

# Lavender Association of Western Colorado Specialty Crop Grant FY 2012 Research Results and Findings of Lavender Essential Oils Produced by Western Colorado Growers

English lavender (*Lavandula angustifolia*) and the hybrid lavandin (*L. x intermedia*) cultivars have been successfully grown and harvested in western Colorado for 10 or more years. While the worldwide market for the essential oil of lavender is substantial, no research has been conducted in Colorado to determine the yield or quality of oil of the various cultivars available for production in Colorado.

This research project was designed to answer:

- which cultivars produce the largest quantity of essential oil;
- which cultivars produce the highest quality of essential oil;

We also researched and compiled information on:

- the average yield of dried buds per variety; and
- winter survival per variety.

#### **Essential Oil Research**

The Lavender Association of Western Colorado, led by Dr. Curtis Swift and his team of research assistants, Travis Bondurant and Honora Carr, spent the summer of 2013 harvesting, distilling and analyzing the results of the GC/MS reports generated on 11 cultivars of English lavender (*Lavandula angustifolia*) and lavandins (*L. x intermedia*).

Nine cultivars were harvested from the main lavender research farm Green Acres U-Pick in Palisade, Colorado. One cultivar 'Maillette' was harvested from Sage Creations Organic Farm in Palisade, Colorado. One cultivar 'Royal Velvet' was harvested from A Pinch of Lavender farm in Palisade, Colorado. Floral stems from each cultivar were harvested in sufficient quantity to provide seven liters of material. The decision was made to use fresh lavender flowers and stems per the ISO standards.

In July of 2013, the floral stems of 11 cultivars of *Lavandula angustifolia* and *L. x intermedia* were distilled and evaluated by Gas Chromatography and Mass Spectrometry to determine the content of their constituents. The results were compared with the International Standards for Lavender (*Lavandula angustifolia*) and Lavandin (*L. x intermedia* 'Grosso').

See accompanying GC-MS tables for results.



## Plant Survival and Bud Yield

Floral stems were harvested in June and July of 2012 with scissors based on their stage of development thus the reason for the extended harvest season. The stems were separated by cultivar, plot number and replication for statistical analysis. Bundles were held together with rubber bands and hung in the garage of Bob and Elaine Korver out of direct sun until they were dry. Bundles were stripped of the floral stems by hand and screened through several sizes of sieves to remove extraneous material. Buds were weighed on an Ohaus Scout gram scale and weights recorded. The data was entered into MSTAT-C, a microcomputer statistical program developed by Michigan State University.

Means were run using MSTAT-C to determine percentage of winter survival and average yield of cleaned buds (calyces) per plant. The results are as follows:

Cultivar	Plant Survival	Average yield per
	(percentage)	plant (grams)
L. angustifolia		
'Betty's Blue'	97	7.9
'Folgate'	74	17.3
'Royal Purple'	33	14.8
'Royal Velvet'	86	16.1
'Twickle Purple'	100	27.8
L. x intermedia		
'Fat Spike'	86	38.0
'Impress Purple'	47	26.2
'True Grosso'	87	31.4
'Super'	60	19.9

#### L. angustifolia:

Based on the above data it is easy to select the highest yielding cultivars for Colorado lavender farms. Only one *L. angustifolia* achieved 100% winter survival; 'Twickle Purple'. 'Twickle Purple' was also the highest yielding angustifolia cultivar with each plant producing 27.8 grams of buds. That is slightly less than one ounce. Based on the average production per plant, a grower is capable of producing 69,500 grams or 2455.8 ounces (153 lbs) of bud from an acre (2500) of this cultivar. 'Betty's Blue' was the low yielder of the cultivars examined and was also found to be very hard to clean due to the sticky trichomes on the buds.

The number of bundles per plant was fairly consistent however the length of floral stems varied greatly.

Growers wishing to increase the harvest of buds will need to improve winter survival by mulching and providing winter water if soil moisture proves inadequate.

#### L. x intermedia

The yield of buds from these hybrid cultivars was greatest for 'Fat Spike' and 'True Grosso' at 38.0 and 31.4 grams respectively. 'Impress Purple' and 'Super' production was less with 'Twickle Purple', an angustifolia, exceeding their yields.

Winter survival was a problem for all the hybrids examined ranging from 47 to 87 percent. Winter protection is recommended for lavender growers in western Colorado.

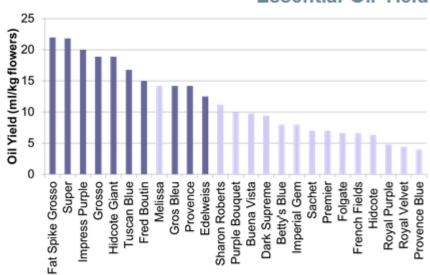
The average number of flower bundles for *L. angustifolia* was 6 and for *L. x intermedia* was 9. The length of stems and number of buds per stem varied but was not recorded. Keep in mind this was the second year after planting and would increase each year until maximum production was achieved. The same would be true for essential oil.

Most commercial lavender plantations are established for essential oil production with Lavandin producing the bulk of these essences. *L. angustifolia* cultivars typically yield up to 15 pounds of essential oil per acre while Lavandin can produce up to 67 pounds of essential oil per acre. (Barstow and Gardner, 2002). Foster (1992) provides more detail indicating *L. angustifolia* produces 300 to 1800 pounds of buds (calyces) per acre and when distilled result in from 12 to 15 pounds of essential oil. Bud production of Lavandin cultivars range from 3500 – 4500 pounds and 53 to 67 pounds per acre. At a wholesale price of \$30 per pound for Lavander and \$20 per pound for Lavandin oil, it is critical to select the highest yielding cultivars. Oil yield is highly veritable (Rabotyagov and Akimov, 1987) with new cultivars reported to produce 30% more flowers and 15 to 20% per more oil (MacTavish and Harris, 2002).

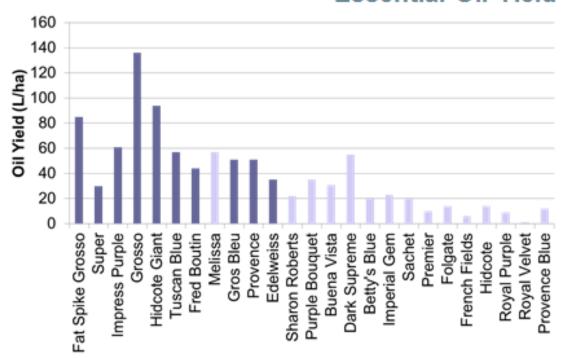
Bud production from the cultivars examined in this study ranged from 7.9 to 27.8 grams per plant for *L. angustifolia* and 26.2 to 38 grams per plant for Lavandin. Based on 2500 plants per acre, the yield of buds in pounds would range from 43.5 to 153.22 for 'Betty's Blue' and 'Twickle Purple' *L. angustifolia*, and 144.4 to 209. 43 for 'Impress Purple' and 'Fat Spike' lavandins respectively.

Dr Sean Westerfeld, Ginseng and Medicinal Herbs Specialist, Ministry of Agriculture and Food, Ontario, Canada conducted a detailed study of the essential oil yield of *L. angustifolia* (light blue) and Lavandins (dark blue). The following was reported at the 2013 US Lavender Growers Association Conference.

# **Essential Oil Yield**



# **Essential Oil Yield**



#### **Goals and Outcomes Achieved:**

There were three goals we wanted to achieve during this project. Goal 1 was to ascertain the components and quality of lavender essential oil of the listed cultivars grown in Western Colorado. Goal 2 was to educate members of LAWC on the best practices of distilling fine quality lavender oil. Finally Goal 3 was to educate members of LAWC on a thorough understanding of essential oil and hydrosol uses, including essential oil ISO standards and what components are important in what quantities in high quality essential oils.

## **Beneficiaries:**

The beneficiaries of this project far exceed our original goals. We hoped the reports and analysis would help our local lavender growers expand their operations by using this information to create higher quality products and market their products to a larger audience. Because of the advertisement of our specialty crop grant and the overwhelming interest from lavender growers across the country, we were able to reach a much wider audience. Instead of 30-40 local lavender growers, closer to 100-150 lavender growers from across the country have been impacted by our research and findings on lavender essential oils and hydrosols. These findings will have an impact on the entire U.S. lavender industry when our final report is published. Our report will help lavender growers better understand how to distill for essential oils and hydrosols, how to use the beneficial properties of essential oils and hydrosols and how to compete globally in the essential oils market by creating higher quality products.

Thank you,

Kathy Kimbrough Grant Manager Lavender Association of Western Colorado (970) 255-1312 kkimbro49@yahoo.com

#### Additional Information:

GC-MS analysis of *L. angustifolia* compared to Fresh Standards ISO GC-MS analysis of *L. x intermedia*Report on Evaluation of the Quality of Essential Oils

Angustifolia comparison to French (Maillette) ISO Standards

	3.5.11		D 1	Royal	m 111		D 44 1	TO 0 6: 1	700 Gt 1	
	Maillette	Folgate	Royal	Velvet	Twickle	Royal	Betty's	ISO Std	ISO Std	New York
	(SCOF)		Velvet	(Nielsen)	Purple	Purple	Blue	France	Other	Standards
cis-B-ocimene	0.24	0.19	0.23	0.2	0.09	0.26	nd	0 - 2.5	1.0 - 10.0	
myrcene	0.42	0.35	0.65	0.22	0.18	0.47	0.58	*	*	
limonene	0.23	0.66	0.85	0.19	0.31	0.88	1.04	03	0 - 1.0	
trans-B-ocimene	0.59	1.02	0.94	1.45	0.95	0.03	1.94	0 - 2.0	.5 - 6.0	
1,8-cineole	0.11	0.45	3.26	1.75	0.44	3.36	1.05	05	0 - 1.0	t-9
linalool	42.65	17.23	40.78	51.45	24.11	26.55	11.38	30 - 45	20 - 43	24-49
lavandulol	nd	nd	0.96	nd	nd	nd	nd	05	0 - 3.0	
hexyl butyrate	0.58	0.72	0.96	0.93	0.8	0.8	0.63	*	*	
camphor	0.71	0.29	0.38	0.23	0.38	0.38	0.22	0 - 1.2	0 - 1.5	t-6
terpinen-4-ol	0.21	9.14	3.73	6.79	1.64	6.43	0.17	0 - 1.5	0 - 8.0	
borneol	2.64	1.33	1.68	0.65	1.44	1.38	1.19	*	*	
a-terpineol	0.94	0.3	0.32	0.09	0.29	0.29	0.31	.5- 1.5	0 - 2.0	
linalyl acetate	35.7	41.56	27.71	24.24	51.71	39.59	53.53	33-46	25 - 47	11-55
lavendulyl acetate	0.58	6.66	1.28	0.72	4.3	2.94	9.02	0 - 1.3	0 - 8.0	
B-Phellandrene	*	*	*	*	*	*	*	02	0 - 1.0	
3 - Octanone	*	*	*	*	*	*	*	1 - 2.5	0 - 3.0	
	85.6	79.9	83.73	88.91	86.64	83.36	81.06	0	0	

<sup>\* =</sup> not tested ND = not detected New York Standards

# L x intermedia GC anaylsis

	Impress Purple	Provence	Fat Spike	True Grosso	Super	ISO
	•		Grosso		*	Standards
cis-B-ocimene	0.63	1	0.55	0.48	0.36	.5 - 1.50
myrcene	1.63	2.48	1.07	0.89	0.93	.3 - 1.0
limonene	1.88	1.8	0.79	0.81	1.26	.5 -1.50
trans-B-ocimene	1.05	nd	nd	0.04	0.84	0 - 1.00
1,8-cineole	14.57	24.82	10.12	9.95	6.29	4.0 - 8.04
linalool	43.13	27.79	24.05	25.68	29.14	24 - 37
lavandulol	0.75	nd	0.69	0.74	nd	.2 - 1.00
hexyl butyrate	nd	0.93	nd	nd	0.61	.350
camphor	7.43	5	8.02	8.53	2.69	6.0 - 8.5
terpinen-4-ol	0.44	5.06	2.31	2.23	0.18	1.5 - 5.01
borneol	10.69	12.7	1.73	2.3	5.85	1.5 - 3.51
a-terpineol	1.06	1.77	0.82	0.83	0.53	.3 - 1.30
linalyl acetate	4.76	2.28	33.49	31.67	38.33	25 - 38
lavendulyl						
acetate	0.48	0.42	4.12	4.1	2.3	1.5 - 3.51
B-Phellandrene	*	*	*	*	*	*
3-Octanone	*	*	*	*	*	*

<sup>\* =</sup> not tested for the ISO ND = not detected

Red = higher Blue = lower

# Evaluation of the Quality of Essential Oils of Lavender (*Lavandula angustifolia*) and Lavandin (*L. x intermedia*) grown and distilled at high altitudes of Western Colorado, U.S.A.

C.E. Swift<sup>i</sup>, G. Dooley<sup>ii</sup>, and K. Kimbrough<sup>iii</sup>

#### **Abstract:**

In the summer of 2013, the floral stems of 11 cultivars of Lavender (*Lavandula angustifolia*) and Lavandin (*L. x intermedia* 'Grosso') were distilled and evaluated by Gas Chromatography and Mass Spectrometry (GC-MS) to determine the concentration of their constituents. The results established base-line data of the essential oils of some *Lavandula* cultivars grown and distilled in western Colorado. The CG-MS of these essential oils are compared with international and other accepted standards. Two of the cultivars evaluated were obtained from locations other than at the Korver research site in Palisade, Colorado.

#### Introduction:

English lavender (*Lavandula angustifolia*) and the hybrid lavandin (*L. x intermedia*) cultivars have been successfully grown and harvested in western Colorado for 10 or more years. While the worldwide market for the essential oil of lavender is substantial no research has been conducted in Colorado to determine the yield or quality of oil of the various cultivars available for production in Colorado. This project was designed to answer the question of which cultivars produce the largest quantity and the highest quality oil.

The essential oil composition of *Lavandula* is known to be affected by species and variety and growing conditions such as latitude and altitude, fertilization, pesticide use (Topalov, 1989), and harvest time (Zheljazkov et al., 2012). While the chemical composition of the oil is largely determined by the genetics of the cultivar, oil quality can also be influenced by all stages of the production system (McGimpsey et al. 1999).

It has been reported high-altitude wild growing plants contain more esters than lavender grown at lower altitudes. The temperature of steam production is lower at higher elevation resulting in the hydrolysis of linally esters happening at a much slower rate creating a higher quality oil. (<a href="http://www.nature-helps.com/agora/lavender.htm">http://www.nature-helps.com/agora/lavender.htm</a>)

During steam distillation molecular rearrangements, hydrolysis of double bonds and deesterification of ester to alcohols and carboxylic acids can result. This changes the levels of linalool and linalyl acetate the key determinants of the fragrance of the oil. Linalool provides the sweetness and linalyl acetate provides the refreshing odor attributed to this oil. The final odor of the oil, the linalool to linally acetate ratio, was reported to be dependent on the length of distillation utilized.

Essential oil of this genus is produced in both the flower heads and foliage in specialized structures known as glandular trichomes or oil glands (Demissie et al., 2012). The largest quantity of oil is found in the calyces. The calyces (calyx – singular) consist of the sepals modified into vase-like structures from which the corolla (flower) protrudes. The calyces are typically called 'buds'.

#### **Materials and Methods:**

A randomized complete block design of 10 cultivars (Table 1) of *Lavandula* spp. were planted in May, 2011 in a furrow-irrigated field on property owned by Bob and Elaine Korver, 3601 G Rd., Palisade, Colorado. Each cultivar was planted six times in blocks of 10 plants each. Border plants were placed on the end of each of the eight rows in the trial. Maintenance provided by the Korver's, Swift, and Colorado State University non-student hourly personnel included irrigation, shearing and shaping, and weed control. Nitrogen fertilizer was applied at the rate of 1 pound per 1000 square foot area once each year after the time of the first bloom period.

In the summer of 2013 floral stems of nine of the ten cultivars evaluated were harvested from the Korver site. Floral stems of 'Royal Velvet' sample #2 were provided by LeAnn Nielsen, Nielsen Vineyard, Palisade, CO for distillation.

- Lavandula angustifolia
  - 'Folgate'
  - 'Twickle Purple'
  - 'Royal Purple'
  - 'Royal Velvet' (2 samples)
  - · 'Buena Vista'
- Lavandula x intermedia
  - 'Impress Purple'
  - 'True Grosso'
  - 'Fat Spike Grosso'
  - 'Super'

McGimpsey et al. recommend the drying of floral bundles prior to distillation. ISO 3515 Oil of lavender (Lavandula angustifolia Mill.) and ISO 8902 Oil of Lavandin Grosso (Lavandula angustifolia Mill. X Lavandula latifolia Medik. French type, however specify the essential oil will be produced by "steam distillation of the recently cut flowering tops." Fresh floral stems were used in this trial. Neither the stage of floral development nor the times for preheating or extraction are given in the ISO standards. The composition of the still, copper or stainless steel is likewise not specified.

Zheljaskov et al. (2012) reported harvest time had a significant effect on essential oil yields, concentration of linalool, and yields of linalool and linalyl acetate. To limit the effect of differing harvest time (floral development) the western Colorado research project used the stages of floral development specified by McGimpsey et al. from 4 and 6 as the time to harvest and distill the various cultivars (Table 1). To meet these requirements floral stems of each cultivar were individually selected for harvest.

**Table 1: Maturity Scale** 

Stage of Maturity	Description of Floral Development
4	Several flowers open, some beginning to wither
5	Approximately equal quantities of withered flowers and buds; some open flowers
6	Few buds left, some open flowers, but mostly withered flowers

McGimpsey, J.A. and Porter, N.G. 1999. Lavender: a growers' guide for commercial production. Pages 61-62.

Floral heads were harvested on the dates noted in Table 2 based on the above floral characteristics. This necessitated several harvest dates for 'Fat Spike', 'Impress Purple', 'Royal Velvet', 'Super', and 'True Grosso'. 'Betty's Blue', 'Folgate', 'Provence', and 'Twickle Purple' had more uniform floral development and floral stems from these cultivars were harvested on the same day.

Table 2: Harvest and Distillation Dates

Cultivar	Harvest and Distillation Date (s) 2013								
	6/17	6/22	6/24	6/27	6/29	7/01	7/02	7/03	7/05
'Betty's Blue		X							
'Fat Spike'					X		X		
'Folgate'	X								
'Impress Purple'						X	X		
'Provence									X
'Royal Velvet'			X	X					
'Super'						X		X	
'True Grosso'	X				X				
'Twickle Purple'				X					
'Maillette' *									

<sup>&#</sup>x27;Maillette' was provided by Sage Creations, Palisade Colorado.

Wesolowska, et al reported the time of distillation of *L. angustifolia* has an effect on the content and composition of the essential oils. They found a maximum essential oil percentage of 2% was obtained after two hours of distillation and the highest concentration of linalool as well as linally acetate was found after one hour of distillation. The lowest concentration of the linally acetate was observed after 40 minutes of

distillation. The minimum amount of essential oil (1%) was obtained after 40 minutes of distillation.

According to Pittman nearly 75% of the total oil yield comes in the first 25 minutes of distillation to give a commercial grade lavender oil. If other molecules are desired in the hydrosol it takes another 50 to 80 minutes of distillation. Statistical analysis of the results showed significant differences between the main constituents of the lavender oil and distillation time.

Note: The sample of 'Maillette' included in this study was steam distilled for ninety minutes using a stainless steel still. The preheating time was not reported.

Guidelines provided by Denny indicate the preheating stage of the distillation process should be no more than five minutes to prevent breakdown of linally acetate to linalool as this would result in a reduction in quality. The duration of the extraction is recommended to be between 25 and 30 minutes. The decision was made to follow Denny's guidelines.

Distillation was accomplished on the same day of harvest using an eight gallon stainless steel essential oil still from Mile High Distilling, Denver, Colorado. Approximate 2 gallons (7 L) of fresh floral stems (buds and stems) were firmly pressed into the section of the still designed to hold the plant material. To ensure proper movement of steam through the plant material (the charge) consisting of stems and flowers were compacted into the charge container. Three gallons of Palisade town water was added to the pot and brought to a vigorous boil before affixing the 'charge' to the still. The pot was refilled as needed. The condenser was supplied with cold water and the condensate was collected in one–liter separatory funnels purchased from NovaTech International. Condensate was generated from the condenser within five minutes of putting each charge on the still. The condensate from the condenser was adjusted to generate a steady stream of liquid by regulating steam production and the temperature of the condenser. A propane burner was used to generate steam in the pot.

When the separatory funnel filled with hydrosol and essential oil, the petcock on the bottom of the separatory funnel was opened and the hydrosol drained off. The essential oil was collected in sterile pint size canning jars. Each container was identified by cultivar name and date of distillation.

The essential oils were placed in the freezer to dewater the samples. The water in the sample froze and the water-free essential oil poured off in another sterile canning jars. Sterile pipettes were used to transfer the e.o. into five-millimeter amber glass bottles with hard plastic caps and chemical resistant liners.

Each collection of amber bottles were identified and placed in a refrigerator until all samples were processed and ready for shipment to the analytical laboratory for testing.

The e.o. samples were mailed on August 9, 2013 to Dr. Gregory Dooley, Assistant Professor, Director of Analytical Services, Center for Environmental Medicine, Dept. of

Environmental and Radiological Health Sciences, at Colorado State University, Fort Collins, CO. for analysis. Essential oil of 'Maillette', provided by Sage Creations Organic Farm was included in this mailing.

The following specifics on the characterization of Lavender Oils by GC-MS were provided by Dr. Dooley.

### **Sample Preparation**

- 1. In a 13 ml conical vial, add 150 mg of desiccated magnesium sulfate to 1ml of lavender oil and vortex
- 2. Cap vial and allow lavender oil to sit at room temperature for 2 hours with intermittent vortexing (every 30 mins)
- 3. Centrifuge sample at 1500 rpm for 5 mins
- 4. Dilute 100ul of lavender oil supernant to 1 ml with acetone in a GC autosampler vial for GC-MS analysis

#### **GC Conditions**

Column: Restek Rxi624Sil MS 30m x 0.25mm x 1.4 um

Carrier Gas: Helium Inlet Temp: 2500°C Flow Rate: 1ml/min

Oven Program: 60°C for 6mins to 310°C at 10°C/min (31min run time)

Injection Volume: 2ul via autosampler

Split Ratio: 200:1

#### **Mass Spectrometer Conditions**

MS: EI+ Full Scan from 40-200 m/z

Source Temp: 220°C

Transfer Line Temp: 280°C Electron Energy: 70eV

#### **Data Analysis**

Peaks were identified by comparison of MS spectra to the NIST MS spectra database. Identifications were considered valid with NIST algorithm Match Scores greater than 800 (out of 100) and Match Probabilities greater than 60%. Each peak in the chromatogram was integrated and % of total area calculated for each peak.

Statistical analysis of the oils was not accomplished due to lack of adequate quantities of oil and funds available for analysis. Ideally each plot of ten plants should have been distilled and analyzed separately. The amount of floral matter available from each plot was limited and had to be combined to fill the charge to the appropriate level. Denny indicates the height of charge is critical to avoid reflux and the hydrolysis and deesterification of the desired components.

# For the CG-MS results see Appendix.

#### Discussion:

The internationally recognized standards 3515 and 8902 provide a benchmark with which to compare oils from new production areas. These standards are often referred to in the world trade.

The International Standards 3515 for *Lavandula angustifolia* refer specifically to oils of lavender produced in France, Bulgaria, the Russian Federation, Australia, and other unnamed locations. 'Maillette' is the only named cultivar included in this ISO for which a specific listing of components are given. Spontaneous (wild-growing or seeded plants) and unnamed cultivars of other countries is also listed with a breakdown of e.o. components.

There are two sets of standards for American growers. The official international standards described above and what American companies are purchasing. The latter are based on what is arriving at the Port of New York (Tucker, 2001).

The purpose of this research was to compare Lavandula cultivar essential oil to the international standards and to develop a base line for cultivars grown in western Colorado. Wesolowska reported all of the oil samples they studied contained less linalyl acetate and cis-β-ocimene than the range specified in ISO Standard 3515. Their samples also contained higher levels of α-terpineol than called for in the specifications. Sean Westerveld, Ginseng and Medicinal Herbs Specialist, Ministry of Agriculture and Food, Ontario, Canada, evaluated the essential oil of 31 cultivars of *Lavandula* and none matched the ISO standards (personal communique). Tucker, 2001, reported the only cultivars that compared favorably with 'Maillette' were 'Munstead' (syn. 'Compacta'), 'Irene Doyle', and 'Twickel Purple'. With the exception of 'Maillette' in the Colorado trials the other cultivars did not meet the requirements of the ISO standards.

Financial Support for this research was provided by a specialty crop grant from the Colorado Department of Agriculture, obtained through the Lavender Association of Western Colorado.

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