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Heavy Metal Contents of St. John's wort (*Hypericum perforatum* L.) Growing in Northern Turkey*

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Abstract: *Hypericum perforatum* L. plants representing a total of 35 wild populations were collected from Black Sea region of Turkey at flowering stage and subsequently assayed for some heavy metal contents, namely Cu, Mn, Fe, Zn and Pb. The results of chemical analysis of plant samples revealed that total individual plant contents in these wild populations were 1.81-10.09 µg Cu g⁻¹ (mean:6.34), 197.88-9.10 µg Mn g⁻¹ (mean:39.26), 2990.21-54.16 µg Fe g⁻¹ (mean:434.34), 181.60-12.91 µg Zn g⁻¹ (mean:68.31) and 124.76-12.04 µg Pb g⁻¹ (mean:47.51), respectively. According to the results of variance analysis, heavy metal contents of *H. perforatum* plants varied with sites to a large scale and this variation was found to be significant statistically.

Key words: *Hypericum perforatum*, Cu, Mn, Fe, Zn, Pb, wild populations, variation

Introduction

The contamination of agricultural lands and irrigation water with toxic pollutants such as inorganic contaminants poses an environmental risk to humans and animals. Heavy metals including copper, manganese, iron, zinc and lead, may accumulate naturally in soil or may originate from industrial and mining processes (Greeger, 1999). Heavy metals accumulate to varying degrees in different plants species and have been shown to impact crop production (Salt *et al.*, 1995).

The recent increase in the popularity of plant-based medicines including herbal products, phytopharmaceuticals and traditional pharmaceuticals derived from leafy plants, has lead to a new segment in crop production and agriculture. However, optimized agricultural practices have not been developed for many of the medicinal species. In contrast to food crops, increased yield or biomass production is important but quality is also assessed in terms of the content of specific phytochemicals composition in medicinal species. Therefore, many agronomic factors should be considered when developing production strategies for medicinal plants (Murch *et al.*, 2003).

Hypericum perforatum L., commonly known as St. John's wort, is one of plants used in drug production industry intensively. It is a well-known traditional medicinal plant that has been used for centuries for the treatment of several diseases, such as skin wounds, eczema, burns, antimicrobial, anti-inflammatory and psychological disorders (Barnes *et al.*, 2001; Sanches-Mateo *et al.*, 2002). Especially

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using of this unique plant as an antidepressant is very popular today (Biffignandi and Bilia, 2000). This is evidenced by the fact that the market for *Hypericum perforatum* has exceeded \$210 million in USA and \$570 million worldwide annually (Sirvent and Walker, 2002).

Recent advances in biochemistry and medicinal sciences have resulted in using medicinal plant extracts or herbal teas for the treatment of a range of ailments. As a result of factors to which plants have exposed during their development, toxic heavy metals in plant tissues are major problems with herbal medicine. Metallic elements are constitutive plant compounds with biological activity as essential or toxic agents in metabolism. Environment in developing countries, pollution in irrigation water, atmosphere, soil, sterilization methods and storage conditions all play an important role in contamination of medicinal plants by metals (Gomez *et al.*, 2004). Metals may contaminate different plants causing serious health hazards such as injury of kidney, symptoms of chronic toxicity, renal failure and liver damage (Andrew *et al.*, 2003). Therefore, metals have been investigated in different plant materials in order to establish their normal concentration range and evaluate their role in plants as part of human medicinal treatment. To authors' knowledge, there is no published report documenting *H. perforatum* plants in terms of metal contents. Due to the lack of such information, the present study focused on determining the contents of some heavy metals namely copper, manganese, iron, zinc and lead from wild populations of *H. perforatum* collected in Northern Turkey where some companies have collected plant materials for export to herbal industry (Çırak *et al.*, 2004). The aim of the research was to document the metal content of *H. perforatum* populations sampled from a total of 35 sites as an initial step to evaluate using of this plant from health care point of view.

Materials and Methods

Plant Material

H. perforatum plants representing a total of 35 samples were collected between July and August of 2003 from the sites located on Black Sea region of Turkey: Çamlıhemşin, Çayeli, Fındıklı, Ardeşen, Arsin, Tonya, Uzungöl, Soğanlı, Bayburt, Aydıntepe, Bolaman, Ordu, Espiye, Eynesil, Asarcık, Samsun, Gümüş, Pazar, Niksar, Hamamözü, Gümüşhacıköy, Daday, Ağlı, Taşköprü, Azdavay, Çatalzeytin, Inebolu, Abana, Seydiler, Küre, Durağan, Ayançık, Türkeli, Bafra and Vezirköprü (Fig. 1). Sampling was randomized from plant crowns that had at least three stems. The top 1/3 of the crown was harvested between 10:00 AM and 2:00 PM. Conditions on the day of collection were clear and sunny at all sites. Temperatures ranged from 24 to 35°C. Samples were dried overnight (or until constant weight) at 65°C, the current temperature used by wildcrafters in USA for industry production (Sirvent and Walker, 2002).

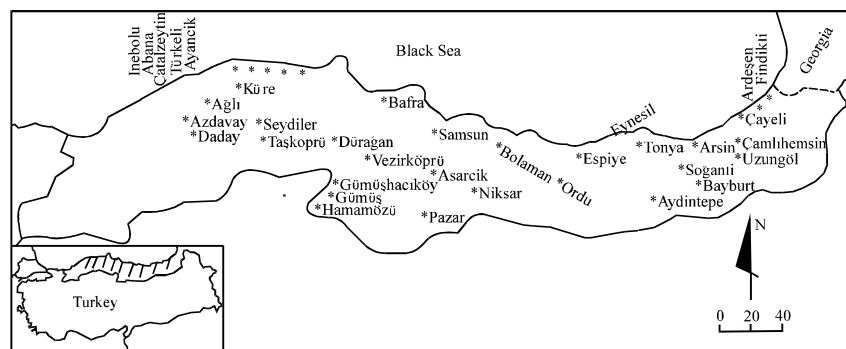


Fig. 1: The map of Black Sea geographical region located in Northern Turkey showing the sites where *Hypericum perforatum* L. plants were collected

Determination of Fe, Cu, Zn, Mn and Pb contents of Samples

Dried and homogenized plant materials were digested with HNO₃:HClO₄ (4:1, v/v) and then Fe, Cu, Zn, Mn and Pb contents in the solutions were determined spectrophotometrically by using atomic absorption (Kacar, 1972).

The data were subjected to ANOVA and differences among samples were tested Duncan Multiple Range Test (Level of significance $p < 0.01$). When necessary, data were normalised using $x' = \sqrt{x+1}$ transformation.

Results and Discussion

According to the results of ANOVA, heavy metal contents of *H. perforatum* plants varied with sites to a large extent and this variation was found to be significant statistically ($p < 0.01$) (Table 1). The results of chemical analysis of plant samples revealed that heavy metal contents of *H. perforatum* plants were 1.81-10.09 $\mu\text{g Cu g}^{-1}$ (mean:6.34), 197.88-9.10 $\mu\text{g Mn g}^{-1}$ (mean:39.26), 2990.21-54.16 $\mu\text{g Fe g}^{-1}$ (mean:434.34), 181.60-12.91 $\mu\text{g Zn g}^{-1}$ (mean:68.31) and 124.76-12.04 $\mu\text{g Pb g}^{-1}$ (mean:47.51), respectively. In case of Cu, Mn, Zn and Pb, the wild populations of *H. perforatum* plants supplied the moderate amounts of the metals and there were 10-20 fold difference between the highest and lowest values. However, Fe content of *H. perforatum* plants varied enormously with populations. The site Soğanlı gave 55 folds higher Fe than that of the site Ayancık which supplied the lowest value (2990.21 and 54.16 $\mu\text{g g}^{-1}$, respectively (Table 1).

To author's knowledge, there is no report documenting toxic and sufficient levels of aforementioned heavy metals for *H. perforatum* except one concerning the metal contents monitoring of *Hypericum* derivatives (Gomez *et al.*, 2004). Since the plant is considered as a noxious weed to a large scale rather than one cultured worldwide (Kirakosyan *et al.*, 2004). In spite of this, the presence of heavy metals like Pb in food and industrial crops are not acceptable in terms of health care even if the plants don't exhibit toxicity due to biomagnification (Kabata-Pendias and Pendias, 1992). Today, *H. perforatum* has become one of the leading plants used in drug industry and is currently supplied by mainly wild-harvested materials (Çırak *et al.*, 2004). Besides, it is used by folks as herbal tea in world-wide on a large scale (Çırak and Kevseroğlu, 2004). Therefore, the presence of heavy metals, especially Pb which has high toxicity level to mammalian in *H. perforatum* plants from wild population has a great importance in health care and agricultural point of view. While lower contents of Cu, Zn, Mn and Fe are useful for plants as nutrition, their high contents which are peculiar to each plant are toxic (Adriano, 1986).

In the present study, plant materials were collected from parts of Northern Turkey where out side of industrial area. Thus, the presence of aforementioned metals in plant samples derived from probably their high contents in soil. Nevertheless, the probability of that metal accumulation in plant samples may be vehicle-originated was omitted and it is possible that Pb found in plant samples may be both vehicle and soil-originated. It is interesting to note that *H. perforatum* plants growing wild in the vicinity of road may be hyper accumulator for some heavy metals like Pb. Because, using of such a plant accumulating toxic elements in drug industry would bring about some health problem. Likewise, Murch *et al.* (2003) reported that exogenous application of Ni (25 and 50 mM NiNO₃) in culture medium resulted in 66 and 140 $\mu\text{g g}^{-1}$ Ni accumulation in leaves of *in vitro* grown *H. perforatum* seedling. Results from the study support that *H. perforatum* may be a hyper accumulator for some heavy metals.

Table 1: Heavy metal contents of *H. perforatum* plants collected from Northern Turkey

Samples	Cu ($\mu\text{g g}^{-1}$)	Mn ($\mu\text{g g}^{-1}$)	Fe ($\mu\text{g g}^{-1}$)	Zn ($\mu\text{g g}^{-1}$)	Pb ($\mu\text{g g}^{-1}$)
Küre	3.90y*	20.04w	67.68z	29.52z	17.33z
Samsun	9.21c	36.17m	431.21k	58.63p	34.41u
Gümüş	8.12h	27.55p	786.79f	38.00y	22.30z
Çamlıhemşin	7.52k	21.58u	122.14z	26.40z	18.13z
Türkeli	4.53w	14.48z	179.36v	90.45j	62.14j
Asarcık	6.49o	110.78c	1175.97c	58.66p	40.30q
Eynesil	5.02z	10.79z	148.98w	69.77m	54.91l
Ayancık	5.58t	9.10z	54.16z	42.45x	29.17y
Daday	8.46f	12.34z	116.45z	77.29l	53.10m
Espiye	9.66b	30.34o	835.59d	122.26f	83.99f
Ordu	10.09a	22.41t	462.93j	133.72d	91.86e
Çayeli	6.08q	17.14y	214.50r	12.91z	10.31z
Ağlı	9.16c	11.85z	100.20z	124.87e	110.76c
Arsin	6.25p	25.32r	825.53e	44.58v	30.62w
Durağan	5.35u	20.65v	123.72z	54.30s	37.30r
Azdavay	1.81z	9.31z	60.80z	17.53z	12.04z
Taşköprü	4.04x	12.46z	82.49z	104.21g	71.59g
Niksar	5.84s	15.22z	259.99p	26.94z	18.51z
Pazar	1.88z	66.20f	139.52x	30.93z	21.25z
Aydıntepe	6.87n	121.56b	2213.03b	50.40t	34.63t
Gümüşhacıköy	9.22c	54.41g	575.52i	83.81k	57.58k
Fındıklı	7.81i	49.10h	752.89h	156.00c	107.17d
Soğanlı	9.00d	197.88a	2990.21a	99.88h	68.62h
Çatalzeytin	8.33g	15.11z	204.19s	181.60a	124.76a
Hamamözü	8.07h	25.00s	263.79o	93.88i	64.49i
Bafra	7.14l	45.23i	771.07g	32.75z	25.83z
İnebolu	7.65j	26.04q	126.90y	56.89q	43.23o
Vezirköprü	6.99m	18.49x	115.41z	54.45r	36.25s
Çayırıcı	8.73e	89.64d	385.86l	36.70z	25.22z
Abana	6.45o	12.59z	203.23t	164.47b	112.99b
Fatsa	6.25p	34.09n	340.96m	63.33n	43.51n
Seydiler	5.95r	67.45e	121.03z	44.71u	30.72u
Bayburt	4.12x	40.08k	221.02q	42.97w	29.52x
Uzungöl	6.24t	38.99l	264.68n	23.15z	13.93z
Ardeşen	2.66z	41.92j	182.05v	60.30o	41.42p
Mean	6.34	39.26	434.34	68.31	47.51

*Values followed by different small letters in columns are significantly different ($p < 0.01$) according to Duncan Multiple Range test

Conclusions

Considering the fact that the use of medicinal plants in both crude and prepared forms has greatly increased in recent years, it is necessary to ensure the safety, quality and efficacy for their consumptions by public. One of the main problems concerning herbal remedies is the contamination by heavy metals in plant materials which may results in serious health injuries. *H. perforatum* has become one of the widely popular herbal remedy worldwide and is currently supplied by cultivated and especially wild-harvested plant materials. Present results indicated that the *Hypericum* plants from wild populations of Northern Turkey include enormous amount of Cu, Mn, Fe, Zn and Pb and this plant may be a hyper accumulator. This is the first report documenting the presence and abundance of metals in wild *H. perforatum* plants. Further studies are needed to determine the critic levels of each heavy metal and to elucidate the accumulations and toxic levels of heavy metals for this plant.

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