Response of Anise Plants to Nitrogen Fertilizer and Foliar Spray of Tryptophan under Agricultural Drainage Water

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Abstract: A field experiment was carried out at Tamiya Province Fayoum Governorate over two growing seasons (2007-2008 and 2008-2009) to study the response of anise plants (*Pimpinella anisum L*) for two nitrogen forms (ammonium nitrate and ammonium sulphate) fertilizer and spraying of tryptophan at rates of (0,25 and50 ppm) on different growth parameters (plant height , shoot , umbels dry weight and seeds yield), oil percentage and chemical composition under irrigation with agricultural drainage water. It had been deduced that application of nitrogen fertilizer was effective in increasing all tested growth parameters and oil percentage compared with unfertilized treatment. The results indicated that the interaction between nitrogen forms and foliar spray of tryptophan at 25 and 50ppm increased oil percentage, N, P, K content and uptake in shoots and seeds compared to the use of nitrogen fertilizer alone. The highest values were noticed when ammonium sulphate was added with tryptophan (25ppm) in shoots and seeds. [New York Science Journal 2010;3(9):120-127]. (ISSN: 1554-0200).

Key words: Anise - nitrogen fertilizer - tryptophan – drainage water – growth – yield oil component – NPK.

1. Introduction

Anise, *Pimpinella anisum L.*, an herbaceous annual native to the Mediterranean region and Egypt, is cultivated in Europe, the Middle East, Mexico, North Africa, India and Russia chiefly for its fruits, called aniseed, the flavour of which resembles that of licorice. Anise was well known to the ancient Egyptians and Romans.

Anise (*Pimpinella anisum L*) is used for the treatment of a carminative, antiseptic, antispasmodic, expectorant, stimulant, and stomachic. In addition, it has been used to promote lactation in nursing mothers and as a medicine against bronchitis and indigestion. Muller-Schwarze, Dietland (2006).

The problem of salinity stress becomes alarming in arid or semiarid, where soils are already saline or irrigation water contains excess of soluble salts. Efforts are being made to overcome this problem primarily by modifying the saline environment, e.g. reclamation , drainage, use of high leaching practices or through use of nutrients, hormones, chemical and physical treatments and biological methods (El-Saidi 1997) The reuse of drainage water is an important strategy to meet the agricultural demands on condition that this reuse exerts minimum side effects on the environment including man health, soil/ ground water quality and crop quality and quantity.

The role of nitrogen in plant nutrition has been studied extensively during the past century and still continuing until data. A great number of researches were carried out on medicinal plants to determine their requirements and their response to ammonium sulphate and ammonium nitrate as will as the balance between these tow forms. Many report demonstrated that low N levels caused a reduction on growth and some chemical constituents (Yeh *et al* 2000)

Amino acids are essential for plant growth as they are involved in the biosynthesis plant compound. Tryptophan is the main precursor of the natural plant auxin (indole acetic acid) consequently; its application may affect growth, yield and the chemical constituents of the plant Abou Dahab and Nahed (2006)

Moreover, there have been reports that foliar application of amino acids (Lysine, ornithine, salicylic acid and tryptophan) enhanced the vegetative growth and chemical constituents Talaat and Youssef (2002) on basil plants, Talaat (2005) on *Pelargonium graveolens L.* and Talaat *et al.*, (2005) on *Catharanthus roseus L.*

Bharti Bhaisare *et al.* (2000) , Singh (2001),Upadhyay (2002), Velu (2002)and Balkcom, and Monks, (2008) found that application of nitrogen alone, or with growth regulators will make the plant photosynthetically more effective, also prevent flower shedding, improving pod setting and consequently increasing the yield.

Shoala (2000) sprayed *Lavendula multifida L* with amino acid (tryptophan) at 0 to 100ppm. The highest value of spike oil percentage was produced from using 50 to 100ppm Tryptophan

Senthil kumar and Jayakumar (2004) and Muthukumar, *et al* (2005) found that application nitrogen with naphthalene acetic acid (NAA) spray increased the growth, yield characters of greengram and corn,

Therefore the present investigation aimed to study the response of anise plants Anise (*Pimpinella anisum L*) for nitrogen fertilizer in addition to foliar spray of tryptophan on plant growth, yield and oil percentage and chemical composition under irrigation agricultural drainage water.

2. Material and Methods

A field experiment was carried out at Tamiya Province Fayoum Governorate over two growing seasons (2007-2008 and 2008-2009). The investigated soil under experimentation was of the following characteristics 24.3% sand, 22.1% silt, 53.6% clay, pH 8.74, EC 4.11 dSm^{-1}at $25^{O}C$, $CaCO_3$ 3.91%. The chemical analysis of drainage water source used for irrigation as shown in Table (1) it was clear that Na^+ and CL^- ions were as high as 8.3 and 8.48 meq / L respectively during the first season, increasing in the second season reaching 8.45meq / L for Na and 8.89meq/ L for CL.

Table (1) chemical analysis of drainage water

Seasons	pH	EC dSm^{-1}	Meq./L							
			Ca	Mg	Na	K	CO3	HCO3	SO4	CL
07/08	7.88	3.12	2.38	3.70	8.3	0.45	-	4.02	3.55	8.48
08/ 09	8.02	3.38	2.94	3.79	8.45	0.40	-	4.12	4.10	8.89

Anise (*pimpinella anisum* L) seeds were sown in the thired week of April. The treatments were arranged in randomized complete block design at four replications. The experimental plot was $10.5m^2$ (3.5m long and 3 m in width) with six ridges, 50cm apart. Recommended dose of phosphorus fertilizer as superphosphate (15.5% P_2O_5) was fully added to the soil during seed preparation at 100 kg P_2O_5 / feddan and Potassium fertilizer at 60kg K_2O feddan incorporated with soil before sowing. Nitrogen fertilizers were applied at two sources (Ammonium nitrate and Ammonium sulphate) at a rate of 100 kg^{-1} N

Seeds of anise were sown in hills 25cm apart within the ridge (3 seeds $hill^{-1}$) the plants were thinned to one plant $hill^{-1}$ after 25 days of sowing. The irrigation was carried out whenever needed using the agriculture drainage water from drainage. Foliar spray of tryptophan at three rate (0, 25 and 50ppm) .Four sprays at 3 weeks intervals were used. The first was after 45 days of cultivation

The experimental treatments can be described as follows:

1 - Control (without fertilizer)

2- Ammonium nitrate (AN)

3- Ammonium nitrate (AN) + 25 ppm of tryptophan

4 - Ammonium nitrate (AN) + 50 ppm of tryptophan

5 - Ammonium sulphate (AS)

6- Ammonium sulphate (AS) + 25 ppm of tryptophan

7 Ammonium sulphate (AS) + 50 ppm of tryptophan

At the harvesting time, vegetative growth parameters[plant height (cm), shoot dry weight (gm / plant) , umbels dry weight (gm / plant) and seeds yield (gm / plant)] which dried at 70 ^{O}C. dry weights , were recorded before grinding. The following chemical analyses were determined nitrogen, phosphorus, and potassium according to the method described by Cottenie *et al* (1982). Collected data was subjected to statistical analysis of variance according to Snedecor and Cochran (1980) The physical and chemical properties of the soil were determined according to Chapman and Pratt (1961)

Extraction of essential oil: seeds yield (50g) were subjected to Hydro distillation for 1h using a Clevenger type apparatus (Clevenger1928). Constituents of essential oil were determined by gas liquid chromatography. The chromatograph (ModelPerkin Elmer3920B) was equipped with a thermal conductivity detector and 2mx0.3cm column packed with10%Carbwax 20Mon 80/100 Chromsorb WAW and hydrogen was used As the carrier gas at$0.5cm^3 s^{-1}$ The column temperature was 130°C and detector and injector temperatures were 200°C.Constituents were identified by retention times and con-junction with known structures.(Adams1995)

3. Results and Discussion

Effect of nitrogen forms and tryptophan on growth parameters of anise plants:

It was clear that the source of water of irrigation (agricultural drainage water) drastically affected the

growth and seed yield of anise plants due to the high Na and CL ions. The plant growth was almost poor compared to treated plants. Tables (1 &2). Salt stress can affect several physiological processes, from seed germination to plant development. The complexity of the plant response to salt stress can be partially explained by the fact that salinity imposes both an ionic and osmotic stress (Pasternak, 1987).

Data in table (2) showed that adding nitrogen as ammonium nitrate and ammonium sulphate on anise plant in both seasons increased all growth parameters (plant height , shoots and umbels dry weight– seed yield) as compared with the control treatment. Generally increasing growth parameters may be explained on the assumption that, with increasing nitrogen supply, the proportion of the carbohydrate used in the portion increase. Data also, revealed that ammonium sulphate high increase in growth parameters as compared with ammonium nitrate (Yeh *et al* 2000)

With regarded to the interaction between foliar application of tryptophan (25 and 50 ppm) and different nitrogen forms, data presented in table (2) revealed that fertilizer with different nitrogen forms (ammonium nitrate and ammonium sulphate) and sprays with tryptophan at the two levels significantly increased growth parameters (plant height, shoots dry weight, and umbels dry weight and seed yield) compared with different nitrogen from individually as well as untreated one. These results are in a good harmony with Barai and Sarkar (1999) on *Capsicum carvi L* and kadiri (1999) on *Capsicum annuum plants.*

The highest values of plant height, shoots dry weight, umbels dry weight and seed yield were obtained by tryptophan (25 and 50 ppm) applied with ammonium sulphate compared to tryptophan (25 and 50 ppm) added with ammonium nitrate . The positive effect of amino acid on growth characters my be due the vital role of these amino acid stimulation on growth plant cell division, cell enlargement and cell elongation (Pareek *et al.*, 2000).

Foliar application of tryptophan (25 ppm) with nitrogen forms were more effective compared tryptophan (50 ppm) with nitrogen forms. According to above mentioned results Shoala (2000) and Salem *et al* 2001and Solaimalai *et al.* 2001) found that growth regulators can improve the effective partitioning and translocation of assimilates from source to sink in the field crops.

It is known that salinity has been shown to reduce the synthesis of DNA,RNA and protein in many plants (Levitt,1980). In this connection, it can be suggested that the stimulation of plant growth under saline condition by growth regulators might be due to its effects on the synthesis of macromolecules, for growth regulators are known to increase nucleic acid synthesis (Smith, 1985), stimulate various processes associated with the synthesis of protein (Datta *et al*, 1986) and promote al cell division (Egea-Cortines and Mizrahi, 1991).

Table (2) Effect of nitrogen forms and tryptophan on growth parameters of anise (*Pimpinella anisum* L) plants under agricultural drainage water (in both season)

Treatments	Plant height cm		Shoot dry weight (g / plant)		Umbels dry weight (g / plant)		Seed yield (g / plant)	
	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
Control (no fertilizer)	45.13	41.82	8.11	9.32	3.55	4.61	2.11	2.50
AN	68.37	63.79	15.97	16.80	7.11	8.84	4.63	5.00
AN + 25ppm Tryptophan	78.60	70.15	22.41	23.61	11.3	11.91	6.11	7.33
AN + 50ppm Tryptophan	71.00	66.31	17.66	19.00	9.71	10.00	5.33	5.60
AS	73.20	66.24	19.00	19.30	8.44	9.17	6.27	6.97
AS + 25ppm Tryptophan	85.41	79.43	25.91	26.44	12.59	13.23	9.34	9.80
AS + 50ppm Tryptophan	77.50	71.11	21.71	23.00	11.37	12.51	7.77	8.46
LSD 0.05								
	11.13	7.96	2.34	4.12	1.17	1.98	1.27	1.35

AN: ammonium nitrate. AS: ammonium sulphate

Chemical composition:

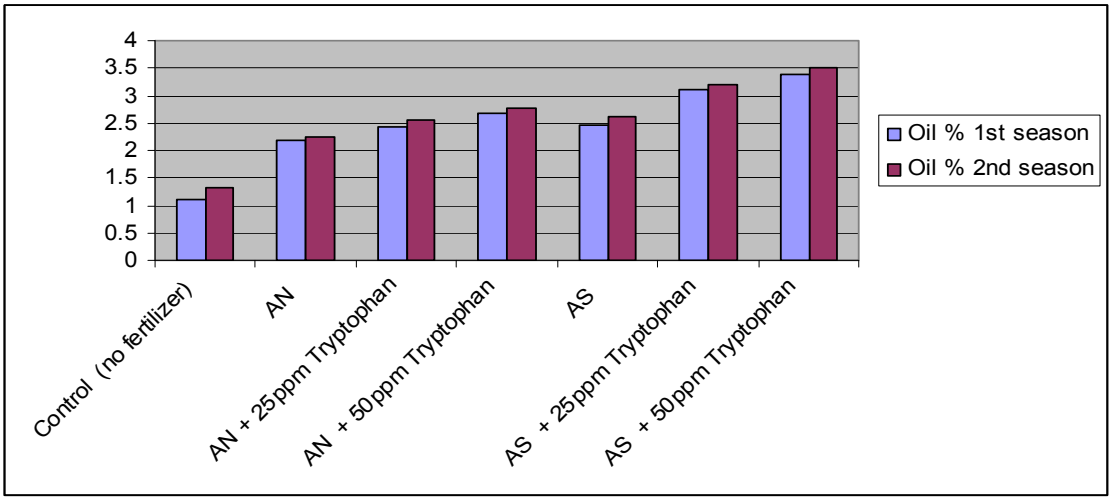
Essential oil percentage:

From the data in Fig (1) indicated that different nitrogen forms and foliar tryptophan (25 and 50 ppm) increased the oil percentage as compared the control and nitrogen from as sole in both seasons .Also, the increment of the concentration of applied resulted in an additional increase in the oil percentage. This conclusion agrees with that reported by Milad (1998) on *Mentha longifolia* and *Menthe viridis* by using tryptophan as a foliar spray at 25,50 and100ppm and Shoala (2000) on(*Lavendula multifida*). and Salem *et al* (2001) on caraway(*Carum carvi,L*) and coriander (*Coriandrum sativum,L*) with tryptophan25 and 50ppm respectively.

Essential oil components:

Fig (2,3&4) showed that the oil components of anise (*Pimpinella anisum* L) plants treated with nitrogen (ammonium nitrate and ammonium sulphate) with tryptophan were fractionated by (GC-MS) analysis. Anise oil contains anethole a pinene and terapenl acetate components. The control plants had the least anethole percentage if compared with other treatments. The most effective treatment was treatment ammonium sulphate + 50ppm of tryptophan in both seasons.

The highest a pinene percentage was determined in the oil distilled from the control plants followed by ammonium sulphate in the first and second seasons. The treatment of ammonium sulphate + 50ppm of tryptophan gave the maximum terapenl acetate percentage followed by ammonium nitrate in two seasons. + 50ppm of tryptophan



Fig(1) Effect of nitrogen forms and tryptophan on essentil oil percentage % on (*Pimpinella anisum* L) plants under agricultural drainage water (in both season)

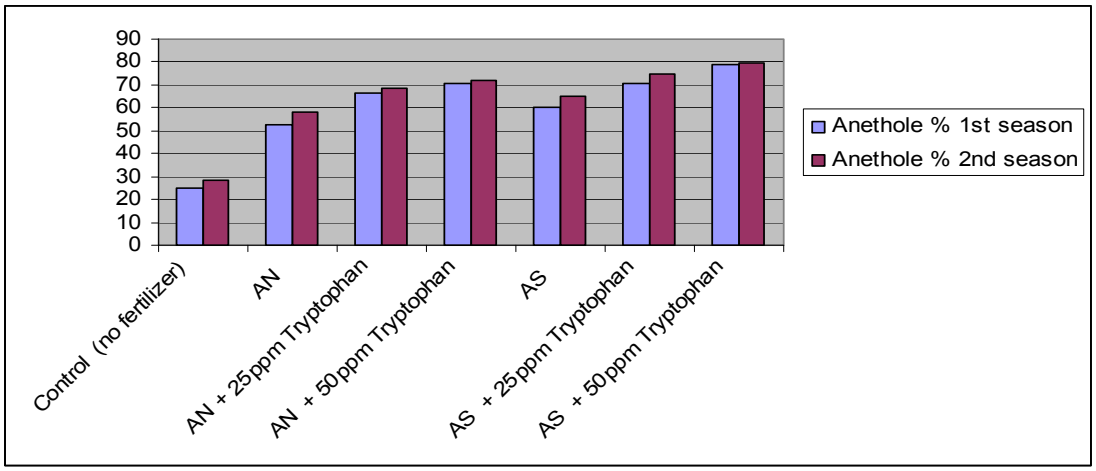


Fig (2) Effect of nitrogen forms and tryptophan on Anethole % (*Pimpinella anisum* L) plants under agricultural drainage water (in both season)

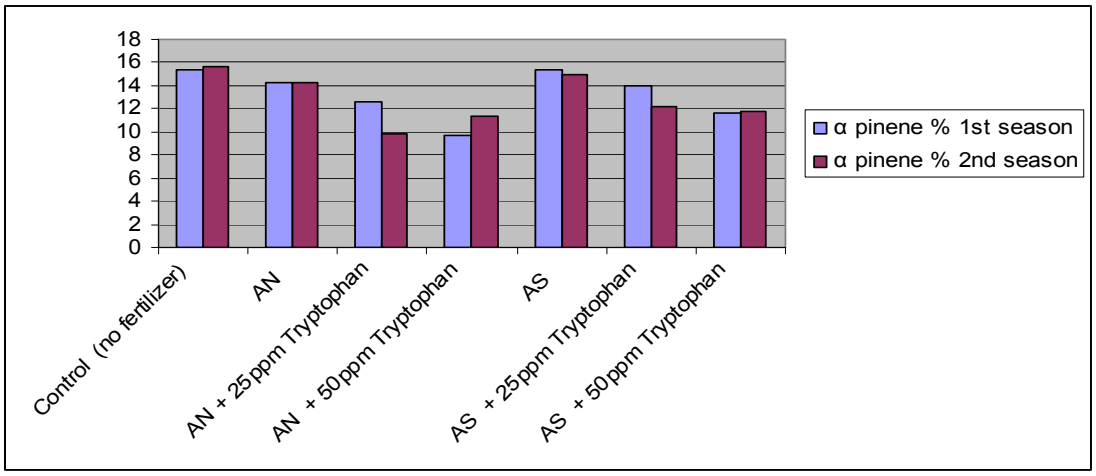


Fig (3) Effect of nitrogen forms and tryptophan on α pinene % (*Pimpinella anisum* L) plants under agricultural drainage water (in both season)

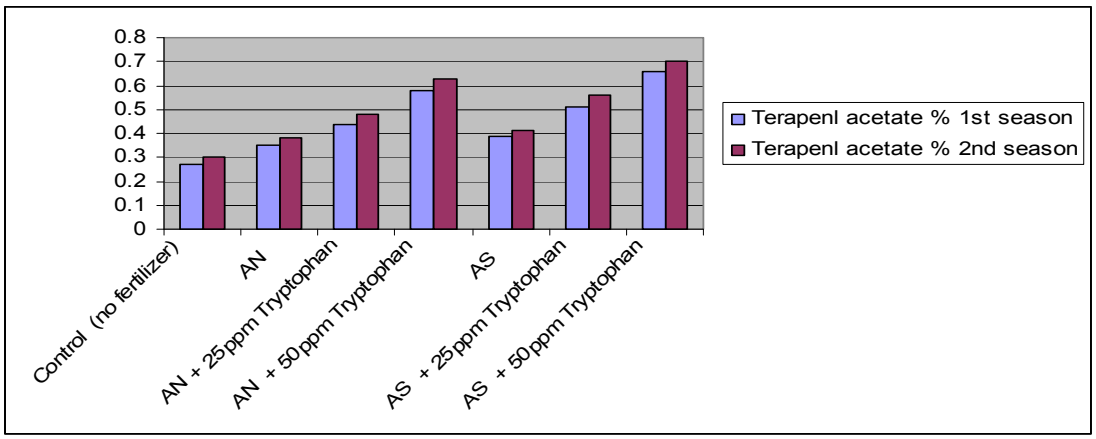


Fig (4) Effect of nitrogen forms and tryptophan on Terapenl acetate % (*Pimpinella anisum* L) plants under agricultural drainage water (in both season)

Nutrient content:

Nitrogen, phosphorus and potassium content and uptake by anise plant as affected by different forms of nitrogen fertilizers and foliar spray of tryptophan under agricultural drainage water are shown in Table (3&4).Results revealed that all treatments tended to increase N, P and K concentration and uptake in the first and second seasons in both shoots and seeds as compared with the control treatment. These results are in a good harmony with Talaat, and. Youssef, (2002)

The maximum N content was noticed when nitrogen as ammonium sulphate + 25ppm of tryptophan (1.52% and 1.64% in shoots) and (2.48 and 2.53% in seeds) in both seasons respectively. Although N uptake 393.8 and 433.6 mg / plant in shoots and 231.6 and 247.9 mg / plant in seeds in the first and second seasons respectively. Data also, showed that N concentration tend to decrease with increasing tryptophan (50ppm) with both ammonium sulphate and ammonium nitrate.

It was evident from the obtained data in Table (3&4) that application of nitrogen fertilizer and foliar spray tryptophan under agriculture drainage increased P and K content and uptake in shoots and seeds as compared with control treatment.

Tryptophan at low level (25 ppm) with different nitrogen forms (ammonium nitrate and ammonium sulphate) were more efficient with the P and K percentage and up take as compared with high level(50ppm) with different nitrogen forms.

The highest level of P content was recorded at ammonium sulphate +50ppm of tryptophan fallowed by ammonium nitrate +50ppm of tryptophan these results due to growth substances may be a result of their roles on regulating ions and modify the movement of nutrients within the plant tissues. The same trend was observed for K content in shoots and seeds in both seasons.

Table (3) Effect of nitrogen forms and tryptophan on N, P and K content (%) of anise (*Pimpinella anisum* L) plants under agricultural drainage water (in both seasons)

Treatments	N%				P%				K%			
	shoots		Seeds		shoots		seeds		shoots		seeds	
	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
Control (no fertilizer)	0.77	0.80	1.12	1.20	0.17	0.20	0.28	0.32	1.32	1.36	1.11	1.15
AN	1.25	1.31	2.00	2.10	0.21	0.23	0.40	0.44	2.51	2.58	1.38	1.42
AN + 25ppm Tryptophan	1.48	1.53	2.32	2.38	0.23	0.24	0.50	0.50	2.60	2.65	1.48	1.51
AN +50ppm Tryptophan	1.43	1.48	2.20	2.37	0.25	0.27	0.58	0.48	2.66	2.72	1.59	1.63
AS	1.36	1.39	2.27	2.30	0.22	0.24	0.44	0.54	2.64	2.71	1.41	1.48
AS + 25ppm Tryptophan	1.52	1.64	2.48	2.53	0.25	0.25	0.55	0.59	2.76	2.80	1.55	1.55
AS + 50ppm Tryptophan	1.44	1.50	2.36	2.41	0.28	0.30	0.64	0.68	2.88	3.00	1.64	1.77
LSD 0.05												
	0.27	0.19	0.40	0.35	0.04	NS	0.09	0.05	0.24	0.37	0.08	0.25

Table (4) Effect of nitrogen forms and tryptophan on N, P and K uptake (mg / plant) of anise (*Pimpinella anisum* L) plants under agricultural drainage water (in both season)

Treatments	N (mg / plant)				P (mg / plant)				K (mg / plant)			
	shoots		Seeds		shoots		seeds		shoots		seeds	
	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
Control (no fertilizer)	62.4	74.5	23.6	30.0	13.7	18.6	5.9	8.0	107.0	126.7	23.4	28.7
AN	199.6	220.0	92.6	105.0	33.5	38.6	18.5	22.0	400.8	433.4	63.8	71.0
AN + 25ppm Tryptophan	331.6	361.2	141.7	174.4	51.5	56.6	30.5	36.6	582.6	625.6	90.4	110.6
AN +50ppm Tryptophan	252.5	281.2	117.2	132.7	44.1	51.3	30.9	26.8	469.7	516.8	84.7	91.2
AS	258.4	268.3	142.3	160.3	41.8	46.3	27.5	37.6	501.6	523.0	88.4	103.1
AS + 25ppm Tryptophan	393.8	433.6	231.6	247.9	64.7	66.1	51.3	57.8	715.1	740.3	144.7	151.9
AS + 50ppm Tryptophan	312.6	345.0	183.3	203.8	60.78	69.0	49.7	57.5	625.2	690.0	127.4	149.7
LSD 0.05												
	20.0	42.2	15.8	23.6	4.9	7.1	2.9	NS.	63.3	83.5	17.1	6.7

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