at P 0.05 levels. Both of the vaccinated groups have shown low moderate and high concentration individuals, Table 3.The immune herd responder types were as; low ,moderate and high responder types. The immune herd plot types were of normal Gaussian distribution plots, Fig. 2.

3.3 IL2 and IL4 Responses Comparative View

In non-scar bearing BCG vaccinee, one case of IL2 lower concentration limits than that of control. There were three different cases in each of IL2 and IL4 cytokine responses were lower than the

mean values of vaccinee and controls. Besides four cases of both IL2 & !L4 were their concentrations lower than the mean concentration values of vaccinated and control.In scar bearing vaccinated child subjects there were two cases of decrease of both IL2 & IL4 than mean concentration values of vaccinated and controls.Table-1.

Table 1. The IL2 and IL 4 cytokine concentration individual values in BCG Vaccinated child subjects

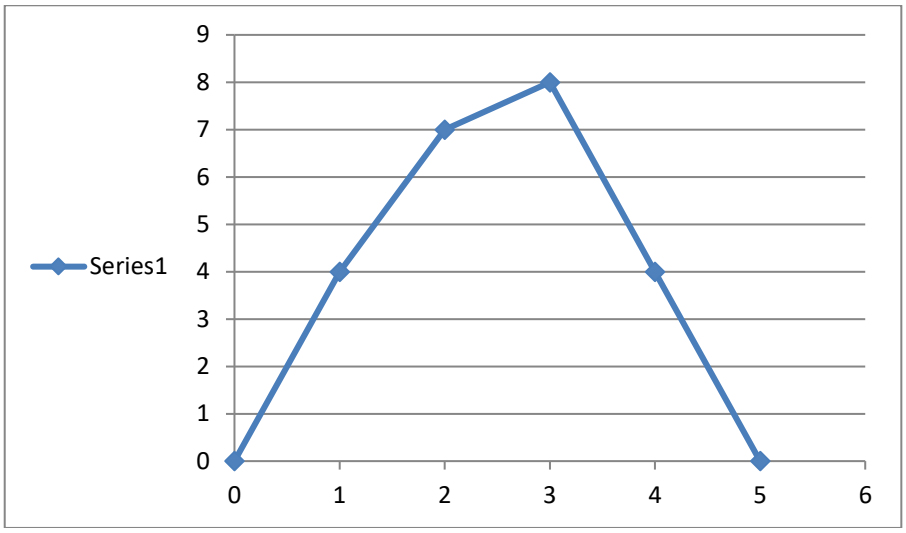
Child subject Sequence	Scar bearing IL2	Non-scar bearing IL2	Scar bearing IL4	Non-scar bearing IL4
1-	658.785	478.428	256.25	66.25
2-	402.357	274.857	78.53	57.343
3-	413.071	1815.929	144.375	314.531
4-	566.285	1733.786	184.531	314.531
5-	569.142	952.714	240	753.281
6-	759.857	1025.214	246.718	506.406
7-	535.857	557.0	187.812	439.218
8-	1053.429	179.5	238.906	353.437
9-	1709.5	353.071	765.781	57.031
10-	304.857	300.214	86.875	100
11-	1033.357	1777.714	355.321	380.468
12-	1887.0	667.714	773.593	599.375
13-	1682	1339.5	705.781	256.406
14-	468.428	1404.5	100.781	459.531
15-	1830.214	1839.857	635.781	698.281
Mean	881.2	938.4	333.385	380.083
Control	548.884	548.884	224.281	224.281

Table 2. Biometery of IL2 and IL4 concentrations in BCG vaccinated child subjects

Feature	BCG scar	BCG non-scar	Control
IL2 variations			**Control variations**
Minimum	308.266	179.5	198.6
Mean	887.2	988.4	548.884
Median	1619.267	191.266	1412.933
Maximum	1784.933	1740.93	1421.933
Range	308.266-1784.933	179.5-1740.93	198.6-1412.933
IL4 variations			**Control variations**
Minimum	78.599	57.031	51.875
Mean	333.375	380.083	224.281
Median	238.906	57.031	468.437
Maximum	773.593	754	468.375
Range	78-774	57-754	52-469

Table 3. IL2 and IL4 cytokine herd responder types in BCG vaccinated child subjects

Responder Type	Scar bearing BCG vaccinee	Non-scar bearing BCG vaccinee	Control Non-vaccinated
IL2 Responders			
Low	308-470	191-400	198-300
Moderate	471-1100	401-1300	301-999
High	1101-1732	1301-1740	1000-1443
IL4 responders			
Low	86-200	51-299	51-199
Moderate	201-599	300-599	200-299
High	600-780	600-760	280-470



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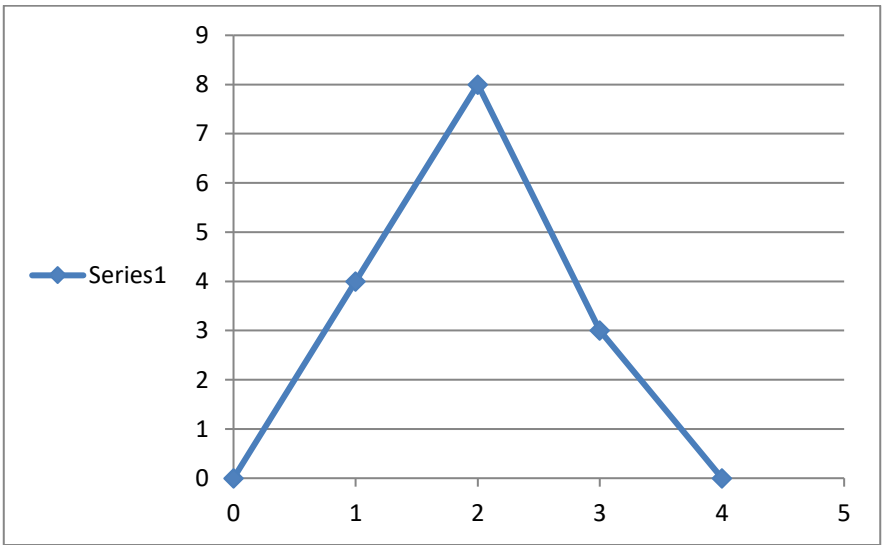

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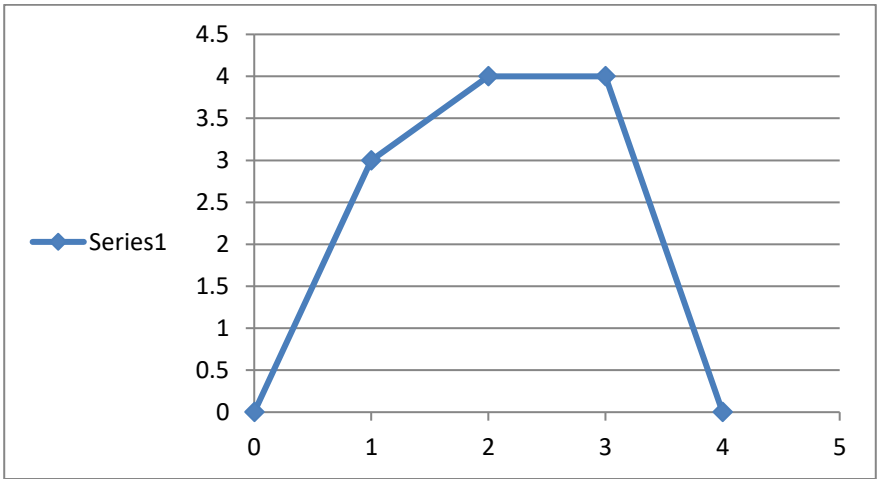

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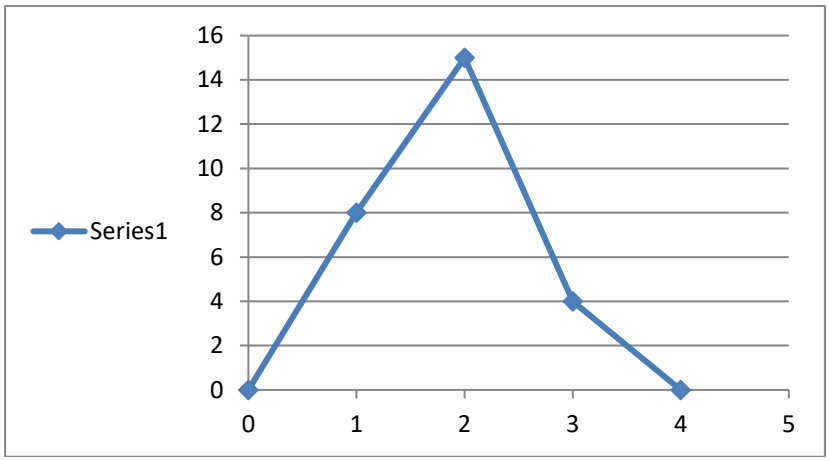
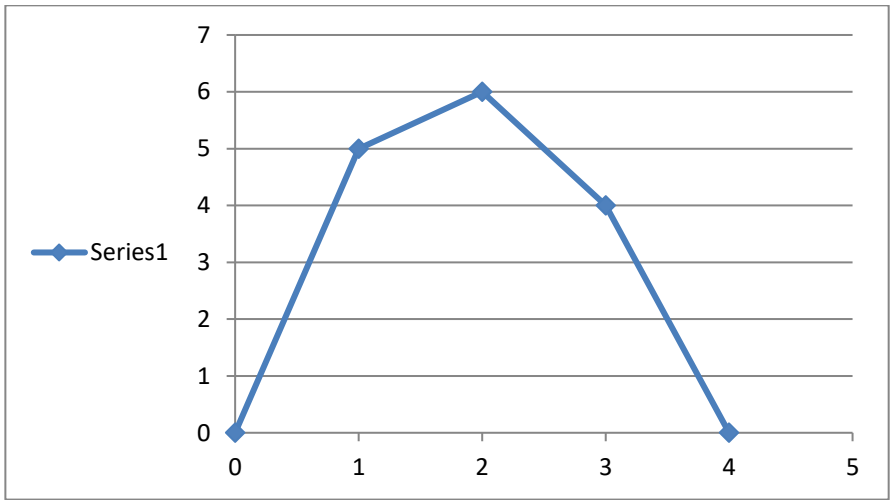


IL2 (4) control

Fig. 1. Immune Herd normal ditribution plots of IL2 responses in (1,scar bearing;2 non-scar bearing;3, cumulated) pediatric BCG vaccinee and IL2 (4) control



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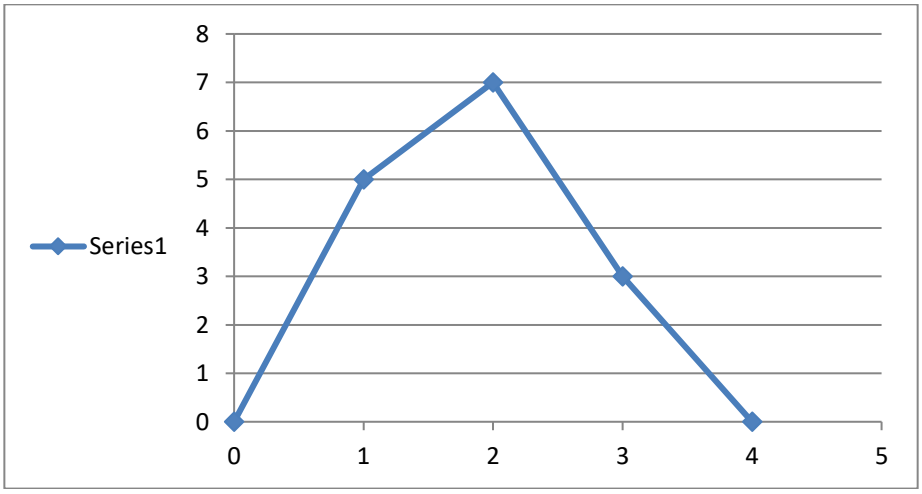

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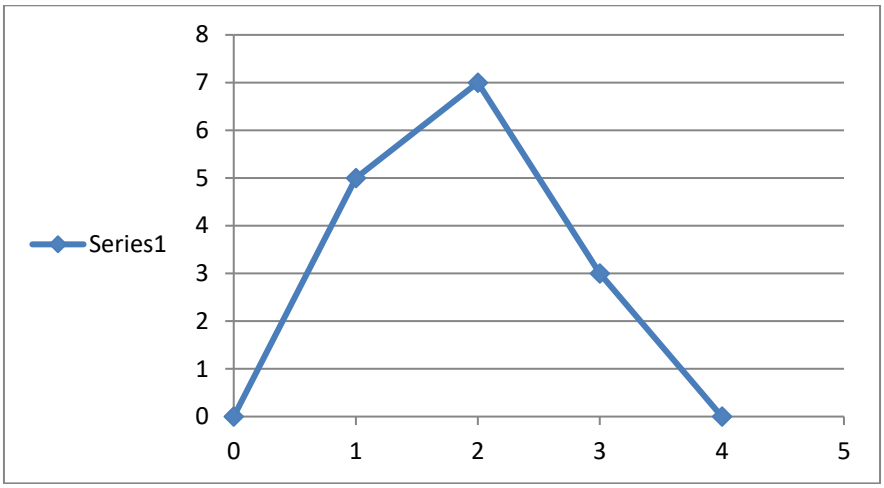
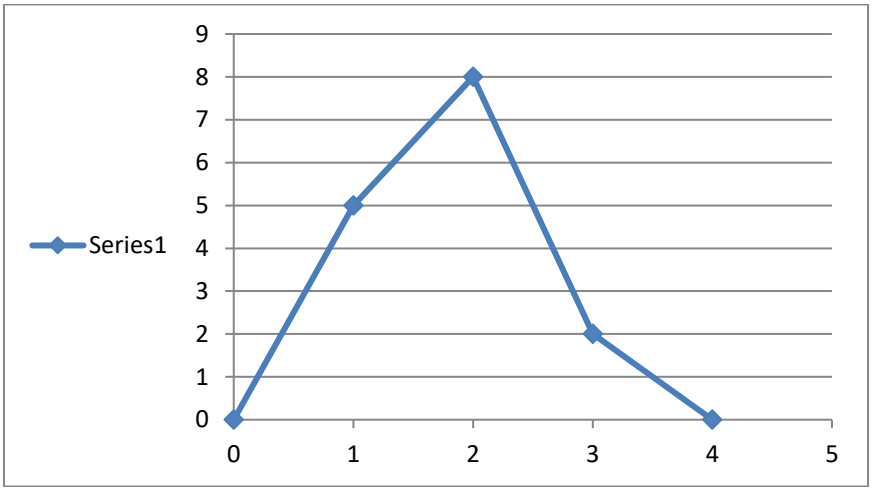


IL 4 (3) cumulated



IL 4 (4) control

Fig. 2. Immune herd normal distribution plots of IL2 responses in (1,scar bearing;2,nonscar bearing; 3,cumulted) pediatric BCG vaccinee and IL4(4) control

4. DISCUSSION

Shnawa and Karim [6], in their work proved that the size of BCG scar in vaccinated child subjects fade up as the child progress in age till five years. The relation was simple negative linear relation. The relation was moderate inverse between age and size of the scar. That is to say waning of gross and impart the cellular immunity did happened as child grew up to five years.Second to this finding the cellular immune reactions of the vaccinated child characterized by heterogeneity of their responses.The apparent heterogeneity of child immune response to BCG was inline with that of other workers[7,8,9]. BCG antigenic makeup is complex multi-epitopic to which;TH1,TH2,allergenic, toleragenic auto-reactive, immune-suppressive and anergic epitopes[10].Within the lymphoid tissue niche continuum of the BCG vaccinated subjects ,macrophages took up the BCG ,process their antigenic epitopes in combination with CD-1 –d MHC molecules to be surface located on the macrophages to be able to activate naïve T cells. Activated naive T cells evolve to be either effector or memory functioning T cells. Effector T cells synthesize and secret TH1, TH2 cytokine array. Among whichIL2 and IL4 [11].

IL2 cytokine was discovered as T cell growth factor thus a key components in immune activation but at times may functions as regulatory T cell initiator thus leading to immune suppression [12]. It is monomeric glycoprotein

that synthesized and secreted by;CD4+,CD8+ and dendritic cells. IL2 played a central role in the activation of regulatory T cells to produce TNF alpha, IFNg and enhance cytolytic activity of natural killer cells .As well as participate in the pathogenesis of infectious disease [13]. Thirteen out of the 30 BCG vaccinee child subjects have shown elevated levels of IL2 than in healthy control this finding is in line with results of other workers [14,15]. The fourteen out of the 30 BCG vaccinated child subjects were with lower IL2 concentration than means of vaccinated and control. This finding was in agreement with that of Kumar etal [2,3] in elderly BCG vaccinee.Both of elderly and childhood are holding the position of life extreme characterized impart by week immune activity.

The antigenic stimulation activate TH2 cells and follicular helper T cells to secret IL4.The follicular helper T cell IL4 control IgE and IgG1 antibody responses and has a role in germinal centers formation in the secondary lymphoid tissue during humoral immune responses [16]. The activation of macrophages by IL4 induces protective innate memory against microbial challenges [17]. IL4 induces naive T cells differentiation into Th2 cells. While in B cells IL4 derives the Ig class switching to IgG1 and IgE. Both of IL13 and IL4 induces alternative macrophage activation [18]. Hence IL4 is critical not only for precise control of Ig production but also related to inflammation, fibrosis, allergic reactions and antitumor activity [19]. Major fraction of BCG vaccinee child subjects were showing high levels of IL4 than control [17]. Other fraction of BCG vaccinated child subjects were showing lower IL4 concentration than concentration means of control. This may be due to heterogeneity, immunosuppressive tissue microenvironment or toleragenic epitope in BCG vaccine [20]. The immune herd plots of both IL2 and IL4 BCG vaccinated childs were of gaussian distribution types this was inline with previous reports in this area [21,22].

One may rise a question as ,is it really the elevation ofIL2 and IL4 cytokines in BCG vaccinated child subjects was more than their concentration levels in non-vaccinated healthy control child subjects due to BCG or due to other inducer?. The answer can be as; So far the inclusion and exclusion criteria were applied firmly then any increase and/or decrease in the IL2 & IL4 concentrations among the vaccinee during the post-vaccination period than that of the baseline concentrations in the healthy non-vaccinated child subjects, the case is logically due to BCG and denoted as immune conversion intensity. The mentioned decrease may be attributed to either of the followings; Presence of inhibitory epitope in some vaccine makes or local tissue immunosuppressive microenvironment in vaccinee hosts [20]. A second question, may be on the waiting cue. Can a BCG scar be a grantee for good cellular immunity in vasccinee .The answer may be phrased as; Not all the BCG scar bearing child have good reactive immunity and not all non-BCG scar bearing child have poor immune reactivity. Part of both of vaccinated groups has potent cellular immunity and the other part was being of poor cellular immunity.IL2 and IL4cytokine herd responses were evident and herd plots were found of Gaussian distribution plots just as that reported by Shnawa et al. [21,22].

5. CONCLUSION

BCG induce cellular immune responses and delayed hypersensitivity responses. Such responses were found to be heterogenic or divergent.IL2 and IL4 cytokine responses in BCG vaccinee child subjects were found including individuals of low ,moderate and high concentrations.IL2 and IL4 cytokine herd responses were found to be as; low ,moderate and high responders.IL2 and IL4 responses were found to be in a balance state. As they express both low and high in same vaccinated child subjects. Low responses can be attributed to an age related immune response waning or presence of toleragenic epitope in BCG antigenic make up.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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