

SZENT ISTVÁN UNIVERSITY

Comparative studies on the effect of farmyard manure and mineral fertilisers on the growth of maize in long-term experiments

Main points of the PhD thesis

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1. BACKGROUND AND AIMS

In modern crop production the main aim up to now has been the maximisation of growth rate and plant yield. Nowadays, however, when the emphasis is on sustainable agriculture, an agro-ecological approach is required, which attaches importance not only to yields, but to the ecological sustainability of the production system. Long-term experiments are an indispensable part of investigations on the effects of crop production systems, treatments and technologies over time (Berzsenyi 2009). It is increasingly accepted that even 10–15 years of trials are insufficient for the reliable detection of changes (Johnston 1988). The long-term experiments set up in Martonvásár by Béla Győrffy in the late 1950s and early 1960s, which are some of the oldest in the country, fully satisfy the methodological criteria (Árendás 1998). The effects of organic and mineral fertilisers on soil fertility have been studied continuously since 1843 in the long-term experiments set up in Rothamsted, UK (Leigh and Johnston 1994). Although the organic matter content of the soil on plots given mineral fertiliser was only about half of that on plots treated with farmyard manure (FYM), the mineral fertiliser treatments nevertheless proved to be better in terms of yield averages (Jenkinson 1991).

Increasing efforts are being made in crop production research not only to measure treatment effects on the basis of the final products, but also to detect changes in the dynamics of photosynthetic production throughout plant growth (Berzsenyi 2000). One important method for examining plant growth and the ecological and agronomic factors that influence it is **growth analysis**. The use of various growth analysis parameters, combined with agronomic, ecological and physiological measurements, makes it possible to evaluate the results of crop production experiments in a scientifically sound manner, based on multiple parameters, as required by current production physiological approaches (Gardner et al. 1985).

The methodology of growth analysis was first elaborated in the 1920s, but at the end of the 1960s the improvements in statistical methods and the spread of computers led to a considerable development both in the methods and in the quality of plant analysis. The introduction of growth analysis into Hungarian research and its use in ecology and crop production can be attributed to Précsényi (1980). Detailed experiments on the dry matter accumulation of maize have been underway in Martonvásár since 1956 (Ferencz 1958, Bajai 1959, Győrffy 1965).

The aim of the present work was to use the results of long-term fertilisation experiments, set up on the principle of active ingredient equivalence in a maize monoculture in 1958, to compare the effect of various levels of farmyard manure (FYM) and mineral fertilisers on:

- 1. the growth of maize plants and the dynamics of growth parameters, using the classical and functional methods of growth analysis;
- the mean and maximum values of growth parameters for individual plants and the whole plant stand;
- the biomass of maize plants, changes in yields and yield components, the nitrogen and chlorophyll contents of maize leaves, and the kernel quality in different years.

The original question raised in the experiments was whether FYM could be replaced by mineral fertiliser if half or all the NPK active ingredients of FYM were supplied in the form of inorganic NPK fertilisers. The results of the long-term experiment were previously published by Győrffy (1979), Berzsenyi and Győrffy (1994) and Berzsenyi et al. (2011). The present paper is a continuation of this work, making use of the results of previous growth analysis research.

2. MATERIALS AND METHODS

2.1. Description of the long-term fertilisation experiment in Martonvásár

The small-plot field experiment on a maize monoculture was set up by Béla Győrffy in the experimental nursery of the Agricultural Research Institute of the Hungarian Academy of Sciences in Martonvásár in 1958. The experiment includes seven treatments, based on the active ingredient equivalence principle (*Table 1*) in seven replications, i.e. in a Latin Square design, on plots measuring $8 \times 10 \text{ m} = 80 \text{ m}^2$. The effects of FYM and mineral fertiliser treatments on the dry matter production and growth parameters of maize were examined in the years 2005–2007.

Table 1. Treatments	in the fertilisation	experiment,	and the	quantities	of active	ingredients
applied. Martonvásár,	, 2005–2007					

Treatments		Active ingredients (kg ha ⁻¹ year ⁻¹)			
		Ν	P_2O_5	K ₂ O	
1	Control	_	_	_	
2	35 t ha ⁻¹ FYM	66	38	75	
3	17,5 t ha ⁻¹ FYM + $N_{1/2}P_{1/2}K_{1/2}$	66	38	75	
4	$N_1P_1K_1$	66	38	75	
5	70 t ha ⁻¹ FYM	132	76	150	
6	$35 \text{ t ha}^{-1} \text{ FYM} + N_1 P_1 K_1$	132	76	150	
7	$N_2P_2K_2$	132	76	150	

2.2. Climatic data of the experimental years

There were very great differences in the rainfall and temperature data of the three years, compared both with each other and with the 30-year mean. After a wet year favourable for maize in 2005, the following year was dry, but could be considered as an average year in terms of rainfall distribution, while the summer of 2007 was extremely hot and dry. The rainfall sum in the vegetation period (Apr.–Sept.) was 526 mm in 2005, 246 mm in 2006 and 265 mm in 2007, while the annual mean temperatures were 9.8°C, 11.0° and 12.8°C, respectively.

2.3. Experimental work

The direct (destructive) and indirect methods of **growth analysis** were applied. Sampling was begun when the maize plants were in the 3–4-leaf stage of development, on the 22–37th day after sowing (depending on the year), and were continued through tasselling and silking until physiological maturity. Samples were taken from all seven treatments in four replications on 10 occasions in 2005 and 2006 and on 9 occasions in 2007, at intervals of 14 days on average.

The plants were analysed individually after division into the following parts: green leaf blade, stalk with leaf husks, tassel, ear husks, ear stalk, cob, and grain yield. The photosynthesising **leaf area** of the plants was measured using ADC AM300 and LI-COR LI-3100A table-top leaf area meters. The separated plant organs were dried at 105°C for 48–72 h in a MEMMERT ULE/800 drying cabinet for the determination of **dry mass**.

The **total leaf area** and the **area of the ear leaf** of the maize plants were recorded after flowering in the plant stand using indirect methods with a LI-COR LI-3000A portable leaf area meter. The **chlorophyll content** of the ear leaf was determined with a portable chlorophyll meter of the Minolta SPAD 502 type at the base of ten ear leaves per plot.

The **total N content** of the whole foliage was determined during the grain-filling period with the Kjeldahl method, using a FIASTAR 5000 spectrophotometer.

The **crude protein, oil and starch contents** of the kernel were recorded using a Perten INFRAMATIC 8600 (NIR) analyser. Harvesting was performed with a Bourgoin plot harvester and the **grain yield** was given in terms of 15% moisture content. The changes caused in the **yield components** (kernel number per ear, thousand kernel weight) by the fertiliser treatments were evaluated from the data of five sample ears per plot.

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2.4. Description of growth parameters and how they were calculated

Growth analysis was performed both for individual plants and for the whole plant stand. Mean, maximum and instantaneous values were recorded for all the growth parameters, and the seasonal dynamics of each parameter was determined. The growth parameters examined for individual plants were: (1) absolute growth rates (AGR, ALGR), (2) relative growth rates (RGR, RLGR), (3) net assimilation rate (NAR), (4) leaf area ratio (LAR), (5) specific leaf area (SLA) and leaf weight ratio (LWR). The growth parameters analysed for the whole stand were: (7) leaf area index (LAI), (8) crop growth rate (CGR), (9) net assimilation rate (NAR), (10) leaf area duration (LAD), (11) biomass duration (BMD) and (12) harvest index (HI).

Both the classical and functional methods of growth analysis were applied in evaluating the growth parameters. When using the **classical method** of growth analysis the mean values of the growth parameters were calculated for the interval between two consecutive samplings from the dry mass and leaf area data determined at each sampling date using the equations given by Précsényi et al. (1976) and Evans (1972). The classical growth analysis program elaborated by Hunt et al. (2002) was used to calculate the mean values of the growth parameters and for the statistical analysis. In the case of the **functional method** of growth analysis, the Hunt–Parsons (1974) growth analysis program (HP model) was applied, which allowed the instantaneous values of the parameters to be calculated. The Hunt–Parsons growth analysis program fits first, second or third degree polynomials to the dry mass of the whole plant (Y) and the total leaf area (Z) as a function of time (X), using the stepwise regression method (Berzsenyi 2000).

2.5. Methods used for the biometric evaluation of the data

Analysis of variance on the data of the Latin square design and the biometric evaluation of the data series were performed using the method of Sváb (1981), with the Microsoft[®] Windows Excel (2003) and MSTAT-C (1991) programs.

Correlations between the grain yield or yield components of maize and the growth parameters were determined by **correlation analysis** using the GenStat 13.1 (2002) statistical program package.

The correlation between the grain yield and the two yield components was evaluated by means of **multiple regression analysis** using the 'Enter' elimination method of the SPSS 11.0 for Windows (2001) statistical program.

The long-term effect of the treatments on the yield between 1958 and 2009 was studied using the **cumulative method** elaborated for long-term experiments (Sváb 1981), analysis of variance, and the multifactorial (AMMI) method of **stability analysis** (Crossa 1990).

3. RESULTS

3.1. Effect of fertiliser treatments and the year on the seasonal dynamics of the dry matter accumulation and leaf area of maize

The effect of the seven fertiliser treatments on the seasonal dynamics of the total dry matter determined using the Hunt–Parsons program could be clearly distinguished in the wet year (2005), while in the dry year (2007) the values of dry matter production obtained in the various treatments varied over a much narrower range (*Fig. 1A*).



Fig 1. Effect of fertiliser treatments and years (2005 and 2007) on the seasonal dynamics of the total dry matter accumulation (A) and leaf area (B) of maize plants using the functional method of growth analysis For treatments, see Table 1.

The HP program used a third-degree exponential function to describe the dynamics of total dry matter incorporation, which did not exhibit a maximum in 2005: growth continued in all the treatments. In the course of statistical

analysis, significant differences between the fertiliser treatments were only observed in favourable years on the basis of values obtained with the classical method, but in all three years in the case of curve fitting.

The HP program fitted second and third degree exponential functions to the dynamics of the growth and decline of the leaf area (*Fig. 1B*). The maximum leaf area values were maintained for a period of five to six weeks in the wet year, while in the dry year the rapid drying down of the foliage due to atmospheric drought had a substantial effect on yield formation. The maximum value of the leaf area was greatly influenced by the year, and significant differences were observed between the years.

3.2. Effect of the year on the distribution of dry matter production between plant organs

In favourable years the dry kernel mass per plant in the period around harvesting was more than twice the dry stalk mass, while in the dry year the kernel mass was hardly any greater than the stalk mass. In the wet year the grain ratio, i.e. HI, was around 50%, while this figure was only 43% in the dry year, so the grain ratio dropped substantially, while that of the stalk and other plant organs (ear stalk, cob, husks, tassel) increased (*Fig. 2*).



Fig. 2. Effect of the year (2005 and 2007) on the dynamics of dry matter distribution between the plant organs, averaged over the fertiliser treatments

3.3. Effect of FYM and mineral fertiliser on the growth parameters of maize plants

According to the Hunt–Parsons model the dynamics of the absolute growth rate of total dry matter could be described with a bell-shaped curve, i.e. the growth rate gradually increased to a maximum (during the leaf development phase) and then declined (*Fig. 3A*). The functional method revealed clear growth dynamics, eliminating the fluctuations observed when using the classical method. In favourable years the AGR_{mean} parameter gave a good description of the fertiliser effects, which differed significantly from each other, while in the dry year no fertiliser effect was observed.



Fig. 3. Effect of fertiliser treatments on the dynamics of the absolute growth rate of (A) total dry matter and (B) leaf area (AGR and ALGR) and on (C) the net assimilation rate (NAR) and (D) the leaf area ratio (LAR) in 2005, using the functional method of growth analysis For treatments, see Table 1.

The dynamics of the absolute growth rate of the leaf area was described by two bell-shaped curves, the first describing the increase in leaf area and the second the drying down stage (*Fig. 3B*). In 2005 drying down was a slower, more uniform process, promoting grain filling. In all the years the fertiliser treatments had a significant effect on the ALGR_{mean} values.

The values of net assimilation rate were almost constant, only reaching a maximum for a short period, irrespective of the year, during the 8–10-leaf stage (*Fig. 3C*). The fertiliser treatments had no significant effect on the NAR_{mean} and NAR_{max} values, based on measured values, while the NAR dynamics could be more precisely described with the functional method, which detected significant treatment effects.

On the basis of the HP model the dynamics of leaf area ratio followed a similar curve, in all the years and treatments, to that revealed by the classical method (*Fig. 3D*). After the initial maximum values there was a gradual decline right up to the end of the vegetation period. On the basis of measured values, both the years and the fertiliser treatments had a significant influence on the LAR_{mean} values.

3.4. Effect of FYM and mineral fertiliser on the growth parameters of the maize stand

In the favourable, wet year the maximum values of leaf area index were significantly different in the individual fertiliser treatments, with the highest values in treatments 6 and 7, the lowest in the control and treatment 2, and intermediate values for treatments 3, 4 and 5 (*Table 2*). In the dry year the LAI_{max} value was only significantly lower in the control treatment, while the values for all the other treatments exhibited no significant differences, i.e. no treatment effect was observed.

The higher rate of fertilisation enhanced the maximum value of the crop growth rate (CGR_{max}), which was almost twice as high in all the fertiliser

treatments in the dry year as in favourable years, rising from approx. 20 g m⁻² day⁻¹ to 40 g m⁻² day⁻¹. Significant differences between the CGR_{max} values were recorded in response to fertiliser treatments, regardless of the year.

The leaf area duration and biomass duration data exhibited similar trends, with the lowest values in the control treatment and the highest at the highest mineral fertiliser rate (treatment 7) in all the years. The LAD, BMD and HI values all gave a good reflection of the fertiliser and year effects. Averaged over the fertiliser treatments, the highest values of these parameters were recorded in the wet year and the lowest in the dry year.

Table 2. Effect of FYM and mineral fertiliser on the growth parameters of the maize stand in 2005

The second second	LAI _{max}	CGR _{max}	LAD	BMD	HI
I reatments	$(m^2 m^{-2})$	$(g m^{-2} day^{-1})$	(cm ² day ⁻¹)	(kg plant ⁻¹)	(%)
1	2,14 c †	13,58 d	155,25 e	10,55 e	48,81 c
2	2,31 c	18,68 c	176,51 e	13,20 d	50,93 bc
3	3,15 ab	20,01 abc	238,47 c	16,44 bc	54,61 ab
4	3,14 ab	22,67 ab	258,12 b	17,57 b	55,24 a
5	2,88 b	19,56 bc	217,43 d	15,31 c	53,57 b
6	3,30 a	22,84 ab	274,51 ab	19,06 a	55,18 a
7	3,35 a	23,12 a	288,03 a	19,62 a	55,07 a
SzD _{treatments}	***	***	***	***	***

Significance levels: ***P=0.1%, **P=1%, *P=5%, ^{NS} = non-significant.

 \dagger Data followed by the same letter within a column did not differ significantly at the LSD_{5%} level based on analysis of variance. For treatments, see Table 1.

3.5. Effect of FYM and mineral fertiliser on the grain yield, yield components and grain quality of maize

In favourable years the fertiliser treatments had a significant effect on the grain yield, yield components and grain quality of maize (*Table 3*). The effect of FYM + mineral fertiliser treatments (treatments 3 and 6) on the yield significantly surpassed that of FYM alone (treatments 2 and 5), but was not as great as that of the equivalent dose of NPK active ingredients in the form of mineral fertiliser alone (treatments 4 and 7). In the dry year the yields achieved with the high fertiliser rate (treatments 6 and 7) did not differ significantly from

those obtained in the control treatment. In this year the significantly highest yield was obtained in treatment 5, i.e. from the application of 70 t ha^{-1} FYM, demonstrating the positive effect of FYM in dry years.

Optimum N supplies and the year effect also made a substantial contribution to trends in yield components. In the wet year the number of kernels per ear in the control treatment was 30% lower than the maximum kernel number obtained in the $N_2P_2K_2$ mineral fertiliser treatment, while in the dry year this difference was only 5.6%. In the dry year there was a 33% drop in the thousand-kernel weight compared to that recorded in the wet year.

The fertiliser treatments had a significant effect on the crude protein content of the grain, and a difference was also observed between the lower and higher fertiliser rates in favourable years. The highest grain protein content was found at the higher rate of NPK mineral fertilisation (treatment 7; 8.96–9.39%), regardless of the year.

Treatments	Grain yield	Seed	Tousand	Grain protein	Grain oil	Grain starch
	(t ha ⁻¹)	number / ear	grain weight (g)	content (%)	content (%)	content (%)
1	4,26 e †	272,67 e	262,02 d	6,28 de	3,55 a	70,16 b
2	5,96 d	325,19 d	267,52 d	6,00 e	3,53 a	71,75 a
3	7,67 b	413,57 bc	333,23 b	6,85 bc	3,46 ab	70,58 ab
4	7,97 b	420,76 abc	328,27 b	7,36 b	3,48 ab	70,23 b
5	6,81 c	405,67 c	303,84 c	6,70 cd	3,51 ab	70,26 b
6	9,22 a	435,71 ab	356,38 a	8,94 a	3,41 bc	68,08 c
7	9,82 a	440,29 a	366,81 a	9,39 a	3,35 c	67,79 c
SzD _{treatment}	***	***	***	***	**	***

Table 3. Effect of FYM and mineral fertiliser on the grain yield, yield components and grain quality of maize in 2005

Significance levels: ***P=0.1%, **P=1%, *P=5%, ^{NS} = non-significant.

 $^{+}$ Data followed by the same letter within a column did not differ significantly at the LSD_{5%} level based on analysis of variance. For treatments, see Table 1.

3.6. Effect of FYM and mineral fertiliser on the leaf nitrogen and chlorophyll contents

A close correlation was detected between the SPAD indexes obtained for chlorophyll measurements on the plant stand and the total nitrogen contents determined in the laboratory for the various fertiliser treatments, based on results averaged over the years (*Fig. 4A, B*). The chlorophyll and nitrogen contents of the leaves of maize plants were significantly influenced by the fertiliser treatments in all the years.



Fig. 4. Chlorophyll content (A) and N content (B) of maize leaves in response to fertiliser treatments (1–7), averaged over the three years (2005–2007) Error bands: at the LSD_{5%} level. For treatments, see Table 1.

3.7. Correlation analysis on the yield, yield components and growth parameters

On the basis of Pearson's correlation coefficient, a very close positive correlation (P=0.1%) was found between the yield and the values of dry matter_{max}, kernel number per ear, thousand-kernel weight and HI. The partial correlation is a stricter criterion, on the basis of which a close positive correlation (P=5%) was only found between the yield and the values of dry matter_{max} and thousand-kernel weight. Both matrixes indicated very close or close positive correlations between CGR and the values of LAI and NAR (P=0.1, 1 and 5%).

Analysis using the 'Enter' method indicated that the two yield components were responsible for 62 and 59% of the grain yield in the wet years $(R^{2}_{2005} = 62.3\%; R^{2}_{2006} = 58.8\%)$, while in the dry year neither the thousandkernel weight nor the number of kernels per ear had a significant effect on the yield $(R^{2}_{2007} = 4.5\%)$. Analysis averaged over treatments and years showed that the effect of the thousand-kernel weight on the yield ($\beta = 0.721$) was approx. 3.75 times as great as that of the kernel number per ear ($\beta = 0.192$).

3.8. New scientific results:

- 1. In the 51-year-old fertilisation experiment carried out in Martonvásár, the Hunt–Parsons (HP) growth analysis model used a third-degree exponential function (ln W = $a + bx + cx^2 + dx^3$) to describe the effect of fertiliser treatments on the total dry matter production and the grain yield, and second- and third-degree exponential functions (ln Z = $a + bx + cx^2$ and ln Z = $a + bx + cx^2 + dx^3$) for the change in leaf area.
- 2. The growth of maize plants as a function of fertiliser treatments and years was clearly described by the instantaneous values and dynamics of the absolute growth rate (AGR), absolute leaf area growth rate (ALGR), leaf area ratio (LAR), leaf area index (LAI), crop growth rate (CGR), leaf area duration (LAD), biomass duration (BMD) and harvest index (HI) parameters, calculated using the HP model.
- 3. The mean values of parameters computed using the calculator devised by Hunt et al. (2002) gave a similarly good description of the effect of fertiliser treatments and years, and of significant differences.
- 4. The combined analysis of the 51 years (1958–2009) showed that the effect of the FYM + NPK mineral fertiliser combination on the yield did not differ significantly from that of NPK mineral fertiliser alone. The smallest yield stability was found for the control treatment and the high dose of

NPK fertiliser alone, and the greatest for the low fertiliser rates and for 70 t ha^{-1} FYM.

- 5. The significantly highest yields in the experimental years (2005–2007) were recorded in the wet years when half or all of the 70 t ha⁻¹ rate of FYM was replaced by NPK mineral fertiliser (9.22 and 9.82 t ha⁻¹), and in the dry year for the lower rate of fertilisation (3.01–3.18 t ha⁻¹) and when 70 t ha⁻¹ FYM was applied (3.35 t ha⁻¹). The data confirmed the yield-enhancing effect of FYM in dry years.
- 6. The crude protein content of the grain and the chlorophyll and total N contents of the leaf were significantly increased by the higher rate of fertilisation, and were also influenced by the year.
- 7. The Hunt-Parsons growth analysis model made it possible to give an accurate description of the dynamics of dry matter production in the whole maize plant and in various plant organs and of the increase in leaf area, both throughout the whole growth period and during various stages of growth.
- 8. Pearson's correlation coefficient and multiple regression analysis revealed a significant positive correlation between the grain yield and the yield components, and between the grain yield and both the maximum value of dry matter production and the harvest index. Correlation analysis demonstrated close correlations between the crop growth rate (CGR) and its components: the leaf area index (LAI) and the net assimilation rate (NAR).

CONCLUSIONS AND RECOMMENDATIONS

The analysis of the year effect primarily demonstrated the yield-limiting effect of rainfall deficiency (yield decreases of 4.91 and 2.91 t ha⁻¹ in 2007, compared with 2005 and 2006), which meant that the effect of the experimental treatments was less evident, if at all, in the yields, though the year effect could be clearly described in terms of the dynamics of dry matter production and the leaf area index. A combination of FYM and NPK mineral fertiliser ensures both high yields and satisfactory yield stability. In the dry year the fertiliser treatments had no significant effect on either the grain yield or the yield components. It was found, however, that the year effect caused significant changes both in the grain yield and yield components and in the leaf chlorophyll content. The results proved that in dry years a high rate of mineral fertiliser resulted in severe yield depression, while the lower rate of mineral fertiliser gave reliable yields, less dependent on the weather.

In agreement with earlier results, it was shown that the SPAD-502 chlorophyll meter is a suitable instrument for the characterisation of maize plant N supplies during the grain-filling period. It was proved that the crude protein content of maize kernels was significantly lower in years with high yields, averaged over the fertiliser treatments (7.35%), than in dry years (8.00%), and that outstanding grain protein contents could be achieved with high rates of mineral fertiliser even in long-term monocultures.

The regular determination of dry matter production (every 14–21 days) allowed the growth dynamics of the maize plants to be compared in the various treatments over the whole of the growth period. Using the Hunt–Parsons growth analysis program, significant differences were detected between the various levels of FYM and mineral fertiliser applied in the experiment and between the years. The growth dynamics of maize in response to treatments and years could best be described using the absolute growth rate (AGR), the crop growth rate

(CGR) and the leaf area index (LAI). In agreement with data in the literature, the use of the functional method of growth analysis revealed significant differences in the values of the net assimilation rate (NAR) between both the years and the fertiliser treatments, while only the year effect was found to be significant using the classical method. When analysing the leaf area ratio (LAR) the treatment effects could not be distinguished any better by fitting functions than with the classical method.

It became clear from the results that the use of the classical and functional methods of growth analysis provided a reliable evaluation of the effects of fertiliser treatments and years on the growth of maize and on the mean and maximum values of growth parameters during the vegetative growth period. Growth analysis research reveals new correlations and provides data for simulation models and precision crop production. The scientific analysis of the growth dynamics and agronomic responses of maize hybrids could make a great contribution to scientifically-based, environment-friendly, efficient maize production.

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- **Micskei, G**., Jócsák, I., Berzsenyi, Z.: 2009. Az istállótrágya és a műtrágya hatása a kukorica növekedésére és növekedési mutatóinak dinamikájára, eltérő évjáratokban. (Effect of farmyard manure and mineral fertiliser on the growth and the dynamics of the growth parameters of maize in different years.) *Növénytermelés.* 58 (4): 45-56.
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