The effect of fertilization on yield components and quality parameters of soybeans [(*Glycine max* (L.) Merr.] seeds

Vplyv hnojenia na vybrané úrodotvorné prvky a kvalitatívne parametre semena sóje [(*Glycine max* (L.) Merr.]

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Abstract

Field experiment was carry out at experimental farm Oponice (West Slovakia, 48° 28´N, 18° 9´E) in 2004-2007. The aim of the work was to evaluate the influence of nitrogen fertilization treatments on yield and yield component and selected quality parameters (proteins, oil, fibre, isoflavones) of soybean seeds variety Korada, in agri-environmental condition of western part of Slovakia. The fertilization treatments were as follows: (I) unfertilized treatment; (II) application of nitrogen fertilizers LAV 27 % (ammonium nitrate with lime - 40 kg*ha^{-1} nitrogen) in the vegetative stage V2; (III) Humix complet in split application of total dose 4 +4 l*ha^{-1} applied in growth stage of V2 and in reproductive growth stage of R1 (beginning bloom). Humix complet contain bioactive ingredient and nutrients for plant nutrition with 2.5% of humic acids, 4.0 % total N, 0.5 P_2O_5, 3 % K_2O, and micronutrients. Result showed the highest variation of pods per plant in dependence of year condition (range 17.8 – 24.7). No significant differences between numbers of seeds were noted (2.13 – 2.29). The split application of Humic complet significantly influence the higher number of pods per plant (24.8) while keeping the number of seeds per pod (2.13). The yield of seeds was significantly influenced by weather conditions (2.84 t*ha^{-1} – 4.68 t*ha^{-1}) and by fertilization treatments. Supplemental ammonium nitrate with lime (treatments II) in the vegetative stage of V2 or Humix complet (treatment III) applied in V2 and R1 stages significantly increased yield of soybean seeds up to 3.91 t*ha^{-1} and 4.27 t*ha^{-1} with comparison to control treatment (3.49 t*ha^{-1}). Content of protein, oil and fibre was not significantly differing between fertilization treatments. In spite of significantly higher soybean yield in Humic complet treatment, content of protein (37.67%), oil (15.83 %) and fibre (9.47 %) was at the same level as lower yielded treatments. Soybean seeds from higher yielded fertilization treatments have significantly less content of isoflavones. Split application of Humic complet is effective method for improving yield of soybean seeds while maintaining the composition of protein oil and fibre.

Keywords: fertilization, isoflavones, protein, soybeans, oil, yield, yield component

Abstrakt

Poľný pokus bol realizovaný na experimentálnej farme v Oponiciach v rokoch 2004-2007. Cieľom práce bolo hodnotiť vplyv dusíkatého hnojenia na úrodu semena a úrodotvorné prvky a vybrané kvalitatívne vlastnosti (bielkoviny, olej, vláknina, izoflavóny) semena sóje odrody Korada v agroklimatických podmienkach západnej časti Slovenska. Boli hodnotené nasledovné varianty hnojenia: (I) nehnojená kontrola; (II) hnojenie LAV 27% (liadok amónny s vápencom – 40 kg*ha^{-1} dusíka) vo vegetatívnej faze V2; (III) delená aplikácia Humix komplet v dávke 4 + 4 l*ha^{-1} vo vegetatívnej faze V2 a v reproduktívnej faze R1 (začiatok kvitnutia). Humix komplet obsahuje bioaktívne látky a živiny s obsahom 2,5% humínových kyselíny, 4,0 % N celkový, 0,5 % P_2O_5, 3 % K_2O a mikroelementy. Najvyššiu premenlivosť mal počet strukov na rastlinu v závislosti od podmienok ročníka (interval 17,8 – 24,7 kusov). Rozdiely medzi počtom zŕn boli nepreukazné a pohybovali sa v intervale 2,13 – 2,29 kusov. Delená aplikácia Humix komplet preukazne podporila najvyšší počet strukov na rastlinu (24,8) pri zachovaní počtu zŕn v struku (2,13). Úroda semien bola preukazne ovplyvnená podmienkami pestovateľského ročníka (2,84 t*ha^{-1} – 4,68 t*ha^{-1}) a hnojením. Dodatková dávka dusíka vo vegetatívnej faze V2 (liadok amónny s vápencom) alebo Humix komplet aplikovaný vo faze V2 a R1 preukazne zvýšil úrodu semena sóje na úroveň 3,91 t* ha^{-1} až 4,27 t*ha^{-1}.v porovnaní s úrodou 3,49 t*ha^{-1} dosiahnutou na kontrolnom variante. Obsah bielkovín, oleja a vlákniny nebol hnojením preukazne ovplyvnený. Napriek preukazne najvyššej úrody sóje na variante s aplikáciou Humic komplet obsah bilekovín (37,67%) oleja (15,83%) a vlákniny (9,47%) zostal na rovnakej úrovni ako u variantov s nižšou úrodou. Semeno sóje získané s hnojených a preukazne úrodnejších variantov malo preukazne nižší obsah izoflavónov. Delená aplikácia Humix komplet je efektívny spôsob pre zlepšenie úrody sóje pri zachovaní obsahu bielkovín, oleja a vlákniny.

Kľúčové slová: bielkoviny, hnojenie, izoflavóny, olej, sója, vláknina, úroda, úrodotvorné prvky

Introduction

Soybeans [(*Glycine max* (L.) Merr.] as a major source of protein, energy, polyunsaturated fat, fibres, isoflavones both for humans and livestock belongs to the most important crop of the world. Soybean seeds contain an average of 36 %-38 % protein and 19 % oil, on dry weight basis, but both genetic and environmental factors can strongly affect the seed composition (Krishnan, 2000). In soybean, isoflavones are principally found in roots and seeds; however, the metabolite has also been isolated from leaf and stem tissue (Romani et al., 2003). Daidzein and genistein, which are isoflavone aglycones are mainly found in soybeans and soy products (Murphy and Barsi, 2005).

Five major soybean producers – the USA, Brazil, Argentina, China and India – produced total 227.7 million metric tons which account for 90.5 % of the global soybean production in 2011 (World Statistic, 2012). In Slovakia, productivity of 1.66 - 2.1 t*ha^{-1} was lower compared to productivity of 2.8-2.9 t*ha^{-1} in best producers. Acreage of growing area of soybean increased from 5.5 thousand hectares in 2008 up to 22 thousand in 2012. For increasing and the stabilization of growing area we need new commercially growing varieties suitable for agro-climatic condition of Slovakia.

Sustainable agriculture is characterised by the introduction of more diversity in crops, and it should ensure fertility of soil and nitrogen balance. Legumes have low input requirement and enhance diversification in crop rotation (Týr et al., 2009; Fazekašová et al., 2011).

Soybean plants can use nitrogen released by mineralization, residual soil nitrogen, fertilizer nitrogen or atmospheric nitrogen, which is converted into a usable form in root nodules through a symbiotic relationship between *Bradyrhizobium japonicum* bacteria and the soybean plant. While the soil is the primary source of nitrogen for many crops, soybean obtains 65 % – 85 % of its needs through the symbiotic nitrogen fixation process (Rao and Reddy, 2010).

The aim of the present study was to evaluate the influence of fertilization treatments on yield and yield component and selected quality parameters - proteins, oil, fibre and isoflavones in agri-environmental condition of south-western part of Slovakia.

Materials and Methods

Field experiment was carry out at experimental farm Oponice (West Slovakia, 48° 28´N, 18° 9´E)) in 2004-2007. The experimental site belongs to warm and moderate arid climatic region. The average rainfall is 607 mm. The average temperature is 9.5 °C, for the growing season 16.1°C. The main soil type is loamy Haplic Luvisols on loess. Forecrop was sugar beet each year. The Canadian non-GMO conventional soybean variety Korada registered in Slovakia was growing. The conventional tillage practices as mouldboard ploughing to the depth 250 mm in autumn and harrowing in the spring were used. Immediately after sowing, pre-emergent herbicides Acenit A 880 EC with acetochlor as active ingredient in dose of 2.2 l*ha^{-1} and Pulzar with imazamox as active ingredient in dose of 1.0 l*ha^{-1} were applied. The sowing/harvest data of growing soybean are as follows: 14 May 2004/08 October 2004; 03 May 2005/10 October 2005; 27 April 2006/20 October 2006; 24 April 2007/01 October 2007. The sowing pattern was 0.6 million of fertile seeds per hectare into the depth 0.05 m, row space was 0.125 m. The randomized complete block design with a three replications was used. The acreage of fertilisation treatment plots was 14 m^2. Before the harvest, plant samples were collected for biometrical analyses.

Seeds were inoculated by HiStick preparation. The fertilization treatments were as follows: (I) unfertilized treatment; (II) application of nitrogen fertilizers LAV 27 % (ammonium nitrate with lime - 40 kg*ha^{-1} nitrogen) in the vegetative stage V2; (III) Humix complet in split application of total dose 4+4 l*ha^{-1} applied in growth stage of V2 and in reproductive growth stage of R1 (beginning bloom). Soybean phenology description with relation to vegetative and reproductive stages was made according Fehr et al. (1971). Soil characteristics of experimental site are described in the table1.

Table 1. The agrochemical soil characteristics (0.3m soil layer) before sowing in 2004 – 2007

Tabuľka 1. Agrochemické vlastnosti pôdy (0,3 m vrstva pôdy) pred sejbou v rokoch 2004 – 2007

Year	Content of element (mg*kg^{-1})							
	N_{an}	$N\text{-}NH_4$	$N\text{-}NO_3$	P	K	Mg	Humus	pH
2004	51.88	14.,05	37.83	89.0	320.6	230.0	2.55	7.06
2005	37.71	11.37	26.34	82.0	323.0	200.0	2.68	7.19
2006	25.94	10.70	15.24	92.0	323.0	430.0	3.20	7.13
2007	27.00	14.82	12.18	60.0	232.0	260.0	2.42	7.48

Humix complet contain bioactive ingredient and nutrients for plant nutrition with 2.5 % of humic acids, 4.0 % total N, 0.5 P_2O_5, 3 % K_2O, and micronutrients: Fe, Cu, B, Co, Zn, Mn, Mo and pH of 11-13. Grain yield and main yield component were analysed. Software package Statistica ver.10 for ANOVA analysis and testing contrasts by Tukey test was used.

For soil testing, Mehlich III method for nutrient analysis was used. For soya seeds analysis the content of oil was determined by Soxhlet Extraction Apparatus, protein by Barstein methods crude fiber by Henneberg-Stohman method. For determination of macro elements (Na, K, Ca, Mg) the Avanta atomic absorption spectrometer was used. Concentration of daidzein and genistein were determined using a high-performance liquid chromatography.

Approximately 1 g of soybean seed from each cultivar was ground to a fine powder. The powder was extracted with 60% methanol. Daidzein and genistein were separated on a HPLC using the following instruments conditions: HPLC, Waters BREEZE with: Waters binary pump 1525, UV-VIS detector 2487, column heater, in-line degasser and Column: PUROSPHER STAR C18 250x4.6 mm, 5 micron particle size. Solvent: A: was 02 % H_3PO_4 and solvent B: was 60 % HPLC grade methanol.

Results and Discussion

The water requirement of soybean varies depending upon the crop cultivar, growing season, rainfall pattern and temperature. The important factors for the legumes are time of stress occurring and relatively different reaction in the particular crop-producing elements on the stress (Krivosudská and Filová, 2013).

The evaluated growing season vary in temperature and rainfall distribution (Figure 1 and Figure 2). The humid condition in 2004 with total amount of rainfall in June and July (127.6 mm), predisposing higher production of pods per plant, but very dry August (19.4 mm) during the seeds filling stage reduced the weight of 1000 seeds (Table 2). Brevedan and Egli (2003) also reported that water stress during the seed-filling stage resulting in earlier maturity and smaller seed size. They noted decrease in pods on the plant from 91 to 86 and decrease in seed size from 233 mg to 156 mg per seed. Crops grown during very hot summer month requires higher amounts of water than that grown during milder mouths (Singh, 2010).

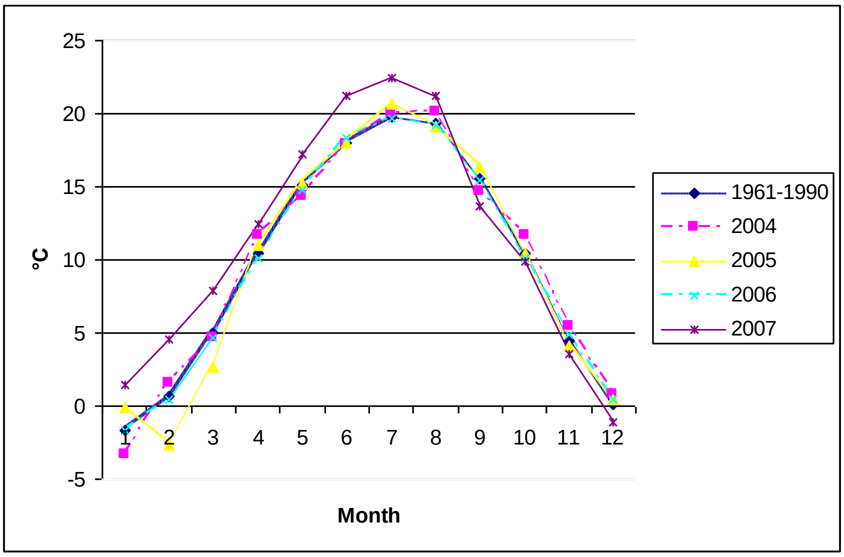

Figure 1. Temperature in year 2004-2007 and LTA 1961-1990

Obrázok 1. Teplota v rokoch 2004-2007 a dlhodobý priemer za 1961-1990

Extraordinary warm July and very warm August 2007 combined with low dose of sum of precipitation (75.3 mm) significantly influenced the lower yield of soybean seeds.

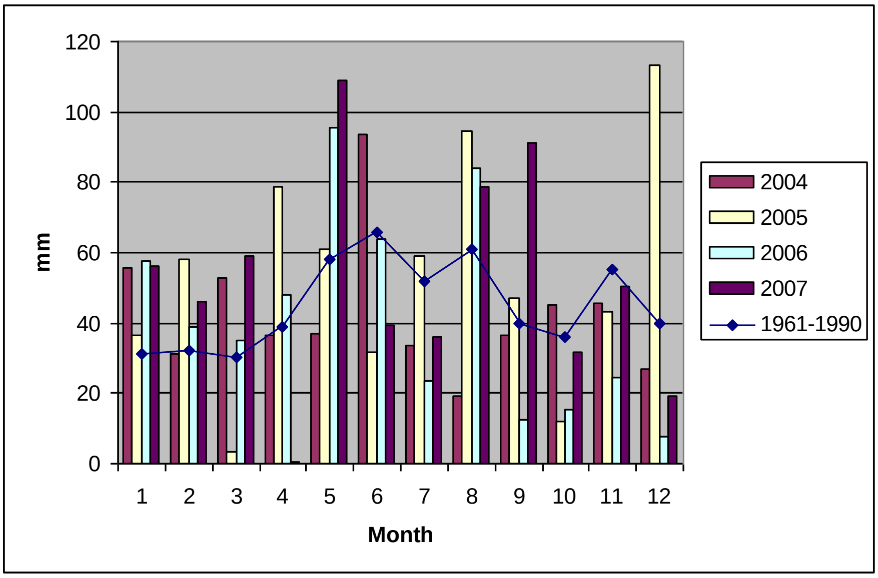

Figure 2. Precipitation pattern in year 2004-2007 and LTA 1961-1990

Obrázok 2. Priebeh zrážok v rokoch 2004-2007 a dlhodobý priemer za 1961-1990

During flowering and pod formation sufficient amount of precipitation for yield production was noted in July and August of 2005. The main season of the year 2006 was characterised by dry July and wet August.

In 2007 the most unsuitable wheatear condition for soybean plant development were noted. Vegetative and reproductive stages reflect the growing condition of specific site. Vegetative growth phase (from emergence to flowering) accounted for 55 days in 2004 up to 64 day in 2006.

In this year the flower and pod abortion was noted and lower number of pods per plant (17.8) was reached in an average (Table 2). Pod abortion occurs in drought-stressed soybean, which may be due to low ability of photosynthate in leaves and impaired ability of pods to utilize sucrose (Liu et al., 2004).

Table 2. Yield forming components of soybean, number of pods per plant, seeds per pod and weight of 1000-seeds, variety Korada in years 2004-2007

Tabuľka 2. Úrodotvorné prvky sóje, počet strukov na rastlinu, počet semien na struk, váha 1000 semien, odroda Korada v rokoch 2004-2007

Factor	Pods per plant	Number of seeds per pod	1000-seeds weight
Year			
2004	24.38b	2.16a	156.52a
2005	24.72b	2.29a	18196c
2006	18.80a	2.13a	184.84c
2007	17.80a	2.15a	168.66b
Fertilisation treatments			
Control	18.42a	2.16ab	167.47a
LAV	21.05ab	2.29b	177.58a
Humix complet	24.80b	2.13a	173.94a

The means between years and fertilization followed by the same letter are not significant at α <0.01 probability level. LAV - application of nitrogen fertilizers; Humix complet in split application of total dose 4+4 l*ha^{-1}

Priemery medzi rokmi a hnojením označené rovnakými písmenami nie sú preukazné na hranici významnosti α <0.01. LAV – aplikácia dusíkatého hnojenia; Humix komplet v delenej aplikácii 4+4 l*ha^{-1}

Production of soybean plant is associated with variation in seed number as a function of pods number and seed number in pods. High positive and significant association existed between seed yield and number of pods per plant as well as the number of seeds per plant (Ikeogu and Nwofia, 2013). Hence good growing condition for the establishment and development of the crop during the flowering and pod stage creates condition for seed number (Egli, 2005). Year 2004 and 2005 has significantly the most favourable conditions for creation of pods number per plant (24.38 – 24.72) with comparison to years condition of 2006 and 2007 (17.80 – 18.80).Variation in seed size is also result of variation in environmental during seed filling stage. Variation in assimilate availability can effect seed growth rate and increase or decrease seed size and yield (Egli, 1997).

The lower yield of soya seeds was reached in 2007 (Table 2 and Table 3) with average yield component as follows: number of pods – 18.80, seed per pods – 2.15 and weight of 1000 seeds – 168.66 g. Similar parameters of pods number (17.8) and seed per pods (2.13) were also noted in 2006, but better seed size parameters (184.84 g) compensate the lower parameters which determined the seed number.

Seed size can partially compensate the yield after fruit and seed numbers are fixed (Egli, 2005).

The split application of Humic complet significantly influence the number of pods per plant (24.80) while keeping the number of seeds per pod (2.13) with comparison to control treatment.

Table 3. Yield of soybean seeds in evaluated years and fertilization treatments, Oponice site, year 2004 – 2007

Tabuľka 3. Úroda semena sóje v sledovaných rokoch a variantoch hnojena, farma Oponice, 2004 – 2007

Factor	Yield (t*ha^{-1})
Year	
2004	4.31c
2005	4.68d
2006	3.73b
2007	2.84a
Fertilization treatments	
Control	3.49a
LAV 27 %	3.91b
Humix komplet	4.27c

The means between years or fertilization followed by the same letter are not significant at α <0.01 probability level; LAV - application of nitrogen fertilizers; Humix complet in split application of total dose 4+4 l*ha^{-1}

Priemery medzi rokmi a hnojením označené rovnakými písmenami nie sú preukazné na hranici významnosti α <0.01. LAV – aplikácia dusíkatého hnojenia; Humix komplet v delenej aplikácii 4+4 l*ha^{-1}

The variability of year condition significantly influenced the seed yield (Table 3). Relatively good growing condition in the year 2004 and 2005, support development of good parameters of yield components and consequently higher yield.

Soybean obtains up to 65 – 85 % of its needs through the symbiotic nitrogen fixation. A high rate of nitrogen fertilizers suppresses nitrogen fixation and most specialists recommended either no fertilizer nitrogen or modest application of 30 – 50 kg*ha^{-1}, either at sowing or just before flowering (Rao and Reddy, 2010).

According Singh et al. (2010) at 45 and 75 days after sowing application of nitrogen had a non-significant effect on number and dry weight of nodules, whereas, at 105 days after sowing and at maturity it resulted in a significant reduction of these attributes than where no nitrogen was applied.

Fertilization treatments significantly influenced the yield of seed (Table 3). We have evaluated only modest dose of nitrogen in LAV fertilization treatment (40 kg*ha^{-1}) and split application of Humix complete (only 4 % of nitrogen content it total dose of 8 l*ha^{-1}).

Table 4. The effect of different fertilization treatments and year condition on content of protein, oil and fibre in soybean seeds variety Korada

Tabuľa 4. Vplyv rôznych spôsobov hnojenia a podmienok ročníka na obsah bielkovín, oleja a vlákniny v semenách sóje odrody Korada

Factor	Proteins %	Oil %	Fibre %
Year			
2005	36.00a	16.02NS	9.49NS
2006	32.21a	16.21NS	9.66NS
2007	44.25b	16.86NS	9.83NS
Fertilization			
Control	36.87NS	16.98NS	9.63NS
LAV	37.92NS	16.28NS	9.87NS
Humix complet	37.67NS	15.83NS	9.47NS

The means between years or fertilization followed by the same letter are not significant at α <0.01 probability level; NS-non significant; LAV - application of nitrogen fertilizers; Humix complet in split application of total dose 4+4 l*ha^{-1}

Priemery medzi rokmi a hnojením označené rovnakými písmenami nie sú preukazné na hranici významnosti α <0.01. LAV – aplikácia dusíkatého hnojenia; Humix komplet v delenej aplikácii 4+4 l*ha^{-1}

Split application of Humix complet in stage V2 and R1 seems to be very effective way for improving yield mainly due to significant increasing of pod per plants (Table 2). Besides nitrogen, Humix complet content also important micronutrients.

Also Chahturvedi et al. (2012) revealed that yield attributes as pods/plant, seeds/pod and hundred seed weight were increased significantly by the addition of micronutrients and FYM at both evaluated fertility levels (50 % and 100 % NPK).

For enhancement of yield attributes of soya plants Mondal and Mondal (2012) recommended application of 1.5 % urea thrice from the beginning of flowering to pod development stage with an interval of 10 days. This treatment which showed superiority in yield attributes (number of pods per plant, number of seeds per pod and 100-seed weight) over other treatments. Soybean is important source of protein and oil. On average, the protein content in commercial cultivars is approximately 40 %, ranging from 34 % – to 48 %, depending upon the genotype, growing conditions and cultural practices of the crop (Kumar et al., 2010).

Variability of protein content was significantly influence by year condition with probability level =0.000527. During investigated period of 2005-2007, protein content of soybean variety Korada range from 36 % in 2005 to 44.25 % in 2007. Significantly higher yield of protein content was reaches in 2007 with comparison to 2005 and 2006 (Table 4). Due to the inverse relationship between protein content and yield (Macák et al., 2010) and very warm and extraordinary warm period of reproductive stage, the high content of protein was reached in 2007 with average yield 2.84 $t*ha^{-1}$. Totally the highest protein yield was reached in 2005 mainly due to the significantly higher yield of soya bean. Oil content was not at the required level.

Content of protein, oil and fibre was not significantly influence by different fertilization treatments. Total yield of protein per hectare was 1609 $kg*ha^{-1}$ in Humic treatment with comparison to 1483 $kg*ha^{-1}$ LAV and 1287 $kg*ha^{-1}$ in control treatment. Soybean seeds from higher yielded fertilization treatments have significantly less content of isoflavones. Content of daidzein/genistein range from 2.85/27.1 $mg*kg^{-1}$ in Humix complet application and 4.3/5.27 $mg*kg^{-1}$ in LAV application of nitrogen fertilizers up to 6.5/71.5 $mg*kg^{-1}$ in control treatment in 2007.

Conclusions

Yield of soybean is associated with variation in seed number as a function of pods number and seed number in pods. Result obtained in this study showed the highest variation of pods per plant in dependence of year condition, but no significant differences between numbers of seeds were noted. The split application of Humic complet significantly influence the higher number of pods per plant while keeping the number of seeds per pod.

The yield of seeds was significantly influenced by weather conditions and by used fertilization treatments during evaluated years. Supplemental nitrogen in the vegetative stage of V2 (ammonium nitrate with lime) or Humix complet in split application of total dose 4+4 $l*ha^{-1}$ applied in V2 and R1 stages significantly increased yield of soybean seeds.

Content of protein, oil and fibre was not significantly differing between fertilization treatments. In spite of significantly higher soybean yield in split plot application of Humic complet, content of protein, oil and fibre was at the same level as lower yielded treatments. Split application of Humic complet is effective method for improving yield of soybean seeds while maintaining the composition of protein oil and fibre.

Environmentally sound low level of fertilizers significantly increases the yield of seeds while maintaining content of basic nutrients of soybean.

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