# Multiannual dynamics of *Perilla frutescens* L. Britton terraneous part productivity and increment during vegetation cycles in Middle Lithuania

Ona Ragažinskienė<sup>1</sup>,

Lina Bumblauskienė<sup>2</sup>,

Erika Šeinauskienė<sup>1</sup>,

Antanas Praškevičius<sup>2</sup>,

## Valdimaras Janulis<sup>2</sup>

<sup>1</sup> Kaunas Botanical Garden of Vytautas Magnus University, Ž. E. Žilibero 6, LT-6324 Kaunas, Lithuania E-mail: o.ragazinskiene@bs.vdu.lt

<sup>2</sup> Kaunas University of Medicine, A. Mickevičiaus 9, LT-44307 Kaunas, Lithuania E-mail: farmakog@kmu.lt The article presents the data of investigations of multiannual dynamics of *Perilla frutescens* L. Britton. terraneous part quantity increment during vegetation cycles in the period from 2001 to 2006 at the Kaunas Botanical Garden of Vytautas Magnus University. *Perilla frutescens* L. Britton. is an annual herbaceous plant, belonging to *Lamiaceae* Lindl. family, originated in East Asia. In recent years, it has been cultivated and introduced into various parts of the world. This species is being introduced into the collection of medicinal plants in a 345 m<sup>2</sup> trial area at the Kaunas Botanical Garden of Vytautas Magnus University, Kaunas, Lithuania. It has been assessed that the average mass of the model plant fresh material is 335.8 g, while that of the dried one is 95.8 g. The average mass of these leaves of the model plant is 248 g, whereas the average mass of dried leaves is 49 g. The mass of the leaves makes up 70 per cent of the total terraneous raw material. Correlative interdependence was established between the number of leaves and hydrothermal coefficient (R<sup>2</sup> = 0.7418). The absolute increment of the mass of the raw material and its growth rate increase up till maximum from the initiation of vegetation up till flowering. At the end of flowering, the rate of this increment suddenly slows down.

The revealed consistent patterns of growth enable to prognosticate the probable quantity of *Perilla frutescens* L. terraneous part, increment and dynamics of increment during vegetation cycles in Lithuania.

Key words: *Perilla frutescens* L. Britton, introduction, multiannual dynamics of increment, meteorological factors

## INTRODUCTION

In recent years, due to the changed tendencies in the usage and preservation of herbal and spice plants as well as increased requirements for the raw material quality and biodiversity preservation, more attention is paid to the cultivation of medicinal plants and to their introduction into the collections and industrial plantations. Furthermore, studying of their properties and utilization are getting more and more relevant both in the Lithuanian and world pharmacy (Lage, 1998; Bernath, 1999; ESCOP Monographs, 2003; Ragažinskienė et al., 2003; Radušienė, 2004; Radušienė et al., 2004).

In dealing with the global problem – health for all in the 21<sup>st</sup> century – the main task is to promote and sustain human health by applying preventive treatment (Ragažinskienė et al., 2004). During recent years, herbs with multipharmacological properties are increasingly being used to treat various allergic, autoimmune, oncologic, and infectious diseases (Ragažinskienė et al., 2004; Gailys, 2005). Such an immunomodulator is *Perilla frutescens* L. Britton. (further, *P. frutescens*), a species, originated in East Asia, with its natural growth areas. Now it is being investigated and cultivated not only in Asia but also in North

America, South Africa, Southeast Europe (Brenner, 1993; Omer, 1998; Hartley, 1996; Nitta et al., 2003).

P. frutescens has also been cultivated and investigated in the medicinal plant collection and trial area at the Kaunas Botanical Garden of Vytautas Magnus University since 1998. P. frutescens is a new perspective medicinal, spice and decorative plant which has already enriched national genetic resources and diversity of species (Ragažinskienė et al., 2006). It has been ascertained that this species passes through all the vegetation cycle up till full fruit ripening in Lithuania (Ragažinskienė et al., 2006). P. frutescens herbal raw material (Perillae folium and Perillae fructus) is included in the Chinese Pharmacopoeia (The Pharmacopoeia..., 1992). During the last decades various experiments confirming the variety of pharmacological effects - antimicrobial (Gailys, et al., 2004), antioxidative (Tada et al., 1996; Nagatsu et al., 1995; Povilaitytė et al., 2000), anti-inflammatory (Naomi et al., 2004), and nutritional properties (Kim 1997; Longvah et al., 1991) - were carried out. The dependence of P. frutescens raw material upon the meteorological factors, the increment of the number of leaves during vegetation in separate cultivation years in Lithuania has not yet been assessed. These consistent patterns permit to prognosticate the productivity of P. frutescens raw material.

The aim of investigations was to determine the multiannual dynamics of increment of the raw material (*Perillae herba*, *Perillae folium*) of the terraneous parts of *P. frutescens* and the number of leaves increment during vegetation as well as to assess their dependence upon the meteorological factors.

#### MATERIALS AND METHODS

*Perilla frutescens* L. Britton. is an annual 80–100 cm height herbaceous plant, belonging to *Lamiaceae* Lindl. family. The stem is branching, hollow, square. Leaves are purple, blades ovate, serrate, acute, crisp. The flowers are pink, bilabiate, androgynous, decussate, each subtended by a single folded bract. Perianth is complexed, five lobed. Inflorescence forms a spike. Fruit is a nutlet (He-Ci-Yu et al., 1997).

Herbal raw material is composed of fresh and dried terraneous parts: leaves (*Perillae folium*), herb (*Perillae herba*), and fruit (*Perillae fructus*) (The Pharmacopoeia..., 1992).

Investigations were carried out in the collection of medicinal plants and in a trial area at the Kaunas Botanical Garden of Vytautas Magnus University during the period from 2001 to 2006. According to climatic zones, it is a part of the central lowlands of Lithuania, where the average yearly temperature is +6.7 °C, the annual sum of active temperatures >10 °C is 2100 to 2300, the average minimal monthly temperature in winter is from -24 °C to -26 °C, and annual precipitation level is 500– 750 mm (Bukantis, 1994; Agrometeorologiniai ... 2001–2006).

The meteorological factors in the period of investigations are presented in Fig. 1. Hydrothermal coefficient (HTC) was calculated according to the formula HTC = H/0,  $1\Sigma T$  (Dirsė, 2001).

In the 345 m<sup>2</sup> experimental area, *P. frutescens* was planted by sprouts with three leaves in rows 50 cm apart; the distance among the rows was 70 cm. The phenological observations and biometrical measurements of the model plant (10 reiterations) were performed during various periods of vegetation: initiation of vegetation, intense growth, pre-flowering, flowering, seed ripening and the end of vegetation.

Herbal raw material was prepared during flowering (August–September), and fruit was collected during the full ripening period. Raw material was dried in a well- ventilated chamber, or in a stove at a temperature of 25–30 °C. Statistical analysis of the obtained data was performed using SPSS 11.0. Linear regression model was analyzed. For the suitability of each regression model, determination coefficient R<sup>2</sup> was obtained. The strength of the linear relationship between variables was meas-

ured by Pearson's correlation coefficient R. The Mann–Whitney test was performed for the hypothesis concerning the equality of distributives. Significance level was chosen  $\alpha = 0.05$  (Miuleris, 1994; Čekanavičius, et al., 2000; Čekanavičius et al., 2002).

### **RESULTS AND DISCUSSION**

The following analyses were performed: the dynamics of terraneous parts (height, projectional cover), the dependence of vegetation rhythmics on the meteorological factors, the optimal time for harvesting raw material, predetermining its quantity and quality. The earliest beginning of vegetation and the optimal climatic conditions for growth were at the time when HTC was 1.80. The optimal condition for flowering and seed ripening was the period when HTC was 1.20. The vegetation of this typical short day plant starts at the earliest on the 125th-130th days of the year (May), and the intense growth is in maximum progress till the 200<sup>th</sup> day (July) and tends to decrease from pre-flowering till massive flowering periods (200th-230th days of the year) (Ragažinskienė et al., 2006). Influence of meteorological factors (active air temperature sum, hydrothermal coefficient, precipitation) on the dynamics of P. frutescens raw material increment (g) and quantity of leaves during vegetation in different days of the year has not yet been investigated.

Multiannual (2001–2006) analysis of the fresh and dried mass quantity of raw material *perillae herba* and *perillae folium* of the *P. frutescens* model plant was studied (Fig. 2). It was assessed that in the years 2003 and 2006 the maximum yield of *P. frutescens herba* was 456 and 539 g of the fresh material, and 110 and 113 g of the dried material, respectively. The minimal yield of the fresh mass was 268 and 258 g, and 71 and 69 g of the dried material. In summary, it was assessed that the average yield of the fresh *P. frutescens herba* was 335.8 g, while that of the dried material was 95.8 g (2001–2006).

The yield of *P. frutescens* leaves constitutes about 70 per cent of the total amount of *P. frutescens* material. It was purposeful to assess the productivity of leaves. The average yield of fresh leaves (2003, 2004, and 2006) was 272.6 g, while the yield of dried leaves amounted to 56.3 g. The maximal yield of fresh and dried leaves (Fig. 3) was 329 g and 70 g in the years 2003 and 2006, whereas the minimal one was 168 g and 35 g, respectively (2004).

In 2006, the raw material quantity analysis was performed, mass dynamics of raw material (*herba* and *folium*) was observed during vegetation, raw material mass increment (Fig. 4) was assessed from initiation of vegetation (181<sup>th</sup>-211<sup>th</sup> days)

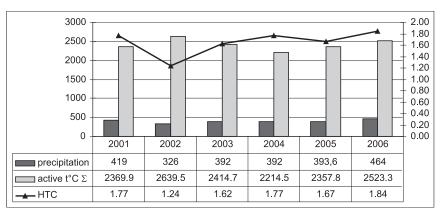
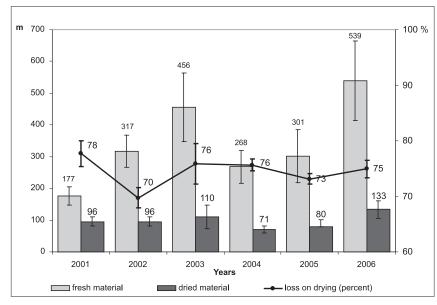
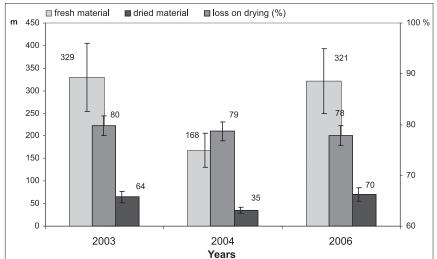
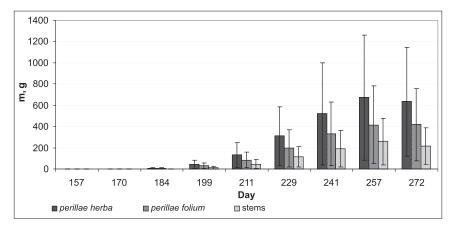
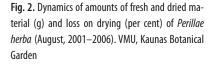


Fig. 1. Dynamics of meteorological factors (sum of average active temperatures  $\Sigma$  t °C, amount of precipitation mm and hydrothermal coefficient HTC) during the vegetation period (May–October, 2001–2006). VMU, Kaunas Botanical Garden

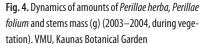








**Fig. 3.** Dynamics of amounts of fresh and dried material (g) and loss on drying (per cent) of *Perillae folium* (August, 2003–2004). VMU, Kaunas Botanical Garden



till flowering (211<sup>th</sup>-257<sup>th</sup> days). Analogical consistent patterns and tendencies were determined in assessing the dynamics of growth parameters (height and projection cover) during vegetation (Ragažinskienė et al., 2006).

The dynamics of absolute increment and increment rate of *P. frutescens* herba, folium and stems was evaluated in 2006, and it was assessed that the increment augmented from the initiation of vegetation and reached maximum during flower-

ing (240<sup>th</sup> day). This increment decreased till minimum after the flowering and full seed ripening at the end of vegetation because vitality of the plant decreased, some leaves and stems died, dried and fell down. The absolute increment and rate of increment of *P. frutescens* leaves is more intense and greater than the increment of stems (Fig. 5–6).

When the dynamics of the average leave quantity of *P. frutescens* model plant was assessed, the tendency of the incre-

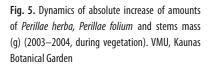
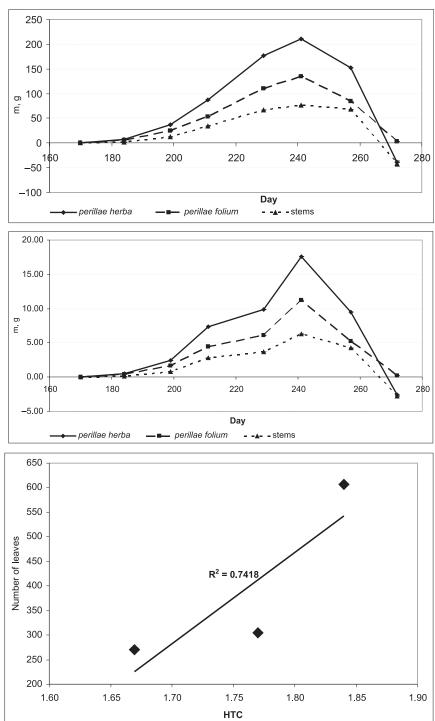


Fig. 6. Dynamics of absolute increase rate of amounts of *Perillae herba, Perillae folium* and stems mass (g) (2003–2004, during vegetation). VMU, Kaunas Botanical Garden

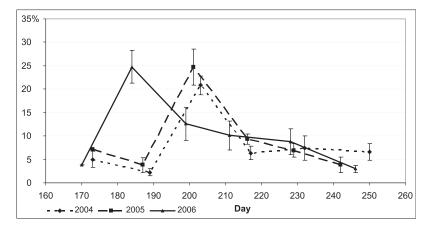


**Fig. 7.** Association between the number of leaves of *Perilla frutescens* L. Britton. and hydrothermal coefficient (HTC)

ment of the number of leaves was observed from the initiation of vegetation (158<sup>th</sup> day of the year) till flowering (250<sup>th</sup> day of the year). The amount of leaves reached maximum during the flowering period in all the years of investigation. The maximum quantity of *P. frutescens* leaves (p < 0.05) was assessed in 2006, when HTC = 1.84. The amount was twice lower in 2004 and 2005, due to the unfavourable growth conditions (Fig. 1). Interdependence was established between the quantity of *P. frutescens* leaves ( $R^2 = 0.7418$ ) and hydrothermal coefficient (Fig. 7).

Slow increment rate of *P. frutescens* leaves was assessed during the initiation of vegetation (170<sup>th</sup>–190<sup>th</sup> days of the year) after the dynamics of the relative increment rate was analyzed (2004– 2006). The rate increased from the 190<sup>th</sup> day (13–20 per cent per day) till the initiation of flowering (200<sup>th</sup> day of the year). The rate of leaves' quantity increment suddenly slowed down (from 13–15 per cent per day till 5–9 per cent per day) during massive flowering and full seed ripening periods (220<sup>th</sup>–250<sup>th</sup> days of the year) and stabilized at the end of vegetation (Fig. 8).

After the dynamics of increment, the increment rate of leaves, and *P. frutescens herba* quantity was assessed, the tendency in augmentation of these indexes was highlighted from the initiation of vegetation till the massive flowering period. Sudden decrease in increment was observed after flowering and at the end of vegetation.



**Fig. 8.** Dynamics of absolute increase rate of amounts of *Perillae folium* quantity (2004–2006, during vegetation). VMU, Kaunas Botanical Garden

The results of investigations obtained and the consistent patterns defined enable us to predict the productivity and its variation of *P. frutescens* terraneous parts depending on the meteorological factors during the vegetation periods.

#### CONCLUSIONS

Assessments of the quantity and increment dynamics of *Perilla frutescens* L. Britton. raw material (*perillae herba, perillae folium*) in multiyear investigations enable to draw inferences from the results obtained:

1. *Perilla frutescens* L. Britton. is an annual herbaceous medicinal plant of *Lamiaceae* Lindl. family which has been cultivated in the collection and exposition of medicinal plants and in the trial area at the Kaunas Botanical Garden of Vytautas Magnus University since 1998.

2. The average yield of *P. frutescens herba* raw material fresh mass is 335.8 g and that of 95.8 g of dried mass. The maximum yield is in favourable growing conditions when HTC = 1.84. The average yield of the fresh and dried material of *P. frutescens* leaves is 248.0 and 49 g, respectively. The yield of *P. frutescens* leaves constitutes about 70 per cent of the total amount of *P. frutescens* material.

3. The absolute increment and increment rate of *P. frutescens* augmented up till maximum from the initiation of vegetation till flowering. The rate of this increment suddenly decreased at the end of vegetation.

4. Interdependence was assessed ( $R^2 = 0.7418$ ) between the number of leaves of *P. frutescens* and hydrothermal coefficient. Statistically significant maximum of leaves was assessed when HTC = 1.84.

5. The results of this study enable us to prognosticate the dynamics of the productivity and its increment of the terraneous part during vegetation in Lithuania.

## ACKNOWLEDGEMENTS

We would like to thank the Lithuanian State Science and Studies Foundation, the Science Fund of Kaunas University of Medicine as well as the staff of the Kaunas Botanical Garden of Vytautas Magnus University for their help in the trial areas.

> Received 10 March 2007 Accepted 22 June 2007

#### References

- 1. Agrometeorologinis biuletenis. 2001–2006. Vilnius. Vol. 1–12.
- Bernath J. 1999. Biological and economical aspects of utilization and exploitation of wild growing medicinal plants in middle and south Europe. Proceedings of the Second World Congress on Medicinal and Aromatic Plants for Human Welfare (WOCMAP II). Biological resources, sustainable use, conservation and ethnobotany. *Acta Horticulturae*. Vol. 500. P. 31–41.
- Brenner D. M. 1993. *Perilla*: botany, uses, and genetic resources. In: J. Janick and J. E. Simon. *New Crops*. N. Y. John Wiley and Sons. P. 322–328.
- 4. Bukantis A. 1994. Lietuvos klimatas. Vilnius.
- Dirsė A. 2001. Žemės ūkio augalų vegetacijos laikotarpių drėgmingumas. Žemės ūkio mokslai. T. 3. P. 51–56.
- Čekanavičius V., Murauskas G. Statistika ir jos taikymai. 2000.Vilnius. TEV. T. 1. P. 239.
- Čekanavičius V., Murauskas G. Statistika ir jos taikymai. 2002. Vilnius. TEV. T. 1. P. 268.
- ESCOP Monographs. 2003. The Scientific Foundation for Herbal Medicinal Products. United Kingdom, Germany, USA.
- Follett J. M., 1995. *Perilla*: production in Japan and potential for New Zealand. *Combined Proceedings*. *International Plant Propagators' Society*. Vol. 45. P. 78–81.
- Gailys V., Benetis R., Pavilionis A. ir kt. 2004. Исследования антимикробной активности in vitro экстрактов листьев перилы. *Immunologija, Allergologija, Infektologija*. T. 3. C. 59–63.
- Gailys V. 2005. The investigation and evaluation of the immunomodulary properties of rattlesnake weed (*Perilla frutescens* L. Britton.). Summary of the Doctoral Dissertation. Kaunas.
- 12. Hartley M. J. 1996. Weed control in *Perilla*. *Proceedings of* 49th N. Z. Plant Protection Conference. P. 165–168.
- He-Ci-Yu, Kosuna K, Haga M. 1997. *The Genus* Perilla. Overseas Publishers Association Netherlands. P. 1–191.
- Kim S. 1997. The *Perilla* oil prevents the excessive growth of visceral adipose tissue in rats by down regulating adipocyte differentiation. *Journal of Nutrition*. Vol. 127. No. 9. P. 1752–757.

- 15. Lage D. 1998. Europe's medicinal and aromatic plants: their use, trade and conservation. A Traffic Network Report *Traffic International*. Cambridge, UK.
- Longvah T., Deosthale Y. G. 1991. Chemical and nutritional studies on Hanshi (*Perilla frutescens*), a traditional oilseed from Northeast India. *Journal of the American Oil Chemists' Society*. Vol. 68. No. 10. P. 781–784.
- 17. Miuleris J. 1994. *Bluzmanas. Augalų stebėjimų ir bandymų metodai*. Vilnius.
- Nagatsu A., Tenmaru K., Matsuura H. et al. 1995. Novel antioxidants from roasted *Perilla* seed. *Chemical & Pharmaceutical Bulletin*. Vol. 43. P. 887–889.
- Osakabe N., Yasuda A., Natsume M., Yoshikawa T. 2004. Rosmarinic acid inhibits epidermal inflammatory responses: anticarcinogenic effect of *Perilla frutescens* extract in the murine two-stage skin model. *Carcinogenesis*. Vol. 25. No. 4. P. 549–557.
- Nitta M., Lee J., Ohnishi O. 2003. Asian *Perilla* crops and their weedy forms: their cultivation, utilization and genetic relationships. *Economic Botany*. Vol. 57. No. 2. P. 245–253.
- 21. Omer E. A., Khattab M. E., Ibrahim M. E. 1998. First cultivation trial of *Perilla frutescens* L. in Egypt. *Flavor and Fragrance Journal*. Vol. 13. P. 221–225.
- Povilaitytė V., Venskutonis P. R. 2000. Investigation of antioxidative activity of purple peril (*Perilla frutescens* L. Britton.), (*Dracocephalum moldavica* L.) and Roman Chammomile (*Anthemis nobilis* L.) extracts in rapeseed oil. *Journal of American Oil Chemist's Society*. Vol. 77. No. 9. P. 951–956.
- Povilaitytė V. 2004. Krūminės perilės (Perilla frutescens L. Britton.), darželinės žiomenės (Dracocephalum moldavica L.) ir tauriojo bobramunio (Anthemis nobilis L.) biologiškai aktyvūs junginiai ir jų savybės. Daktaro disertacijos santrauka. Kaunas.
- Radušienė J. 2004. Trade, use and conservation of medicinal and aromatic plants in Lithuania. *Acta Horticulturae*. Vol. 629. P. 31–37.
- Radušienė J., Janulis V. 2004. Vaistinių ir aromatinių augalų įvairovės tyrimo, jų panaudojimo bei išsaugojimo tendencijos. *Medicina*. T. 40. Nr. 8. P. 705–709.
- Ragažinskienė O., Gailys V. ir kt. 2004. Krūminė perilė (*Perilla frutescens* L. Britton) – perspektyvus imunomoduliatorius. *Medicina*. T. 40. Nr. 3. P. 220–224.
- Ragažinskienė O., Maruška A., Kornyševa O. 2006. Genetic resources and phytochemical analysis of medicinal and spice (aromatic) plants in Kaunas Botanical Garden of Vytautas Magnus University. *Vytauto Didžiojo* universiteto Botanikos sodo raštai. Kaunas. T. XI. P. 86–91.

- Ragažinskienė O., Rimkienė S. 2003. Journal of Medicinal & Spice Plants. Vol. 8. No. 4. P. 89–91.
- Ragažinskienė O., Šeinauskienė E., Janulis V., Jankauskaitė L. Milašius A. 2006. The influence of meteorological factors on growth and vegetation process of *Perilla frutescens* in Lithuania. *Medicina*. Vol. 42. No. 8. T. 667–670.
- Tada M., Matsumoto R., Yamaguchi H., Chiba K. 1996. Novel antioxidants isolated from *Perilla frutescens* Britton var. Crispa (Thunb.). *Bioscience, Biotechnology and Biochemistry*. Vol. 60. T. 1093–1095.
- The Pharmacopoeia Commision of PRC 1992. *Pharmacopoeia of The People's Republic of China*. China. Vol. 18. P. 75–78.
- WHO traditional medicine strategy 2002–2005. Document WHO/EDM/TRM/2002.1.

Ona Ragažinskienė, Lina Bumblauskienė, Erika Šeinauskienė, Antanas Praškevičius, Valdimaras Janulis

## PERILLA FRUTESCENS (L.) BRITTON ANTŽEMINĖS DALIES DAUGIAMETĖ PRIEAUGIO DINAMIKA VEGETACIJOS TARPSNIAIS VIDURIO LIETUVOJE

#### Santrauka

Straipsnyje pateikiami 2001–2006 m. Vytauto Didžiojo universiteto Kauno botanikos sode *Perilla frutescens* L. Britton antžeminės dalies vaistinės žaliavos (*Perillae herba, Perillae folium*) kiekio ir jos prieaugio daugiametės dinamikos vegetacijos tarpsniais tyrimų duomenys.

*P. frutescens* (*Lamiaceae* Lindl.) vienametis, žolinis, vaistinis, prieskoninis, dekoratyvinis augalas, kilęs iš Rytų Azijos, o pastaraisiais metais auginamas bei introdukuojamas įvairiose pasaulio šalyse. Ši nauja rūšis nuo 1998 m. iki šiol introdukuojama Vidurio Lietuvoje, Vytauto Didžiojo universiteto Kauno botanikos sodo vaistinių augalų kolekcijoje ir bandymo plote. Nustatyta *P. frutescens* žolės vidutinis modelinio augalo šviežios masės kiekis 335,8 g, orasausės – 95,8 g, *P. frutescens* vidutinis modelinio augalo lapų šviežios masės kiekis 248 g, orasausės – 49 g; lapų masė sudaro 70% bendros antžeminės dalies vaistinės žaliavos. Nustatytas *P. frutescens* lapų kiekio priklausomumas ( $R^2 = 0,7418$ ) nuo kritulių kiekio ir oro temperatūrų sumos. *P. frutescens* vaistinės žaliavos masės absoliutus prieaugis ir šio prieaugio tempas greitėja iki maksimumo nuo vegetacijos pradžios iki žydėjimo tarpsnio, šio prieaugio tempas staiga sulėtėja žydėjimo–vegetacijos pabaigoje.

Tyrimų duomenų dėsningumai pagrindžia *P. frutescens* antžeminės dalies vaistinės žaliavos produktyvumą, prognozes, dinamiką bei prieaugį vegetacijos tarpsniais Lietuvoje.

Raktažodžiai: *Perilla frutescens* L. Britton, introdukcija, daugiametė prieaugio dinamika, meteorologiniai veiksniai