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THE ROLE OF ORGANIC FERTILIZER AND AMINO ACID (PROLINE) IN THE GROWTH AND PRODUCTION OF CAYENNE PEPPER PLANTS (*CAPSICUM ANNUUM*. L) AND THE ACCUMULATION OF THEIR MEDICINAL COMPOUNDS (CAPSAICIN AND DIHYDROCAPSAICIN) UNDER WATER STRESS CIRCUMSTANCES

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Abstract

The study was conducted in the fields of Al-Dujaila-Kut for the two seasons 2014-2015, in order to study the effect of organic fertilizer and amino acid (proline) in the growth and production of cayenne pepper plants and the accumulation their medicinal compounds (Capsaicin and Dihydrocapsaicin) in the fruit of hybrid cayenne pepper (Ampala) under water stress circumstances. The experiment was conducted according to Randomized Complete Blocks Design (RCBD), with three replicates and the averages were compared according to L. S.D. At the 5% probability level. The experiment included 10 treatments, a combination of the addition of organic manure (poultry manure) to the soil at a rate of 5 and 10 tons.h^{-1} and the treatment of 100% and 150 mg.L^{-1} of proline by alone or in combination with half of the recommended chemical fertilizer for crop within 100% and 50% field capacity. The treatment of T_{10} showed significantly superiority over the rest of the other treatments in both the dry weight of the root system and the reduction of nitrate levels and the accumulation of medicinal compounds (Capsaicin and Dihydrocapsaicin) in the fruits for both seasons. occupied the second level in an increase of the total vegetative and total yield of fruits after the treatment of chemical fertilization, with non-significant differences in both seasons. We conclude from this that the efficiency of using the T_{10} combination in the total yield is equivalent to the efficiency of mineral fertilization, as well as giving good quality fruits free of chemical pollutants (nitrates) and The accumulation of medicinal compounds in their fruits.

Key words : Organic fertilizer, Amino Acid, *Capsicum annuum*. L

Introduction

Cayenne pepper (*Capsicum annuum.L*) is one of the most important commodities in international trade (Schaumann and Thiele-Bruhn, 2011) not only economically but also because of its various preventive and curative uses. The fruit contains a group of alkaloids called Capsaicinoids, most notably capsaicin ($C_{18}H_{27}NO_3$) in terms of quantity of 69% and Dihydrocapsaicin about 22% of the total mass of Capsaicinoids (Supalkova *et al*., 2007), with medical importance to its numerous physiological effects on human health as antioxidant such as treating some cancers and treating rheumatism, as well as in weight loss by burning fat and lowering cholesterol. Making its fruit used in many foods and food industry in many countries of the world. The problem of water stress or drought is one of world environmental problems that limit the production of different crops, threaten world food security, and the climate changes and the resulting increases in the temperature of the atmosphere, which led to increased frequency of dry cycles, its Intensity, which increased the seriousness of the problem. Plants are exposed to a variety of non-vital stresses such as drought. Drought stresses cause oxidative stress within the plant's cells due to the high leakage of electrons towards Molecular Oxygen (O_2) during the metabolic processes of photosynthesis and respiration, which increases the concentrations of reactive oxygen (ROS), these free radicals can directly attack cell components such as cellular membranes, nucleic acids, proteins, lipids and plant pigments, thus exposing the cell to death (Cui and Zhao, 2011). In order to resist drought and high levels of ROS, the plants have developed a defense system that limits the high evaporation rate and the oxidative effects of the ROS group. The components of this system include enzymatic antioxidants, including the medical capsinicoid and proline (Gill and Tuteja, 2010). The chemical fertilization rates used for growing vegetables compared to other crops have increased because they can be grown in more than one season per year, which has exacerbated and increased the adverse effects on health and the environment, in particular the remaining effect of nitrates, which is one of the most dangerous compounds to human health (Zandonadi *et al*., 2013), this also led to the weakness of the Iraqi soil in its construction because its low content of organic matter, which does not exceed 1% for high temperatures, and lack of rainfall, It is difficult to increase the proportion of organic matter in Iraqi soil without human intervention. The conservation of appropriate levels of soil organic substance is aim in itself because of its importance in agricultural production and Soil maintenance through its role in improving physical, chemical and biological soil properties (Sartip *et al*., 2015). The statistics indicated that 25% of the soil of Asia and Africa and 20% of the soil of the continent of Europe and 5% of the soil of the North American continent has deteriorated over the last 50 years (Bongaarts, 1994). Therefore, the current agricultural system is described in many countries of the world that it does not have the status of permanence and low productivity over time (Bationo *et al*., 1997). Therefore, the current agricultural system is described in many countries of the world that it does not have the status of permanence and low productivity over time (Bationo *et al*., 1997). As a result of these adverse effects of the use of chemical fertilizers and the phenomenon of drought, Attention has been paid in many countries to encourage the use of compost to meet some of the needs of the global market of products After having identified problems caused by traditional products In addition to what the availability of organic agriculture of improve the physical and chemical of soil traits so as to maintain soil moisture and the availability of mineral elements for the longest period Possible for plant, which led to the expansion of cultivated areas and the increase in organic production to the degree of its competition for conventional production in the world markets, especially developed countries US organic food market revenues were 1 billion $ in 1994 and 13 billion $ in 2003. For the importance of cayenne pepper crop, the idea of its production came accordance with the organic farming system, which is an indispensable aspect in maintaining soil fertility and supplying developing plants with the need of various nutrients under the prohibition of the use of random chemical fertilizers (Arun, 2001), Based on the above, and the position occupied by the pepper crop in the open and protected crops in our country and the lack of studies around it, this study came to test:

1. Rationalization in the use of chemical fertilizers to obtain good quality fruits free of chemical pollutants (nitrates).
2. The role of organic fertilizer and amino acid (proline) in reducing the effects of water stress.
3. The accumulation of Capsaicin and Dihydrocapsaicin in fruits.

Materials and Methods

The experiment was conducted in one of the special fields of Al-Dujaila area in Wasit Province during the spring season 2014-2015 to study the effect of organic fertilizer and the amino acid proline in the quantity and quality of cayenne pepper product under the water stress condition. Table 1 shows the physical and chemical properties of the soil before cultivating, As analyzed in the analytical laboratory of the Department of Chemistry at the University of Baghdad.

Table 1: Physical and chemical traits to the soil of the experiment field before cultivating for both seasons.

Physical and chemical traits to the soil		seasons		measuring unit
		2014	2015	
Soil separates	Clay	372	352	$g.kg^{-1}$
	Silt	400	452	
	Sand	188	198	
O. M		10.6	15.2	-
E. C		2.3	2.8	$ds.m^{-1}$
pH		7.64	7.28	-
Availability of nutrients elements	N	66.6	70.02	$mg.kg^{-1}$
	P	20.11	17.5	
	K	221.0	210.4	
	Cl	14	12	$mg.kg^{-1}$
	Mg	0.5	2.4	
	Na	169.8	117.6	
	Ca	26.5	10.5	
Soil Texture	sandy clay loam			

* Analysis was conducted in the laboratories of the Department of Soil and Water Sciences - college of Agriculture - University of Baghdad.

After the field was divided into a plot and calibrated, cayenne pepper (Ampala) were cultivated, it is a hybrid produced by the French company (Clause), at the line of calibration and 40 cm between seedlings and other (Boras *et al*., 2011) and the cultivation date in 10/3 and 12/3 for the seasons sequentially, and the experiment was terminated on 8/1 for both seasons. After preparing the soil, a drip irrigation system consisting of the main irrigation pipe with diameter of 5 cm and the sub-tubes of 2.5 cm diameter (3 meters away from each other and identical to the dimensions between the sectors) and the field pipes (bearing the dotted) with diameter of 1.3 cm, while the distance between dotted 40 cm. The experimental land was divided into 36 experimental units, dimensions of each 4 x 3 m^2,each experimental unit included 4 furrows, each furrow had 9 plants, and the distance between plant and another 40 cm, was left 1 m between experimental units as a insulator to prevent mixing between the treatments, the experiment was conducted using Randomized Complete Block Design (R.C.B.D) with three replicates and 36 experimental units. Randomized randomized whole-scale randomized. The averages were compared using a less significant difference of LSD at the probability level of 0.05 (Alsahoki and Wahib, 1990).Treatments were as follows:

- T_1 - Irrigation level 100% Field capacity only.
- T_2- 100% recommended mineral fertilizer + irrigation level 100% field capacity.
- T_3- 50% recommended mineral fertilizer + irrigation level 100% field capacity.
- T_4- poultry manure 5 $tons.h^{-1}$ + level of irrigation 100% field capacity.
- T_5 - Poultry manure 10 $tons.h^{-1}$ + level of irrigation 100% field capacity.
- T_6 - add 100 $mg.L^{-1}$ Proline + irrigation level 100% field capacity.
- T_7 - Poultry manure 5 $tons.h^{-1}$ 50% + mineral fertilizer + addition of 100 $mg.L^{-1}$ Proline + irrigation level 50% field capacity.
- T_8 - Poultry manure 5 $tons.h^{-1}$ + 50% mineral fertilizer + add 150 $mg.L^{-1}$ Proline + irrigation level 50% field capacity.
- T_9- Poultry manure 10 $tons.h^{-1}$ 50% + mineral fertilizer + addition of 100 $mg.L^{-1}$ Proline + irrigation level 50% field capacity.
- T_{10} - Poultry manure 10 $tons.h^{-1}$ + 50% mineral fertilizer + add 150 $mg.L^{-1}$ Proline + irrigation level 50% field capacity.

In the country, especially the province of Wasit significant amounts of poultry manure, which was used to prepare the organic manure by air fermentation method on 21/11/2011, one tons of poultry manure was prepared after it was fully soaked with spraying with soil suspension to accelerate the process of microbial decomposition, Then the heap was covered with nylon perforated, and the heap was then stirred and moistened every 10 days until the end of the fermentation process. Table (2) shows the chemical traits of the organic fertilizer after decomposition. Poultry manures with a depth of 15 cm were added homogeneously to the soil and the addition was before 10 days of cultivating. The level of amino acid (proline) (100 and 150 $mg.L^{-1}$) was added by spraying 6 times, the first level was added after one month of transfer of the seedlings to the field and the second addition was after the first addition by 15 days and so the rest of the additives, Spraying was done early in the morning and make sure the leaves are fully wet. The recommended mineral fertilizer (160 kg N + 160 kg P_2O_5 + 144 kg $K_2O).h^{-1}$ [9], Use diammonium phosphate fertilizer (DAP) fertilizer was used: K: P: N (46: 46), fertilizers of urea (46)% N and potassium sulphate fertilizer (50% K_2O). The addition was on two batches in a feeding method, the first after 20 days of implantation and the second was one month after the first addition.

Table 2: Chemical traits of organic fertilizer (poultry manure) after decomposition.

Traits	Unit	Poultry manure	
		2014	2015
EC	$ds.m^{-1}$	2.58	2.32
pH	-	6.5	6.4
Organic Carbon	$g.kg^{-1}$	321	356
Total nitrogen	$g.kg^{-1}$	31	35
N / C ratio	-	10.35	10.17
Total phosphorus	$g.kg^{-1}$	17.2	18.1
Total potassium	$g.kg^{-1}$	23.1	24.2

* Organic fertilizers were analyzed in laboratories of the Water Treatment Department of the Ministry of Science and Technology

The irrigation process was conducted on the basis of moisture depletion for depth of 0 -20 cm from agriculture until the vegetative growth stage, then the depth of irrigation water was increased on the basis of moisture depletion for depth 0-30 cm to the beginning of the flowering stage, then increased the depth of irrigation water also on the basis of moisture depletion for depth of 0-40 cm to the end of the physiological maturity to reach moisture content approaching the field capacity, So that the Moisture weight is measured and then turned to the depth of irrigation water according to the stress used in the experiment and irrigation when it loses 50% of the availability water, Where the weigh method was used sampling by soil driller more than two days ago to determine soil moisture content for irrigation purposes according to treatments. The availability water was determined by the difference between the humidity at the two stresses of 33 and 1500 KPa. In order to calculate the amount of added water according the treatments to compensate moisture depletion at the field capacity, the following equation (Sutcliffe, 1979) was used:

$$\mathbf{d = [\theta_{f.c} - \theta_{bi}]\ D}$$

where

d = depth of added water (cm)

$\boldsymbol{\theta_{f.c}}$ = volumetric humidity at field capacity.

$\boldsymbol{\theta_{bi}}$ = volumetric humidity before irrigation,

D = Soil depth at the desired root system (cm).

At the end of the harvest season, 20 plants were randomly selected from each experimental unit to study the dry weight of the vegetative system and the root system ($g.plant^{-1}$) by weight method, after placing it in an electric oven under 65 m for 72 hours and weight after stabilizing using a sensitive balance, Nitrate was measured using method (Cataldo *et al*., 1975). Capsaicin and Dihydrocapsaicin were estimated by the spectrometer of the blue solution resulting from the preparation method (Ademoyegun *et al*., 2011) and by total production according to the following equation:

$$\text{Total production (tons.h}^{-1}) = \frac{\text{Experimental unit yield} \times \text{Area of hectare}}{\text{Area of experimental unit}}$$

Results and Discussion

The results indicated that the treatment T_{10} (poultry manures 10 tons.h^{-1} + 50% fertilizer + 150 mg.L^{-1} proline + irrigation level 50% field capacity) was excelled in most traits studied for both seasons compared to treatment (Irrigation 100% field capacity only) their effect on productivity was close to the effect of mineral fertilization. The differences were insignificant between the two treatments (T_2). The results showed in Table (3) that the treatment of T_{10} was significantly excelled than the other treatments in giving the highest dry weight of the root system of 37.54 and 35.59 g.plant^{-1}, respectively. However, the differences were non-significant between them and between the two treatments T_2 and T_5, which gave 34.24 and 32.96 g.plant^{-1}, respectively in the first season and the treatment of T_2 in the second season, which gave 33.52 g.plant^{-1}, as the results show in the same table, the treatment of mineral fertilization (T_2) has achieved the highest dry weight of the total vegetative reached 222.7 and 213.4 g.plant^{-1}with a significant increase on other treatments except for the T_{10} treatment, which gave 211.5 and 212.4 g.plant^{-1} for both seasons respectively, while the lowest value in the treatment of T_1 (irrigation 100% field capacity only) for both seasons was 108.2 and 85.71 g.plant^{-1}, respectively. The results of Table (4) indicate that the treatment of mineral fertilizer T_2 significantly excelled the other factors by giving the highest total production of 21.60 and 21.13 tons.h^{-1} for the two consecutive seasons except T_{10} and T_4 for both seasons, with productivity at 21.50 and 21.32 tons.h^{-1}, respectively for the first season, 20.88 and 20.36 tons.h^{-1} respectively for the second season, while the lowest was obtained at treatment T_1 (irrigation 100% field capacity only) amounted to 8.82 and 8.04 tons.h^{-1} for consecutive seasons. Table (4) show a decrease in the nitrate ratio in fruits for plants treated with T10 treatment reached to 66% and 52.5% respectively the two seasons compared to the treatment of mineral fertilization, which reached the proportion of nitrates 0.906 and 1.046 mg.g^{-1}. Table (5) shows that there was a significant effect of treatment T_{10} by giving the highest value of capsaicin reached 403.7 and 392.3 mg.kg^{-1} for the two seasons respectively, with a significant increase of 35.9% and 17.4% respectively for the treatment of mineral fertilization. The same treatment (T_{10}) was excelled by giving the highest dihydrocascaicin increase of 149.3 and 145.5 mg.Kg^{-1} for two seasons respectively, with a significant increase of 31.2% and 15.3% respectively for the treatment of mineral fertilization.

Table 3: Effect of the addition of organic fertilizer and amino acid (proline) in the dry weight rate of the total vegetative and root under the condition of water stress.

Treatments	Dry weight for the vegetative		Dry weight for the root system	
	First season	Second season	First season	Second season
T_1	108.2	85.71	20.48	20.60
T_2	222.7	213.4	34.24	33.52
T_3	161.1	166.2	27.91	24.26
T_4	189.6	177.2	29.56	28.85
T_5	200.5	188.1	32.96	33.12
T_6	177.1	169.9	25.50	21.75
T_7	152.6	169.2	24.35	22.77
T_8	168.6	141.6	21.84	20.59
T_9	179.9	182.0	24.44	25.84
T_{10}	211.5	212.4	37.54	35.59
LSD 0.05%	11.75	7.53	5.11	2.41

Table 4: Effect of the addition of organic fertilizer and amino acid (proline) in the total yield (tons.h^{-1}) and the concentration of nitrates in the fruit (mg.g^{-1}) under the water stress condition.

Treatments	Total yield (tons.h^{-1})		Nitrates (mg.g^{-1})	
	First season	Second season	First season	Second season
T_1	8.82	8.04	0.716	0.686
T_2	21.60	21.13	0.906	1.046
T_3	15.79	15.77	0.796	0.826
T_4	21.32	20.36	0.806	0.766
T_5	18.50	18.73	0.836	0.796
T_6	14.40	14.06	0.856	0.736
T_7	18.87	18.79	0.606	0.696
T_8	18.28	18.80	0.716	0.726
T_9	19.91	18.84	0.626	0.696
T_{10}	21.50	20.88	0.546	0.686
LSD 0.05%	1.82	1.77	0.230	0.295

The results were compared between the treatment of mineral fertilization and the treatment of T_{10} (poultry waste 10 tons.h^{-1} + 50% mineral fertilizer + addition of 150 mg.L^{-1} proline + irrigation level 50% field capacity) despite the irrigation level of 50% field capacity and the addition of 50% of the mineral fertilizer, has excelled on the rest of the treatments in most of the studied traits, due to the role of organic fertilizer in improving physical, chemical and biological soil traits (Ullah *et al*., 2008) and increasing availability and the availability of many nutrients and the important role of these elements as they are involved in many processes The activities of phylogeny and vitality, or stimulate them to do, which are related to the process of photosynthesis and food manufacturing in the plant (Schaumann and Thiele-Bruhn, 2011). As well as stimulate cell division and elongation on the one hand, And to the role of water stress in stimulating the root to deepen and spread in an attempt to obtain sufficient water and elements, this is one of the images of plant adaptation to bear drought, Thus forming a good and efficient root mass capable of absorbing water and mineral elements and transferring them to the upper parts of the plant (Suganya and Sivasamy, 2006). In addition, the role of proline as an osmosis regulator reduces the water stress in plant leaves and thus reduces the evaporation rate and as a result the plant's ability to resist moisture stress and continue to perform its various physiological functions. (Vandenbroucke *et al*., 2013). Therefore, Which affect in the increase of vegetative growth represented by the branches number of the plant, leaf area and leaf content of chlorophyll, which increases the carbon representation and materials accumulated in the plant such as carbohydrates and proteins and then increase the dry weight of the vegetative and root system (Zhao *et al*., 2017), which reflected positively on the increase of total yield.

Table 5: Effect of adding organic fertilizer and amino acid (proline) in fruit content of Capsaicin and Dihydrocapsaicin (mg.kg^{-1}) under water stress condition.

Treatments	Capsaicin (mg.kg^{-1})		Dihydrocapsaicin (mg.kg^{-1})	
	First season	Second season	First season	Second season
T_1	322.7	340.8	122.3	128.3
T_2	297.1	334.3	113.8	126.2
T_3	303.1	346.1	115.8	130.1
T_4	295.7	339.9	113.3	128.0
T_5	297.1	341.2	113.8	128.5
T_6	354.4	346.7	132.9	130.3
T_7	323.9	360.7	122.7	135.0
T_8	394.1	360.6	146.1	134.9
T_9	334.1	368.0	126.1	137.4
T_{10}	403.7	392.3	149.3	145.5
LSD 0.05%	30.22	N.S	15.65	12.85

The increase in Nitrate ratio in T_2 treatment is due to the fact that the nitrogen sources added to the soil are converted into nitrate and in a few days when the environment conditions are favorable for the nitration process and the use of the process is not used. Increasing chemical fertilization does not only increase nitrate levels in the plant but also increases the ratio of free amino acids and oxalate (Hemanth and Kambiranda, 2011). Or the reason for the low nitrate ratio in the treatment of T_{10} to organic manure and the production of humic acids (the humic acid and Fulvic acid) increase the absorption of nutrients in a balanced manner, including nitrogen by the plant, allowing good growth of the plant without any accumulation of any material limits In the plant (IFOAM, 2006). Since alkaloids are nitrogen compounds, the superiority of the T_{10} treatment in giving the highest value of Capsaicin and Dihydrocapsaicin is attributed to the role of the treatment in the formation of a good and efficient root system capable of absorbing elements, including nitrogen, or perhaps exploiting the amino acid (proline) to provide the nitrogen element Which enters the formation of alkaloids (Aniszewski, 2015). Or perhaps due to the effect of water stress (50% field capacity), which increased the amount of Capsaicin and increased its quantity by 3.8 times compared with plants not exposed to water stress (Sung et al, 2005). This may be due to the exposure of plants to stress (Water, thermal, photovoltaic) results in the production of Reactive oxygen species (ROS) or free radicals such as (O^{-2}, H_2O_2, O_3, O^-) which is a very powerful oxidizing agent and rapidly attacks biomolecules such as DNA molecules, leading to severe malfunction in the processes of reactive oxygen species (ROS). Metabolism and dysfunction cannot be repaired or compensated. In order to remove the poisonous effect from the ROS reactions, the plant produces anti-oxidant (alkaloids) compounds which is enzymatically or non-enzymatically (Sartip *et al*., 2015). This is confirmed by (Sung et al, 2005). that there is an increase in the activity of enzymes of Phenylalanine ammonia-lyase (PAL) and cinnamic acid-4-hydroxylase (C_4H) as well as increased activity of the enzyme compound Capsaicinoid synthetase (CS) Capsaicinoids as shown in Figure (1), which were 1.45-1.58 times that of plants not exposed to water stress. These results are consistent with (Cazares and Garay, 2011) that the buildup of alkaloids is inversely proportional to the amount of water added, or the reason for the increase is that the exposure of the plant to the stress conditions leads to a decrease in the content of phosphorus, which inhibits the reactions of the cycle Leads to the accumulation of pyrovic acid (Sartip *et al*., 2015), which is the base material in the biological synthesis of the Valine compound responsible for the first pathway of the biosynthesis of Capsaicin as shown in Figure (2).In addition, water stress leads to disturbances in cellular food transitions. For example, the rate of destruction (Sartip *et al*., 2015), Leading to increase in Phosphoenolpyruvate production, which helps the Shikimate path as shown in Figure (3) when interacting with Erythrose 4-phosphate to produce Chorismate

(Swer *et al*., 2011), a key to the formation of Phenylalanine as shown in Figure (4) responsible for the second pathway of the biosynthesis of Capsaicin and Dihydrocapsaicin as shown in Figure (1).We conclude from the above that the processes containing organic fertilizer, all of which have been characterized by particular treatment of T10 (poultry manure 10 tons.h^{-1} + 50% mineral fertilizer + addition of 150 mg.L^{-1} proline + irrigation level 50% field capacity) by giving it the lowest level of Nitrate and highest level in Capsaicin and Dihydrocapsaicin in fruits compared with the treatment of mineral fertilization, and this is very important to reduce chemical residues harmful to health and pollution of the environment when high additions of fertilizers. The treatment T10 was characterized by giving an economic value of good quality equivalent to the value when using mineral fertilization.

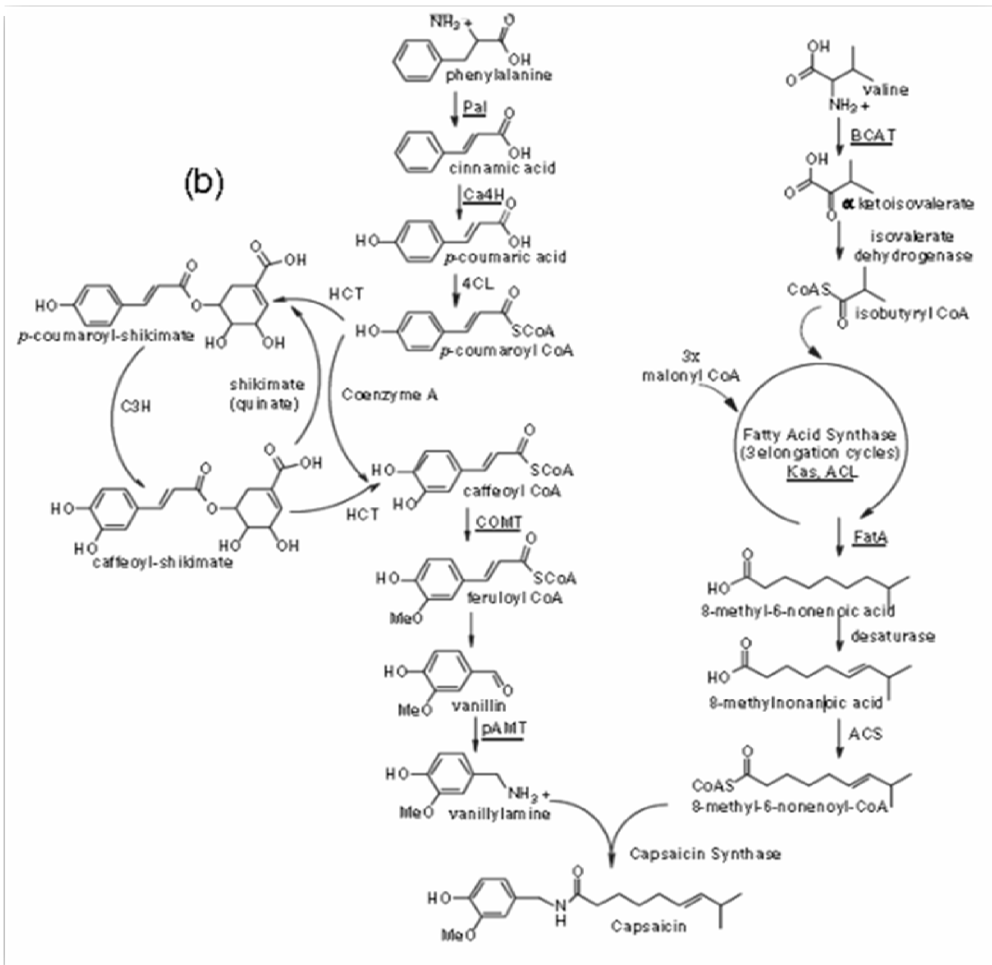

Fig. 1: illustrates the pathway of the biological construction of Capsaicin in the fruit of the cayenne peppers [22].

Phenylpropanoid = Phenylalanine
Phenylalanine ammonia –lyase =PALcinnamic acid-4- hydroxylase = Ca4H
4CL = 4-coumarate CoA ligase HCL = hydroxycinnamoyl transferas
ρ-coumaric acid-3-hydroxylase = C3Hcaffeic acid-o-methyltranferase = COMT
ρAMT = aminotransferase BCAT = branched-chain amino acid transferase
KAS = ß-ketocyl ACP synthase ACL = acyl carrier protein
FAT = acyl- ACP thioesterase synthaseACS = acyl- CoA
Ca3H= coumaricacid 3- CS = capsaicin synthase
HCHL= hydroxycinnamoyl-CoA hydratase/lyase

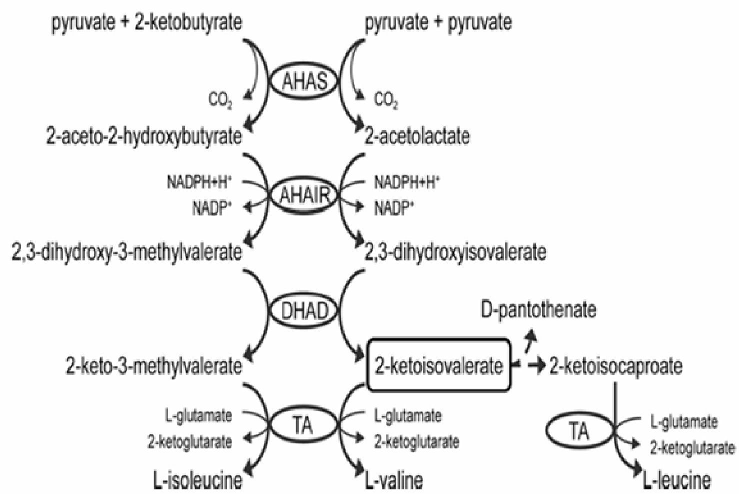

FIG. 1. 2-Ketoisovalerate, L-valine, and L-isoleucine synthesis from two molecules of pyruvate or from pyruvate plus 2-ketobutyrate, respectively, in *C. glutamicum*. Abbreviations: AHAS, acetohydroxyacid synthase; AHAIR, acetohydroxyacid isomeroreductase; DHAD, dihydoxyacid dehydratase; TA, transaminase B.

Fig. 2: illustrates the pathway of the vital construction of Valine [11].

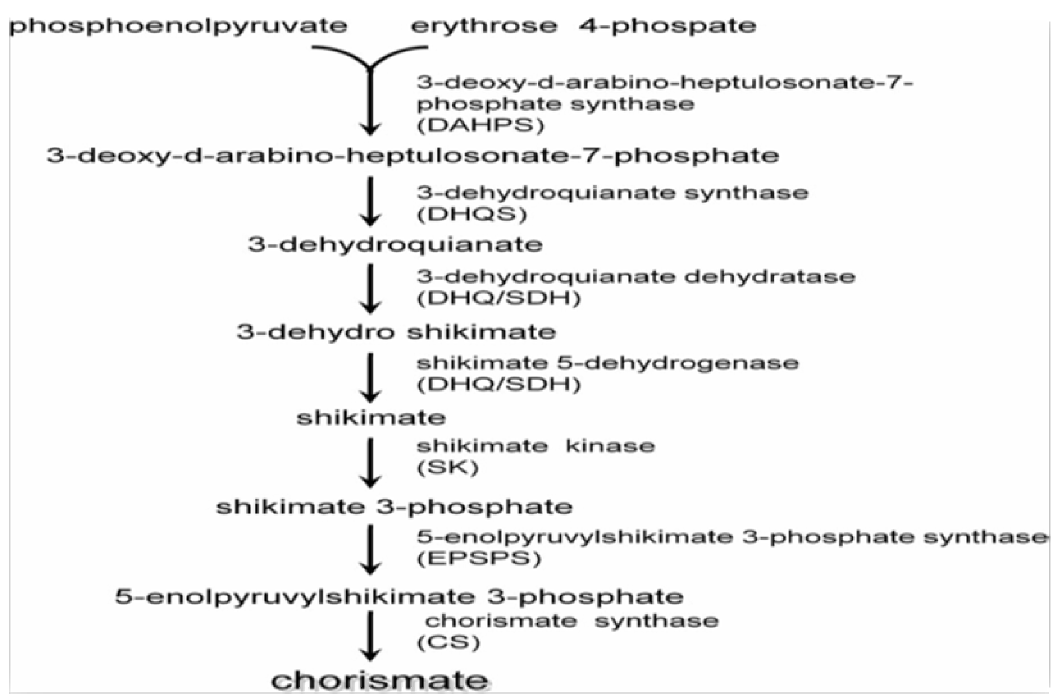

Figure 3: illustrates the dynamic construction path of Shikimate [24].

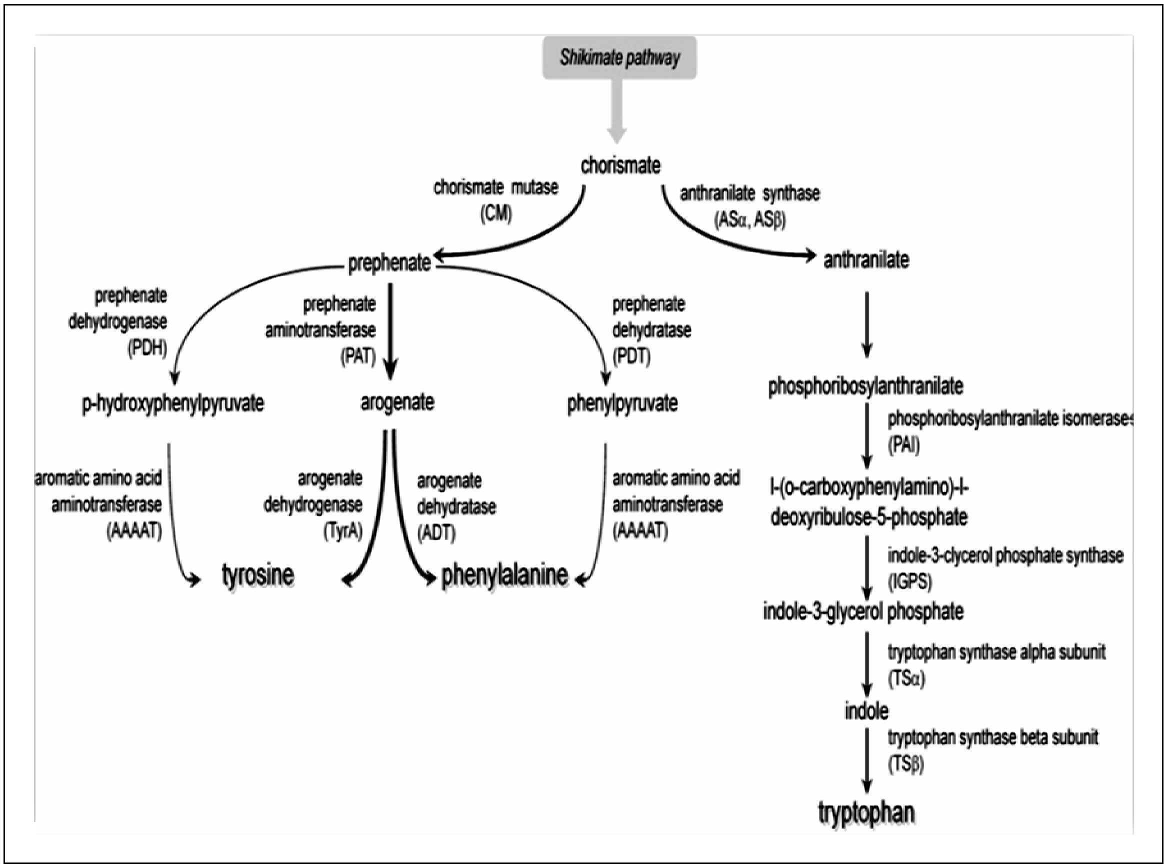

Fig. 4: illustrates the pathway of phenylalanine biosynthesis through the Shikimate path [24].

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